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WHEN TO TEST

Criteria for Susceptibility Testing

I. Introduction

This section lists the susceptibility testing methods and required antimicrobials for each significant organism appropriate to the site of isolation. Perform susceptibility testing on pure cultures ONLY.

II. Reagents/Materials/Media

Analytical Process – Bacteriology Reagents_Materials_Media List QPCMI10001

III. Method

1. Select significant organisms as per procedure manual of body sites.

2. Identify the selected isolate as perBacteria and Yeast Workup Manual.

3. For identical organisms, as defined in Bacteria and Yeast Workup Manual – Minimal workup, isolated within 1 day (24 hours) from blood and sterile sites for bacteria OR 7 days from blood and sterile sites for yeasts OR bacteria within 3 days from other sites do not require repeat susceptibility testing, use statement "Susceptibility testing not done. Please refer to ___ collected on date ___ and time _____"
    i. **EXCEPTION**: oxacillin and vancomycin screen for *Staphylococcus*, vancomycin screen for *Enterococcus* and meropenem screen for Enterobacterales

4. Refer susceptibility results back to like sites only and NEVER refer a sterile site to a non-sterile site. NEVER refer clinical isolates to isolates from infection control screens or vice versa.

5. For Infection Control Screens isolates of identical organisms (identified by minimal tests-see IC manual), full susceptibility only needs to be performed if there were no identical isolates in the past 3 months.

6. Follow the table below as a guide for the appropriate method(s)/antimicrobial(s) to be setup

7. If the Vitek susceptibility panel or drug(s) are terminated, please set up alternate method

8. Please read your KB first then accept your VITEK results – this will allow for reporting rules in SOFT to work.
9. Minocycline (MH),Chloramphenicol (C), and Cefiderocol (FDC) must be set up on plain MH agar (NOT MH agar plus)

WHAT TO TEST:

Organisms	Site	Method	Antimicrobial(s)
Enterobacterales Non-SPICE [4]	Non-sterile sites - E. coli or P. mirabilis NICU patient	Vitek [7] Add KB	astn391 [2, 3, 5] AMP
	Sterile sites [1]	Vitek [7] Add KB	astn391 [2, 3] AMP, KZ[9], ETP, TOB, AK
	If initial AST results suggest the isolate is multi-drug resistant [6]		
Enterobacterales SPICE [8]	All sites	Vitek [7] Add KB	astn391 [3] CIP, GM, TOB, AK, ETP
	If initial AST results suggest the isolate is multi-drug resistant [6]		

Additional Testing Notes for Enterobacterales (SPICE/non-SPICE)		
[1] Early growth of *E. coli, K. pneumonia, K. oxytoca* or *P. mirabilis* from Blood culture or Sterile Fluid	*B*LACTA [2]	See How to detect ESBL
[2] *E. coli, K. pneumonia, K. oxytoca* or *P. mirabilis*: If CAZ or CRO=I or R or *B*LACTA+ **ONLY** from MSH newborn:D1-M13/Female 12-50yrs	+ KB-ESBL	AMC, ATM, CRO, CAZ, CPD, FOX, TZP, FEP,ETP,MEM
[3] If erta = I/R and mero MIC ≤0.25mg/L	mero screen	mems - See How to detect CRE
If Meropenem mic ≥0.5 mg/L or	*β*CARBA →	See How to detect CRE
If Meropenem screen ≤25mm	*β*CARBA →	
[4] Proteus non-vulgaris sp	KB →	AMC (on request only)
[5] If I or R to CIP, SXT, 3rd gen cephs and Gent	KB →	AMP, KZ[9], ETP, TOB, AK
[6] *If multi-drug resistant:* If I or R to all of the following from urine site: AMP, AMC, cephalexin, CIP, nitro and SXT	KB (**kbxeru**)	FOS, DO
If resistant to **all** routinely tested antimicrobials (**disregarding** aminoglycoside results) — Urines	KB →	FOS, DO , ATM, TE, FEP,CZA, FDC(using plain MH agar)
	etest→	C/T
	PHL→	CO
If resistant to **all** routinely tested antimicrobials (**disregarding** aminoglycoside results) — All other sites	KB (kbxdr)	FOS, DO , ATM, , TE, FEP,CZA, MH,FDC, C (using plain MH agar)
	etest (etresis)	TGC, C/T
	PHL→	CO
If resistant to **all** the above **All** sites(disregarding colistin)	KB→	I/R
	etest→	M/V
	If oxa-48, NDM, VIM or IMP detected +DoubleDisk KB→	CZA, ATM
[7] Enterobacterales not growing on Vitek from all	KB →	AMP, KZ[9], CRO, CIP, SXT, CN, TZP,

Organisms	Site	Method	Antimicrobial(s)
sites		TOB, CAZ, ETP, MEM, CPD, AK, F, AMC	
[8] SPICE group includes Serratia, Providencia, Morganella, Proteus vulgaris, Proteus penneri, Citrobacter, Enterobacter including Klebsiella aerogenes, Hafnia, Pantoea			
[9] Do not setup or report on Proteus mirabilis			
E. coli O:157	Enteric sites	Not tested	
Salmonella typhi	Enterics sites – routine	KB→ + etest→	AMP, SXT, AZ CI,
	If resistant to **all** routinely tested antimicrobials	KB →	CRO, ETP, MEM, DO
	Non-Enteric sites – routine	KB →	AMP, CRO, SXT, AZ(except CSF/Urine)
		+ etest	CI
	If resistant to **ceftriaxone**	KB→	ETP(except CSF), MEM, DO (except CSF)
Salmonella species other than *S. typhi*	Enterics sites – routine	Not tested	
	On request **ONLY** upon microbiologist approval	KB→ + etest →	AMP, SXT, CI,
	If resistant to **all** routinely tested antimicrobials	KB→	CRO, ETP,MEM, DO
	Non-enteric sites	KB → + etest →	AMP, CRO, SXT CI
	If resistant to **ceftriaxone**	KB →	ETP(except CSF), MEM, DO(except CSF)

Organisms	Site	Method	Antimicrobial(s)
Shigella species	Enterics sites – routine	KB → + etest→	AMP, SXT, AZ CI
	If resistant to **all** routinely tested antimicrobials	KB→	CRO, ETP, MEM, DO,
	Non-Enteric sites – routine	KB → + etest →	AMP, CRO, SXT, AZ(except CSF/Urine) CI
	If resistant to **ceftriaxone**	KB→	ETP(except CSF), MEM, DO(except CSF),
Vibrio species	Enteric sites Sterile sites	Not tested → PHL	
Acinetobacter species	All sites	Vitek → **Add** KB →	astn391 AK, TOB
		If Vitek mero=I/R, KB	MEM
		If KB MEM I/R	Send to NML for PCR
	If resistant to **all** routinely tested antimicrobials (**excluding** aminoglycosides)	etest (etresa)	TGC **(Except Urine)**
	If resistant to **all** routinely tested antimicrobials (**including** aminoglycosides)	Non-urine etest (etresis) KB (kbedxa) → PHL	 TGC ATM, DO, MH, FEP, CO
		Urine samples KB (kbedxa) → → PHL	 ATM, DO, TE, FEP, CO
Pseudomonas aeruginosa	All sites	KB	TZP, CAZ, CIP, AN, TOB, MEM
	If resistant to **all** routinely tested antimicrobials (**disregarding** aminoglycosides and colistin)	KB (kbxdrpa) etest (etresa) → PHL	ATM, FEP, CZA C/T CO
	If resistant to **all** the above (disregarding colistin)	KB → etest → +DoubleDisk KB→	I/R, FOS M/V CZA,ATM

Organisms	Site	Method	Antimicrobial(s)
Pseudomonas sp., *Plesiomonas* sp. and other afermenters *	All sites	→ PHL	
	* Other afermenters include -but not limited to- *Moraxella* (see below for specific section for catarrhalis), *Oligella, Chryseobacterium, Elizabethkingia, Myroides, Sphingobacterium, Weeksella*		
Aeromonas species	Non-Enteric Sites	KB →	CRO, CIP, SXT, AK, ETP, MEM, TZP, CN, TE
	Enteric Sites - routine	Not tested	
	Enteric Sites - On request ONLY upon microbiologist approval	KB →	CRO, CIP, SXT, AK, ETP, MEM, TZP, CN, TE
	If resistant to all routinely tested antimicrobials	KB →	C **(Except Urine)**
Stenotrophomonas maltophilia	All sites ***except:***	KB →	LVX, SXT
	Urine/CSF/VP Shunt and Brain	KB →	MH(using plain MH agar)
		+ e-test →	TS
	Urine Site	KB →	LVX, SXT
		+ e-test →	TS
	CSF/VP Shunt and Brain	KB →	SXT
		+ e-test →	TS
	If e-test and KB for sxt disagree	PHL for MIC →	SXT
	If only susceptible to one of the routinely tested antimicrobials	KB →	FDC(using plain MH agar)
		+ DoubleDisk KB	CZA, ATM
		+ etest →	TGC **(Except Urine)**
		PHL for MIC →	CO

Organisms	Site	Method	Antimicrobial(s)
Burkholderia cepacia Complex including [1]: B. cepacia/gladioli/ vietnamiensis/multivorans /pyrrocinia/ubonensis /stabilis/ambifaria	Urine site	PHL for MIC→	CAZ, SXT, MEM, LEV, CO
	CSF/VP Shunt & brain	PHL for MIC→	CAZ, SXT, MEM,TGC, CO,
	Other sites	PHL for MIC→	CAZ, SXT, MEM,TGC LEV, CO, MH
Haemophilus species	All sites	beta-lactamase	
	Blood and Sterile sites	beta-lactamase +KB →	CRO, CIP, AMP
Helicobacter pylori	Gastric biology - On request ONLY	Send to Mayo Clinic	
Moraxella catarrhalis	All sites Blood and Sterile Sites	Not tested → PHL	
Neisseria gonorrhoeae	All sites	→ PHL	
Neisseria meningitides	All sites	→ PHL	
Other fastidious Gram negatives (e.g. HACEK **, *Capnocytophaga, Pasteurella*)	All sites Blood and Sterile Sites	Not tested → PHL	
	** HACEK group includes *Aggregatibacter, Cardiobacterium, Eikenella, Kingella*, for *Haemophilus* see specific section above.		
Campylobacter species	All sites	Not tested	

Organisms	Site	Method	Antimicrobial(s)
Gram Positive:			
Staphylococcus aureus **NOTE: Vancomycin can only be tested by etest as there are no KB interpretations.**	All sites	Vitek → Oxacillin Screen Vanco Screen	astp580 ox va
	Early growth from bloods sterile sites	+ PBP2a →	
	If Vitek SXT = I/R	**add** KB →	SXT
	If MRSA	**add** KB →	MUP 200
	For MRSA all sites, on request ONLY	**add** e-test →	BPR
	If Vancomycin is ≥2 mg/L from Vitek OR growth onVanco Screen plate	Add macro-e-test **add** e-test →	VA, TP VA, TP
	All sites ***except*** Resp samples If MRSA/BORSA **AND**: • Vancomycin MIC >=2 mg/L	**add** KB → **add** e-test →	LZD DPC
	• I/R to All other antimicrobial agents	**add e-test** →	TGC(Except Urine) BPR
	If e-test DPC is non-susceptible,	PHL for MIC	DPC
	If not growing on Vitek from all sites	**add** KB panel→ **add** Breakpoint panel →	kbgpc etstaav
Coagulase-negative *Staphylococcus* NOT *Staphylococcus lugdunensis* **NOTE: Vancomycin can only be tested by etest as there are no KB interpretations.**	Blood Cultures	Not tested If BC endocarditis	astp580
	Urine	Not tested	
	All other sites	Vitek →	astp580
	If not growing on Vitek from all sites	**add** KB panel→ **add** Breakpoint panel →	kbgpc etstanv

Organisms	Site	Method	Antimicrobial(s)
Staphylococcus lugdunensis/ pseudointermedius/ intermedius	All sites	Vitek →	astp580
	If not growing on Vitek from all sites	**add** KB panel→ **add** Breakpoint panel →	kbgpc etstanv
Micrococcus species	All sites	Not tested	
Aerococcus species	Blood & Sterile sites	→ PHL	
	All other sites	Not tested	
Enterococcus species	Urines	Vitek + Screen plate	astgp67 va
		If Amp I/R/Pen allergic AND Nitro = I/R **add** KB →	 FOS
		if Amp, Nitro, Tet AND Levo = I/R, **add** KB	 FOS, LZD, DO
	Blood	KB → + Screen plate	AMP High level gm500 and st2000, va
	Non-Blood Sterile sites	KB → + Screen plate **Add** KB	AMP va DO, LVX
	Blood Cultures: *E.faecium* vanco=S	Cepheid VRE PCR	
	Persistent Enterococcus faecalis positive from the same sterile site (eg. Blood or CSF)- **on request ONLY**	Beta-Lactamase Testing	Cefinase disk
	All other sites	KB → + Screen plate	AMP va
	All sites, if VA=R or vanA/vanB determinant positive	add KB → add e-test →	LZD DPC (excluding respiratory)

Organisms	Site	Method	Antimicrobial(s)
Streptococcus pneumoniae	Blood & Sterile sites	Sensititre → If Clindamycin is requested,DoubleDisk KB	TX, PG, LX, EM, VA, DA, E
	All other sites	DoubleDiskKB + KB → If OX=R then Sensititre →	DA, E OX, LVX, VA TX, PG, LX, EM, VA
Group A, B, C, G *Streptococcus*	Blood	DoubleDisk KB + KB →	DA, E P, VA
	Sterile sites	DoubleDisk KB + KB → If Bone/Joint **Add** KB	DA,E P,VA TE**, LVX**
	Urine for Group A, C, G on request ONLY	KB →	LVX, VA
	Urine, GBS on request ONLY: - female >12 and <60 years old (with significant amount) - female >12 and <60 years old (with insignificant amount) - male or female <12 or >60 years old	KB → DoubleDisk KB KB → DoubleDisk KB KB →	LVX, VA DA, E VA DA, E LVX
	- Vaginal GBS screens, on request ONLY or patient is Penicillin allergic.	DoubleDisk KB + KB	DA, E VA
	Other sites, on request ONLY	DoubleDisk KB + KB	DA, E LVX, VA

Organisms	Site	Method	Antimicrobial(s)
Streptococcus bovis, viridans *Streptococcus*	Blood One morphotype → >1 morphotype →	DoubleDisk KB e-test → Not tested	DA, E TX, PG, VA
	Sterile Sites	DoubleDisk KB e-test → **Add** KB	DA, E TX, PG, VA TET, **LVX**
	Urine, on request ONLY	KB → + e-test →	VA, LVX PG
	Other sites, on request ONLY	KB → + e-test →	VA, LVX PG
Streptococcus anginosus group and small colony-ß-haemolytic *Streptococcus*	Blood	DoubleDisk KB + KB → + e-test	DA, E LVX, VA PG, TX
	Sterile sites	DoubleDisk KB + KB → + e-test **Add KB**	DA, E LVX, VA PG, TX TET
	Urine, on request ONLY	KB → + e-test →	LVX PG
	Other sites, on request ONLY	DoubleDisk KB + KB → + e-test	DA, E LVX, VA PG, TX
Listeria species	All sites	Not tested	
Corynebacterium species	All sites	Not tested	
Bacillus species	All sites	Not tested	
Nocardia species	All sites	Not tested	Send to PHL on special request
Anaerobes	All sites	Not tested	Send to PHL on special request
Yeasts	Blood and Sterile sites	Not tested	Send to PHL
	Other non-sterile sites	Not tested	Send to PHL on special request
Filamentous Fungus [1]	All sites	Not tested	Send to PHL on special request

Note:
[1] For all requests, ask the requesting physician to contact Dr. Julianne Kus (julianne.kus@oahpp.ca) or PHOL covering microbiologist via email for approval. Once approval has been obtained, notify microbiology.specialqueries@sinaihealth.ca to send the isolate to PHOL. On the requisition, please indicate that approval by Julianne Kus (or the covering microbiologist) has been obtained.

Reference:

Coenye T, Vandamme P, Govan JR, Lipuma JJ. Taxonomy and identification of the Burkholderia cepacia complex. J Clin Microbiol. 2001;39(10):3427–36.

WHAT TO REPORT:

Urine – Gram Positive Susceptibility Reporting 1 – *Staphylococcus* species, MRSA/BORSA

Antimicrobial Agent	*Staphylococcus species*	MRSA/BORSA
Ampicillin	X[1]	X[1, 5]
Cefazolin	X[2]	X[2, 5]
Ceftobiprole		X[13, 16]
Cloxacillin	X[2]	X[2, 5]
Daptomycin		X[14,15]
Doxycycline	X[3,11]	X[3, 6, 11]
Linezolid		X[15]
Mupirocin		X[6, 8]
Nitrofurantoin	X	X[5]
Rifampin		X[6]
Trimethoprim/Sulfa	X[12]	X[12, 16]
Vancomycin	X[4, 9]	X[5, 9]

[1] Base on Penicillin or beta-lactamase result
[2] Base on Oxacillin/cefoxitin result; for *Staphylococcus pseudointermedius* base on Oxacillin result
[3] Report only for age>7 y; base on Tetracycline result
[4] Report if patient is allergic to Penicillin OR if *Staphylococcus* species is resistant to **All** other antimicrobial agents.
[5] DO NOT report if isolated from Infection Control Screening test
[6] For Infection Control Screen, include Isolate Comment "Susceptibility results are provided for infection control purposes only."
[8] For KB result that shows any zone, report as Isolate Comment "No high-level mupirocin resistance detected"
For KB result that shows no zone, report as Isolate Comment "High-level mupirocin resistance detected"
[9] For *S. aureus* or MRSA, vancomycin MIC=2.0 mg/L, result with ISOLATE comment: "This isolate has a vancomycin MIC of 2 mg/L which is associated with an increased risk of vancomycin treatment failures. Consultation with infectious diseases or medical microbiology is advised."
[10] Report if I/R to **All** other antimicrobial agents **OR** if requested.
[11] If I/R, add comment "Doxycycline results are based on testing tetracycline which may overcall doxycycline resistance. If you wish this isolate to be tested with doxycycline directly, please contact the microbiology laboratory."
[12] Base on KB results if Vitek = I/R, do not report on patients <2 months old
[13] Report with comment:
There are no CLSI standards for this drug. EUCAST suggests MICs <=2 mg/L correlate with susceptibility. Please consult the microbiologist-on-call with any questions. For research use only.
There are no CLSI standards for this drug. EUCAST suggests MICs >2 mg/L correlate with susceptibility. Please consult the microbiologist-on-call with any questions. For research use only.

[14] Report with comment:

Daptomycin MIC = xx mg/L
CLSI recommends MIC <= 1 mg/L correlates with susceptibility.
CAUTION: Daptomycin MIC results within 2-fold dilutions of this breakpoint may not be reliable given variance in media calcium concentration which may result in false susceptibility or false resistance.

Daptomycin MIC = xx mg/L
CLSI recommends MIC > 1 mg/L correlates with non-susceptibility.
CAUTION: Daptomycin MIC results within 2-fold dilutions of this breakpoint may not be reliable given variance in media calcium concentration which may result in false susceptibility or false resistance.

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[15] Report if MRSA/BORSA **AND** vanco MIC of 2mg/L
[16] Report if MRSA/BORSA **AND** vanco MIC of 2mg/L **AND** daptomcycin MIC >1mg/L.

Note: ***S. saprophyticus*** and coagulase-negative-***Staphylococcus*** – **DO NOT** report susceptibilities. Report with Isolate Comment – "Susceptibility testing of this organism is not routinely done because infections respond to concentrations achieved in urine of antimicrobial agents commonly used to treat acute, uncomplicated urinary tract infections e.g. nitrofurantoin, trimethoprim/sulfa or fluoroquinolones. Suggest repeat specimen with request for susceptibility testing if patient does not respond to empiric therapy."

Note: If all antimicrobial agents are resistant, inform the Microbiologist on-call.

Urine – Gram Positive Susceptibility Reporting – 2 – *Enterococcus* species, *Streptococcus* species, *Aerococcus* species

Antimicrobial Agent	*Enterococcus* species[5]	Group A, B, C, G *Streptococcus*	*Streptococcus anginosus* group	*Aerococcus* species
		Routinely not tested See Below[7] For special requests:	Routinely not tested See Below[13] For special request:	Routinely not tested See Below[18]
Ampicillin	X[20]			
Clindamycin		X[10, 11]		
Daptomycin	X[15]			
Doxycycline	X[1, 16]			
Erythromycin				
Fosfomycin	X[19]			
Levofloxacin	X[4]	X[6, 9, 17]	X[6]	
Linezolid	X[2,16]			
Nitrofurantoin	X[20]			
Penicillin G			X	
Tetracycline	X[1,20]			
Vancomycin	X[3, 8]	X[12]		

[1] Report only for age >7 y
[2] Report if Vancomycin is **R**, except for *E. gallinarum* and *E. casseliflavus*.
[3] Test but **DO NOT** report unless Vancomycin R or Enterococcus resistant to **All** other antimicrobial agents
[4] Report if Ampicillin, Nitrofurantoin and Tetracycline are ALL I/R.
[5] If isolated from Infection Control Screening test, include Isolate Comment "Susceptibility results are provided for infection control purposes only."
[6] Adults only (>18y)
[7] Report "This organism is intrinsically susceptible to penicillin. If treatment is required AND this patient cannot be treated with penicillin, please contact the Microbiology Department within 48 hours to request sensitivity testing.
[8] *E. gallinarum and E. casseliflavus*, report as **R** with the statement "This organism always has intrinsic non-transmissible resistance to vancomycin. The patient does not require isolation."
[9] For male or female <12 or >60 years old, report with additional isolate comment "Susceptibility completed as requested" (do not remove original comments). If Levofloxacin is R or patient is <18y, consult the Microbiologist.
[10] For female >12 and <60 years old (**with significant amount**) reported with the isolate comment "Susceptibility testing completed as requested. Note: clindamycin should NOT be used to treat bacteriuria, they are provided to help guide intrapartum chemoprophylaxis (if this patient is pregnant)." (do not remove original comments)
[11] For female >12 and <60 years old (**with insignificant amount),** report with additional isolate comment "Susceptibility testing completed as requested for intrapartum chemoprophylaxis" (do not remove original comments).
[12] Report if R to clindamycin
[13] *Streptococcus anginosus* group are generally susceptible to penicillin and levofloxacin. If susceptibility testing for this organism is required, please contact the microbiology laboratory within 48 hours.
[14] Report if I/R to **All** other antimicrobial agents **OR** if requested.
[15] Report if requested base on etest results
[16] Report if Ampicillin, Nitrofurantoin, Tetracycline and Levofloxacin are ALL I/R
[17] For female >12 and <60 years old (with insignificant amount) do NOT report.

[18] "Aerococcus species are usually susceptible to beta-lactams and vancomycin. If you would like susceptibility testing to be completed, please contact the Microbiology Laboratory."

[19] Report for I/R to ampicillin and nitrofurantoin. For *E. faecalis* report interpretation. For *E. faecium* report with with zone diameter in isolate canned comment\FseS.

[20] if "S" for *E.faecalis* add Isolate Message "*E. faecalis* is generally susceptible to fosfomycin for treatment of acute uncomplicated cystitis."

Note: If all antimicrobial agents are resistant, inform the Microbiologist on-call.

Urine – Gram Negative Susceptibility Reporting [27]

Antimicrobial Agent	**Enterobacterales** [16] **excluding *Salmonella* spp. And *Shigella* spp.**	***Salmonella* spp.**	***Shigella* spp.**	***Acinetobacter* spp.**[16]	***P. aeruginosa***[16]	***Aeromonas spp.***[25]	***S. maltophilia****	***B. cepacia***
Amikacin	X[13]			X[13]	X[13]	X[13]		
Ampicillin	X [7, 10, 11]	X	X					
Amoxicillin/Clavulanate	X							
Aztreonam	X[32]				X[17]			
Cefepime	X[32]				X[17]			
Cefiderocol	X[32]						X[24]	
Cephalexin[15]	X[8, 10]							
Ceftazidime					X		X[5]	X
Ceftazidime-avibactam	X [32]				X[17]			
Ceftazidime-avibactam+Aztreonam combination	X[30,31]				X[24, 30]		X[24, 30]	
Ceftriaxone	X[6, 10, 11, 12]	X[28]	X[28]	X		X[12]		
Ceftolozane/Tazobactam	X[14, 32]				X[17]			
Ciprofloxacin	X[1]	X[1]	X[1]	X[1]	X [1]	X[1]		
Colistin	X[32]			X[17]	X[17]		X[17]	X[17]
Doxycycline	X[18]	X[29]	X[29]					
Ertapenem	X[3]	X[29]	X[29]					
Fosfomycin	X[23]				X[17]			
Gentamicin	X			X		X		
Imipenem-Relebactam	X[24]				X[24]			
Levofloxacin							X[1]	X[5]
Meropenem	X[3]	X[29]	X[29]	X	X[4]	X[2]		X
Meropenem-Vaborbactam	X[24]				X[24]			
Nitrofurantoin	X							
Piperacillin/Tazobactam	X[9, 10, 12]			X	X	X[12]		
Tetracycline	X,[21, 32]					X[21, 22]		
Trimethoprim/Sulfa [26]	X	X	X	X		X	X	X

Tobramycin	X			X	X			

[1] Adults only (>18 y)
[2] Report if I/R
[3] Report if I/R **OR** if ≤1 of the antimicrobial agents is susceptible: aminoglycosides , ciprofloxacin, TMP/SMX, 3rd generation cephalosporins (ceftriaxone or ceftazidime) **OR** if requested
[4] Report if I/R **OR** if I/R to 3 of the 4 antimicrobial agents: aminoglycosides , ciprofloxacin, 3rd Generation Cephalosporins, Piperacillin/tazobactam **OR** if requested
[5] Report MIC from PHL if I/R **OR** if I/R to **All** other antimicrobial agents
[6] Report ceftriaxone only if I/R to Cephalexin
[7] *Klebsiella* spp., and SPICE always report Amp as R.
[8] Report for *E. coli, Klebsiella pneumonia* & *Proteus mirabilis* only.
[9] Do not report for Salmonella species.
[10] For *E. coli, Klebsiella* species and *Proteus* species that are confirmed to have an ESBL of any class, report all penicillins and first, second and third generation cephalosporins and piperacillin/tazobactam as R.
[11] For *Enterobacterales* other than *E. coli, Klebsiella* species and *Proteus* species where ESBL testing is not done, if any one of cefotaxime/ceftriaxone or ceftazidime=R, report all penicillins and first, second and third generation cephalosporins and piperacillin/tazobactam as R
[12] For SPICE, report all penicillins and first, second and third generation cephalosporins and piperacillin/tazobactam as R; report with comment "Resistance to extended-spectrum penicillins, beta-lactam/beta-lactamase inhibitor combinations (e.g. piperacillin-tazobactam), and cephalosporins may develop during therapy. These agents should be avoided and will be reported as resistant regardless of their in vitro susceptibility results. If you have questions, please contact the medical microbiologist on call.."
[13] Report if both Gentamicin and Tobramycin are I/R.
[14] Report with comment if I/R to **All** other Antimicrobial Agents **OR** if only aminoglycoside is S **OR** if requested
[15] Reflex from Cefazolin tested in Vitek2
[16] If isolated from Infection Control Screening test, include Isolate Comment "Susceptibility results are provided for infection control purposes only."
[17] Report if I/R to **All** other Antimicrobial Agents (disregarding aminoglycosides) **OR** if requested.
[18] Report if I/R to **All** other Antimicrobial Agents **including** aminoglycoside **OR** if requested.
[19] Report with comment if I/R to **All** other Antimicrobial Agents **including** aminoglycoside **OR** if requested.
[20] Report if I/R to **all** routinely tested antimicrobials (disregarding aminoglycosides)
[21] Report only for age >7 y
[22] Report if I/R to **All** ciprofloxacin, amoxicillin/clavalacnic acid and trimethoprim/sulfamethoxazole
[23] Report if I/R to all of the following: amoxicillin/ampicillin, amox/clav, cephalexin, ciprofloxacin, nitrofurantoin and TMP/SMX. Report *E. coli* with interpretation. Report other *Enterobacterales* with zone diameter and Isolate Message. For *E. coli* where fosfomycin is not reported, add Isolate Message "*E. coli* is generally susceptible to fosfomycin for treatment of acute uncomplicated cystitis."
[24] Report if I/R to **All** other Antimicrobial Agents
[25] Report with: "Resistance to beta-lactam antimicrobials may develop in *Aeromonas* species during therapy. Choosing a non-beta-lactam antimicrobial and considering combination therapy is recommended for serious infections. Consultation with infectious diseases or medical microbiology is advised."
[26] Do not report on patients <2 months old
[27] Pseudomonas species (other than P. aeruginosa), fastidious gram-negative bacteria & non–fermenters – Send to PHOL and report results as tested.
[28] Report only for age >1m
[29] Report if I/R to ceftriaxone
[30] Report CZA+ATM combination result based on double disc diffusion between CZA and ATM
[31] Report if I/R to All other Antimicrobial Agents **AND** if oxa-48, NDM, VIM or IMP detected
[32] Report if I/R to all **routinely** tested antimicrobials (***disregarding*** *aminoglycosides*)

*For *Stenotrophomonas maltophilia* isolated from **all** sites and susceptibility result is reported, add comment "If treatment is deemed clinically warranted for *S. maltophilia*, combination therapy with two effective antimicrobials is recommended until clinical improvement is observed.

If all antimicrobial agents are resistant, inform the Microbiologist on-call.

Enterics

Antimicrobial Agent	*Shigella* species[1,2]	*Salmonella* species other than *Salmonella typhi* [2]	*Salmonella typhi*	*Vibrio cholerae*[3]	*Aeromonas* species[5,9]
Amikacin					X[6]
Amoxicillin/Clavulanic acid					
Ampicillin	X	X[5]	X	X	
Azithromycin	X		X		
Ceftriaxone	X[13]	X[5,14]	X[13]		X
Chloramphenicol					X[7]
Ciprofloxacin	X[1,10]	X[5]	X[1]	X[1]	X
Doxycycline	X[4, 13]	X[4, 5, 14]	X[4, 13]		
Ertapenem	X[13]	X[5, 14]	X[13]		X[6]
Gentamicin					X[6]
Meropenem	X[13]	X[5, 14]	X[13]		X[6]
Piperacillin/Tazobactam					X[6]
Trimethoprim/Sulfa [11]	X	X[5]	X	X	X
Tetracycline				X[4]	X[4, 8]

[1] Adults only (>18 y).
[2] Not tested or reported from enteric isolates
[4] Report only for age >7 y
[5] On request, ONLY upon Microbiologist approval
[6] Report if intermediate or resistant to all: Amoxicillin/Clavulanic acid, Ceftriaxone, Ciprofloxacin, Trimethoprim/Sulfa, Tetracycline.
[7] Report if I/R to **All** other Antimicrobial Agents
[8] Report if I/R to **All** ciprofloxacin, amoxicillin/clavalacnic acid and trimethoprim/sulfamethoxazole
[9] Report with "Resistance to beta-lactam antimicrobials may develop in *Aeromonas* species during therapy. Choosing a non-beta-lactam antimicrobial and considering combination therapy is recommended for serious infections. Consultation with infectious diseases or medical microbiology is advised."

[10] Report MIC with comment \Shig "This isolate has a ciprofloxacin MIC of mg/L. There is the risk of ciprofloxacin treatment failures in infections caused by ciprofloxacin-susceptible Shigella with ciprofloxacin MICs between 0.125 and 1mg/L. Consultation with medical microbiology or infectious diseases is advised."
[11] Do not report on patients <2 months old
[12] Report with "Susceptibility testing has been reported for surveillance purposes. Please only treat with antimicrobials if deemed clinically warranted (eg. Immunocompromised patients, severe disease, etc.). Unnecessary treatment may select for resistant strains."
[13] Report if I/R to all: Ampicillin, Trimethoprim/Sulfa, Ciprofloxacin, Azithromcyin
[14] Report if I/R to all: Ampicillin, Trimethoprim/Sulfa, Ciprofloxacin

Note: ***E. coli* O157, *Campylobacter* spp., and *Yersinia* spp. –** DO NOT report susceptibility result. Report with ISOLATE comment "In vitro susceptibility testing for this organism is not routinely performed and/or is unreliable. If advice on antimicrobial therapy is required, please contact the Medical Microbiologist".

If all antimicrobial agents are resistant, inform the Microbiologist on-call.

Respiratory and Miscellaneous Non-Sterile Sites – Gram Positive Susceptibility Reporting – 1 – *Staphylococcus*

Antimicrobial Agent	*Staphylococcus* species	MRSA
Cefazolin	X[2]	X[2, 6]
Ceftobiprole		X[13]
Clindamycin	X	X[6, 12]
Cloxacillin	X[2]	X[2, 6]
Doxycycline	X[5 10]	X[3, 5, 10]
Erythromycin	X	X[6]
Mupirocin		X[3, 8]
Rifampin		X[3]
Tigecycline	X[9]	X[9]
Trimethoprim/Sulfa	X[11]	X[11]

Vancomycin	X[1, 4]	X[6, 4]

[1] Report if Oxacillin R
[2] Base on Oxacillin/Cefoxitin result; for *Staphylococcus pseudointermedius* base on Oxacillin result
[3] For Infection Control Screen, include Isolate Comment "Susceptibility results are provided for infection control purposes only."
[4] For *S. aureus* or MRSA, vancomycin MIC=2.0 mg/L, result with ISOLATE comment: "This isolate has a vancomycin MIC of 2 mg/L which is associated with an increased risk of vancomycin treatment failures. Consultation with infectious diseases or medical microbiology is advised."
[5] Report only for age >7 y; base on Tetracycline result. DO NOT report on respiratory specimen.
[6] DO NOT report if isolated from Infection Control Screen.
[8] For KB result that shows any zone, report as Isolate Comment "No high-level mupirocin resistance detected"
For KB result that shows no zone, report as Isolate Comment "High-level mupirocin resistance detected"
[9] Report if I/R to All other antimicrobial agents OR if requested.
[10] If I/R, add comment "Doxycycline results are based on testing tetracycline which may overcall doxycycline resistance. If you wish this isolate to be tested with doxycycline directly, please contact the microbiology laboratory."
[11] Base on KB result if Vitek = I/R, do not report on patients <2 months old
[12] Do not report if Vitek result = ICR-neg/clindamycin=S/erythromycin=R. Report with comment: "If clindamycin susceptibility testing is required, please contact the microbiology laboratory within 48 hours."
[13] Report with comment:
There are no CLSI standards for this drug. EUCAST suggests MICs <=2 mg/L correlate with susceptibility. Please consult the microbiologist-on-call with any questions. For research use only.
There are no CLSI standards for this drug. EUCAST suggests MICs >2 mg/L correlate with resistance. Please consult the microbiologist-on-call with any questions. For research use only.

Note: For organisms isolated from **ears and eyes** and susceptibility result is reported, add comment "These susceptibility testing results are based on guidelines for systemic antimicrobial agents and may not accurately represent activity of topical agents."
If all antimicrobial agents are resistant, inform the Microbiologist on-call.

Respiratory and Miscellaneous Non-Sterile Sites – Gram Positive Susceptibility Reporting – 2 – *Enterococcus, Streptococcus, Corynebacterium* spp.[22], *Bacillus* spp.[22], viridans *Streptococcus*[22], *Listeria* spp.[20], *Aerococcus* species [21]

Antimicrobial Agent	*Enterococcus*[1]	*S. pneumoniae*	Group A, B, C, G *Streptococcus*	*S. anginosus* group	*Aerococcus* species
			Routinely not tested.[7] For special request:	Routinely not tested.[18] For special request:	Routinely not tested. [21]
Ampicillin	X				
Ceftriaxone-meningitis		X[6]			
Ceftriaxone-non-meningitis		X[6]			
Ceftriaxone				X[6]	
Clindamycin		X[4]	X[4, 12]	X	
Daptomycin	X[14]				
Doxycycline	X[23]				
Erythromycin		X[4, 5]	X[2, 4, 12]	X	
Levofloxacin	X[23]	X[10, 11]	X[2, 11, 12, 15]	X	
Linezolid	X[13]				
Moxifloxacin		X[9, 11]			
Penicillin G		X[8]		X	
Penicillin-oral		X[17]			
Penicillin-IV meningitis		X[17]			
Pencillin-IV non-meningitis		X[17]			
Tigecycline	X[19]				
Vancomycin	X[3]	X[6]	X[12, 16]		

[1] If isolated from Infection Control Screening test, include Isolate Comment "Susceptibility results are provided for infection control purposes only.

[2] DO NOT report on GBS Screen or vaginal swab

[3] *E. gallinarum and E. casseliflavus,* report as **R** with the statement "This organism always has intrinsic non-transmissible resistance to vancomycin. The patient does not require isolation."

[4] Report as R if D-zone is present

[5] Report Erythromycin for respiratory and eye specimens

[6] Report if Pen I or R

[7] Report "This organism is intrinsically susceptible to penicillin. If treatment is required and this patient cannot be treated with penicillin, please contact the Microbiology Department within 48 hours to request sensitivity testing."

[8] Base on Oxacillin result if S. **OR**
if Oxacillin is R, base on Penicillin MIC

[9] Base on Levofloxacin result. Report on MSH and UHN patients.

[10] DO NOT report on MSH, UHN patients.
[11] Adults only (>18 yrs)
[12] Report with additional isolate comment "Susceptibility completed as requested" (do not remove original comments).
[13] If Vancomycin and Ampicillin are R except for *E. gallinarum* and *E. casseliflavus.*
[14] If requested (excluding respiratory), base on etest result.
[15] If Levofloxacin is R or patient is <18y, consult the Microbiologist.
[16] Report only if either Clindamycin or Erythromycin are I or R.
[17] Base on Penicillin MIC from TREK.
[18] **"***Streptococcus anginosus* group are generally susceptible to penicillin, clindamycin, and levofloxacin. If susceptibility testing for this organism is required, please contact the microbiology laboratory within 48 hours."
[19] Report if I/R to **All** other antimicrobial agents **OR** if requested.
[20] ***Listeria* species** – DO NOT report susceptibility result. Report with ISOLATE comment –"Routine in vitro susceptibility testing of this organism is unreliable. *Listeria* spp. Should be considered resistant to all cephalosporins. The recommended regimen for therapy is ampicillin. If additional advice on antimicrobial therapy is required, please contact the Medical Microbiologist."
[21] "Aerococcus species are usually susceptible to beta-lactams and vancomycin. If you would like susceptibility testing to be completed, please contact the Microbiology Laboratory."
[22] ***Corynebacterium* species, *Bacillus* species, viridans *Streptococcus* –** DO NOT report susceptibility result. Report with ISOLATE comment "In vitro susceptibility testing for this organism is not routinely performed and/or is unreliable. If advice on antimicrobial therapy is required, please contact the Medical Microbiologist".
For organisms isolated from **ears and eyes** and susceptibility result is reported, add comment "These susceptibility testing results are based on guidelines for systemic antimicrobial agents and may not accurately represent activity of topical agents."
If levofloxacin or doxycycline is requested on viridans streptococcus, setup and report as per sterile site.
[23] If requested, setup and report as per sterile site.

If all antimicrobial agents are resistant, inform the Microbiologist on-call.

Respiratory and Miscellaneous Non-Sterile Sites – Gram Negative Susceptibility Reporting – 1 – Enterobacterales, *Acinetobater* species, *Pseudomonas aeruginosa*, *Aeromonas* species [25]

Antimicrobial Agent	*Enterobacterales*[16] excluding *Salmonella* spp. And *Shigella* spp.	*Salmonella* spp.	*Shigella* spp.	*Acinetobacter* spp.[16]	*Pseudomonas aeruginosa*[16]	*Aeromonas* spp.[23]
Amikacin	X[13]			X[13]	X[13]	X[13]
Ampicillin	X[2, 9, 10]	X	X			
Amoxicillin/Clavulanate	X[11]					
Azithromycin		X[26] *(S.typi **only**)*	X			
Aztreonam	X[31]				X[17]	
Cefazolin	X[3, 9, 10, 11]					
Cefiderocol	X[31]					
Cefepime	X[31]				X[17]	
Ceftazidime				X[4]	X	
Ceftazidime-avibactam	X[31]				X[17]	
Ceftazidime-avibactam+Aztreonam combination	X[22,29], [30]				X22,29	
Ceftriaxone	X[7, 9, 10, 28]	X[28]	X[28]	X[28]		X[28]
Ceftolozane/ Tazobactam	X[14,31]				X[17]	
Chloramphenicol	X[31]					X[22]
Ciprofloxacin	X[1]	X[1]	X[1]	X[1]	X[1]	X[1]
Colistin	X[31]			X[15]	X[15]	
Doxycycline	X[31]	X[27]	X[27]			
Ertapenem	X[5]	X[27]	X[27]			
Fosfomycin	X[31]				X19	
Gentamicin	X[11]			X		X
Imipenem-Relebactam	X[22]				X[22]	
Minocycline	X[31]					
Meropenem	X[5]	X[27]	X[27]	X	X[12]	X[8]
Meropenem-Vaborbactam	X[22]				X[22]	
Piperacillin/Tazobactam	X[7, 9, 11]			X	X	X
Tetracycline	X[14,20, 31]					X[20], [21]
Tigecycline	X[14, 31]			X[14]		
Trimethoprim/Sulfa [24]	X	X	X	X		X
Tobramycin	X[11]			X	X	

[1] Adults only (>18 y)
[2] Always report *Klebsiella* spp., and SPICE as R.
[3] Always report SPICE as R.
[4] Report only if R. For *Enterobacterales* if cefotaxime/ceftriaxone or ceftazidime R, report both as R
[5] Report if I/R **OR** if ≤1 of the antimicrobial agents is susceptible: amikacin, ciprofloxacin, TMP/SMX, 3rd generation cephalosporins (ceftriaxone or ceftazidime) **OR** if requested
[6] Report if Genta is R
[7] For SPICE, report all penicillins and first, second and third generation cephalosporins and piperacillin/tazobactam as R; report with comment "Resistance to extended-spectrum penicillins, beta-lactam/beta-lactamase inhibitor combinations (e.g. piperacillin-tazobactam), and cephalosporins may develop during therapy. These agents should be avoided and will be reported as resistant regardless of their in vitro susceptibility results. If you have questions, please contact the medical microbiologist on call."
[8] Report only if I or R
[9] For *E. coli, Klebsiella* species and *Proteus* species that are confirmed to have an ESBL of any class, report all penicillins and first, second and third generation cephalosporins and piperacillin/tazobactam as R.
[10] For *Enterobacterales* other than *E. coli, Klebsiella* species and *Proteus* species where ESBL testing is not done, if any one of cefotaxime/ceftriaxone or ceftazidime=I/R, report all penicillins and first, second and third generation cephalosporins and piperacillin/tazobactam as R
[11] Do not report for *Salmonella* species.
[12] Report if I/R **OR** if I/R to 3 of the 4 antimicrobial agents:aminoglycosides, ciprofloxacin, 3rd Generation Cephalosporins, Piperacillin/tazobactam **OR** if requested
[13] Report if both Gentamicin and Tobramycin are I/R.
[14] Report with comment if I/R to **All** other Antimicrobial Agents **OR** if only aminoglycoside is S **OR** if requested.
[15] Report if I/R to **All** other Antimicrobial Agents **OR** if requested.
[16] If isolated from Infection Control Screening test, include Isolate Comment "Susceptibility results are provided for infection control purposes only."
[17] Report if I/R to **all** routinely tested antimicrobials disregarding aminoglycosides and colistin)
[18] Report if I/R to **All** other Antimicrobial Agents **including** aminoglycoside **OR** if requested.
[19] Report with comment if I/R to **All** other Antimicrobial Agents (disregarding aminoglycosides and colistin) **OR** if requested.
[20] Report only for age >7 y
[21] Report if I/R to **All** ciprofloxacin, amoxicillin/clavulanic acid and trimethoprim/sulfamethoxazole
[22] Report if I/R to **All** other Antimicrobial Agents (disregarding aminoglycosides and colistin)
[23] Report with "Resistance to beta-lactam antimicrobials may develop in *Aeromonas* species during therapy. Choosing a non-beta-lactam antimicrobial and considering combination therapy is recommended for serious infections. Consultation with infectious diseases or medical microbiology is advised."
[24] Do not report on patients <2 months old
[25] *Pseudomonas* species (other than *P. aeruginosa*), fastidious Gram-negative bacteria & non–fermenters – Send to PHOL and report results as tested.
[26] Report for *Salmonella typhi* ONLY
[27] Report if I/R to ceftriaxone
[28] Report only for age >1m
[29] Report CZA+ATM combination result based on double disc diffusion between CZA and ATM
[30] Report if I/R to All other Antimicrobial Agents **AND** if oxa-48, NDM, VIM or IMP detected
[31] Report if I/R to all **routinely** tested antimicrobials (***disregarding*** *aminoglycosides*)

For organisms isolated from **ears and eyes** and susceptibility result is reported, add comment "These susceptibility testing results are based on guidelines for systemic antimicrobial agents and may not accurately represent activity of topical agents."

If all antimicrobial agents are resistant, inform the Microbiologist on-call.

Respiratory and Miscellaneous Non-Sterile Sites – Gram Negative Susceptibility Reporting – 2 – *Haemophilus* species, *M. catarrhalis, S. maltophilia, B. cepacia, Pseudomonas* species (other than *P. aeruginosa*)[13], fastidious gram-negative bacteria, non-fermenters, *Neisseria meningitides, H. pylori.*

Antimicrobial Agent	*Haemophilus* species	*S. maltophilia**	*Burkholderia cepacia*	*H. pylori* [8]
Amoxicillin				
Beta-lactamase	X[2]			
Cefiderocol		X[6]		
Ceftazidime			X	
Ceftazidime-avibactam+Aztreonam combination		X[6, 15]		
Ciprofloxacin				
Clarithromycin				
Colistin		X[3]	X[3]	
Levofloxacin		X[1, 4]		
Meropenem			X	
Metronidazole				
Minocycline		X[14]	X[14]	
Rifampin				
Tetracycline				
Tigecycline		X[6]		
Trimethoprim/Sulfa [12]		X	X	

[1] Adults only (>18 y)
[2] If beta-lactamase is negative, report with comment "This isolate is beta-lactamase negative. Beta-lactamase negative isolates are generally susceptible to amoxicillin.Susceptibility testing can be completed if requested."
If beta-lactamase is positive, report with comment "This isolate is beta-lactamase positive. Beta-lactamase positive isolates are resistant to ampicillin but generally susceptible to amoxicillin-clavulanic acid. Susceptibility testing can be completed if requested."
[3] Report with comment if I/R to all other drugs; report without interpretation;
[4] Report with comment "NOTE: There are no standardized interpretive breakpoints for *Stenotrophomonas maltophilia* and moxifloxacin but in general, levofloxacin and moxifloxacin minimum inhibitory concentrations (MICs) correlate well with each other. Ref: J Chemother. 2008 Feb;20(1):38-42."
[5] Report with comment if I/R to all other drugs base on PHL MIC result
[6] Report with comment if I/R to all other drugs
[8] Report all Mayo Clinic MIC results:
Report Mayo clinic MIC for antimicrobial agent within isolate comment:
"Amoxicillin MIC ____mg/L
Metronidazole MIC ____mg/L
Ciprofloxacin MIC ____mg/L

Clarithromycin MIC ____mg/L
Tetracycline MIC ____mg/L
as reported by the Mayo Clinic Mayo Medical Laboratories
Rochester Main Campus, 200 First Street SW, Rochester, MN
55905. Mayo Clinic Specimen No_____.

There are no CLSI standards for the following drugs. EUCAST ECOFF states the following MICs correlate with wild-type organisms:

Amoxicillin MIC <= 0.125 mg/L
Metronidazole MIC <= 8 mg/L
Levofloxacin MIC <= 1 mg/L*
Tetracycline MIC <=1 mg/L
*There is not a wild-type ECOFF for ciprofloxacin."

[12] Do not report on patients <2 months old

[13] *Pseudomonas* species (other than *P. aeruginosa*), fastidious Gram-negative bacteria & non–fermenters – Send to PHOL and report results as tested.

[14] Adults only (>13 y)

[15] Report CZA+ATM combination result based on double disc diffusion between CZA and ATM

*For *Stenotrophomonas maltophilia* isolated from **all** sites and susceptibility result is reported, add comment "If treatment is deemed clinically warranted for *S. maltophilia*, combination therapy with two effective antimicrobials is recommended until clinical improvement is observed.

For organisms isolated from **ears and eyes** and susceptibility result is reported, add comment "These susceptibility testing results are based on guidelines for systemic antimicrobial agents and may not accurately represent activity of topical agents."

If all antimicrobial agents are resistant, inform the Microbiologist on-call.

Spinal Fluids, including VP shunts and Brain Tissue – Gram Positive Susceptibility Reporting

Antimicrobial Agent	*Staphylococcus* species	*Enterococcus* species	*Strep. Pneumoniae*	viridans *Strep.* *S. bovis*	*Strep. Anginosus* group	Group A,B,C,G *Streptococcus*
Ampicillin		X				
Ceftobiprole	X[17]					
Ceftriaxone				X[4]	X[4]	
Ceftriaxone-meningitis			X			
Cloxacillin	X[15]					
Daptomycin	X[12]	X[13]				
HLGR[3]		X				
HLSR[3]		X[2]				
Linezolid	X[12]	X[7]				
Penicillin IV-meningitis			X	X	X	X
Tigecycline	X[12]					
Trimethoprim/Sulfa	X[14, 16]					
Vancomycin	X[1, 10]	X[6]	X	X[4]	X[4]	X[4]

[1] Report if Oxacillin R
[2] Report only if requested.
[3] HLGR = High Level Gentamicin Resistant; HLSR = High Level Streptomycin Resistant. Report based on HLGR using canned message (See Blood HLGR Results Reporting).
[4] Report only if Pen is I/R
[5] Report based on Ampicillin result
[6] *E. gallinarum and E. casseliflavus* report as **R** with the statement: "This organism always has intrinsic non-transmissible resistance to vancomycin. The patient does not require isolation."
[7] Report if Vancomycin and Ampicillin are R except *E. gallinarum* and *E. casseliflavus*.
[10] For *S. aureus* or MRSA, vancomycin MIC=2.0 mcg/L, result with ISOLATE comment: "This isolate has a vancomycin MIC of 2 mg/L which is associated with an increased risk of vancomycin treatment failures. Consultation with infectious diseases or medical microbiology is advised."
[12] Report if I/R to **All** other antimicrobial agents **OR** if requested.
[13] Report if requested, base on etest result
[14] Base on KB result if Vitek = I/R, do not report SXT on patients <2 months old
[15] Base on Oxacillin/cefoxitin result; for *Staphylococcus pseudointermedius* base on Oxacillin result
[16] Report if I/R to Ceftriaxone
[17] For MRSA only; report with comment:
There are no CLSI standards for this drug. EUCAST suggests MICs <=2 mg/L correlate with susceptibility. Please consult the microbiologist-on-call with any questions. For research use only.
There are no CLSI standards for this drug. EUCAST suggests MICs >2 mg/L correlate with resistance. Please consult the microbiologist-on-call with any questions. For research use only.

Note: ***Listeria* species** – DO NOT report susceptibility result. Report with ISOLATE comment –"Routine in vitro susceptibility testing of this organism is unreliable. *Listeria* spp. Should be considered resistant to all cephalosporins. The recommended regimen for therapy is ampicillin. If additional advice on antimicrobial therapy is required, please contact the Medical Microbiologist."

***Corynebacterium* species, *Bacillus* species** – DO NOT report susceptibility result. Report with ISOLATE comment "In vitro susceptibility testing for this organism is not routinely performed and/or is unreliable. If advice on antimicrobial therapy is required, please contact the Medical Microbiologist".
If all antimicrobial agents are resistant, inform the Microbiologist on-call

Spinal Fluids, including VP shunts and Brain Tissue – Gram Negative Susceptibility Reporting – 1 – Enterobacterales and *Acinetobacter* spp., *Salmonella* species including *S. typhi*, *Shigella* species

Antimicrobial Agent	Enterobacterales excluding *Salmonella* species and *Shigella* species	*Acinetobacter* species	*Salmonella* species including *S. typhi*	*Shigella* species	*Aeromonas* species[17]
Amikacin		X[8]			X[8]
Ampicillin	X[4, 5, 6]		X	X	
Ceftazidime		X[2]			
Ceftolozane/Tazobactam	X[13]				
Ceftriaxone	X[5, 6, 7]	X	X	X	X
Chloramphenicol	X[13]	X[13]	X[13]	X[13]	X[13]
Ciprofloxacin	X[1] X[3]	X[1] X[3]	X[1] X[3]	X[1] X[3]	X[1] X[3]
Colistin	X[13]	X[12]	X[13]	X[13]	X[13]
Gentamicin	X[16]	X[16]	X[16]	X[16]	X[16]
Meropenem	X[3]	X[3] X[12]	X[3]	X[3]	X[3]
Tetracycline					
Trimethoprim/Sulfa	X[9]	X[9]	X[9]	X[9]	X[9]
Tobramycin		X[9]			

[1] Adults only (>18 y)
[2] Report only if R.
[3] Report if I/R to ceftriaxone **OR or if I/R to meropenem or** if requested.
[4] Always report *Klebsiella* spp., and SPICE as R.
[5] For *E. coli*, *Klebsiella* species and *Proteus* species that are confirmed to have an ESBL of any class, report all penicillins and third generation cephalosporins and piperacillin/tazobactam as R.
[6] For *Enterobacterales* other than *E. coli*, *Klebsiella* species and *Proteus* species where ESBL testing is not done, if any one of cefotaxime/ceftriaxone or ceftazidime=R, report all penicillins and third generation cephalosporins as R
[7] For SPICE, report all penicillins and first, second and third generation cephalosporins and piperacillin/tazobactam as R; report with comment "Resistance to extended-spectrum penicillins, beta-lactam/beta-lactamase inhibitor combinations (e.g. piperacillin-tazobactam), and cephalosporins may develop during therapy. These agents should be avoided and will be reported as resistant regardless of their in vitro susceptibility results. If you have questions, please contact the medical microbiologist on call.."
[8] Report if both Gentamicin and Tobramycin are I/R
[9] do not report SXT on patients <2 months old
13 Report on A. baumanii complex only
[13] Report with comment if I/R to **All** other antimicrobial agents (disregarding aminoglycosides) **OR** if requested.
14Report only for age>7 y
D. Report if I/R to Report with omment if I/R to **All** other routinely tested Antimicrobial Agents
[17] Report with "Resistance to beta-lactam antimicrobials may develop in Aeromonas species during therapy. Choosing a non-beta-lactam antimicrobial and considering combination therapy is recommended for serious infections. Consultation with infectious diseases or medical microbiology is advised."
[18] Report if I/R to ceftriaxone

Note: If all antimicrobial agents are resistant, inform the Microbiologist on-call

Spinal Fluids, including VP shunts and Brain Tissue – Gram Negative Susceptibility Reporting – 2 – *Pseudomonas aeruginosa*, *Pseudomonas* spp. (other than P. Aeruginosa), fastidious Gram-negative bacteria, non-fermenters, *M. Catarrhalis*, *N. Meningitidis*, *Stenotrophomonas maltophilia*, *Burkholderia cepacia*, *Haemophilus* species. [9]

Antimicrobial Agent	*P. aeruginosa*	*S. maltophilia**	*B. cepacia*	*Haemophilus* species
Amikacin	X[3]			
Ampicillin				X[1]
Aztreonam	X[7]			
Cefepime	X[7]			
Cefiderocol		X[4]		
Ceftazidime	X		X	
Ceftazidime-avibactam	X[7]			
Ceftazidime-avibactam+Aztreonam combination		X[4],[11]		
Ceftolozane/Tazobactam	X[7]			
Ceftriaxone				X
Colistin	X[5]	X[4]	X[4]	
Meropenem	X[2]		X	
Trimethoprim/Sulfa		X[8]	X[8]	
Tobramycin	X[8]			

[1] Base on beta-lactamase result and KB Ampicillin
[2] Report if I/R **OR** if I/R to 3 of the 4 antimicrobial agents:aminoglycosides , ciprofloxacin, 3rd Generation Cephalosporins, Piperacillin/tazobactam **OR** if requested
[3] Report if both Gentamicin and Tobramycin are R.
[4] Report with comment if I/R to all other drugs
[5] Report if I/R to **All** routinely tested antimicrobial agents (disregarding aminoglycosides)
[6] Report with comment if I/R to all other drugs base on PHL MIC result
[7] Report if I/R to **all** routinely tested antimicrobials (disregarding aminoglycosides and colistin))
[8] Report if I/R to Ceftriaxone, do not report SXT on patients <2 months old
[9] *Pseudomonas* species (other than *P. aeruginosa*), fastidious Gram-negative bacteria & non–fermenters – Send to PHOL and report results as tested.
[10] For ***M. catarrhalis*** – DO NOT report susceptibility result. Report with ISOLATE comment: "The majority of *Moraxella catarrhalis* are resistant to ampicillin. In vitro susceptibility testing for this organism is not routinely performed and/or is unreliable. If advice on antimicrobial therapy is required, please contact the Medical Microbiologist".
[11] Report CZA+ATM combination result based on double disc diffusion between CZA and ATM

*For *Stenotrophomonas maltophilia* isolated from all sites and susceptibility result is reported, add comment "If treatment is deemed clinically warranted for *S. maltophilia*, combination therapy with two effective antimicrobials is recommended until clinical improvement is observed.

If all antimicrobial agents are resistant, inform the Microbiologist on-call.

Blood and other Sterile Sites, excluding Spinal Fluids/VP Shunts or Brain Tissue – Gram Positive Susceptibility Reporting – 1 – *Staphylococcus aureus, Staphylococcus lugdunensis, Enterococcus*, Other CNST from sterile sites

Antimicrobial Agent	*Staphylococcus aureus, Staphylococcus lugdunensis*, Other CNST (from sterile sites and bloods if requested)	*Enterococcus* species
Ampicillin		X[9]
Cefazolin	X[2]	
Ceftobiprole	X[15]	
Cloxacillin	X[2]	
Daptomycin	X[10]	X[7]
Doxycycline	X[13, 14]	X[17]
HLGR[3]		X
HLSR[3]		X[4]
Levofloxacin		X[16]
Linezolid	X[10]	X[7]
Moxifloxacin	X[13]	
Piperacillin/Tazobactam		~~X~~[1]
Rifampin	X[12]	
Tigecycline	X[10]	X[10]
Vancomycin	X[8, 5]	X[6]

[2] Base on Oxacillin/cefoxitin result; for *Staphylococcus pseudointermedius* base on Oxacillin result
[3] HLGR = High Level Gentamicin Resistant; HLSR = High Level Streptomycin Resistant
Report based on HLGR using canned message (See Blood HLGR Results Reporting).
[4] Report only if requested.
[5] For *S. aureus* or MRSA, vancomycin MIC=2.0 mg/L, result with ISOLATE comment: "This isolate has a vancomycin MIC of 2 mg/L which is associated with an increased risk of vancomycin treatment failures. Consultation with infectious diseases or medical microbiology is advised."
[6] *E. gallinarum and E. casseliflavus* report as **R** with the statement "This organism always has intrinsic non-transmissible resistance to vancomycin. The patient does not require isolation."
[7] Report if Vancomycin and Ampicillin are R OR if the isolate is *E. gallinarum* or *E. casseliflavus*.
[8] Only if Oxacillin=R.
[9] If beta-lactamase testing is requested and is positive, report Ampicillin as R
[10] Report if I/R to **All** other antimicrobial agents **OR** if requested.
[11]
[12] Report if requested with comments: "This organism is susceptible to rifampin. Rifampin should NOT be used as monotherapy given the risk of resistance. If rifampin combination therapy is being considered, consultation with infectious diseases or medical microbiology is advised." "This organism is intermediate to rifampin." OR "This organism is resistant to rifampin."
13 Report on Bone or Joint and sterile site specimens. **DO NOT** report on blood culture.

[14] If doxycycline is I/R, include comment "Doxycycline results are based on testing tetracycline which may overcall doxycycline resistance. If you wish this isolate to be tested with doxycycline directly, please contact the microbiology laboratory." Do not report on blood.

[15] For MRSA only; report with comment:
There are no CLSI standards for this drug. EUCAST suggests MICs <=2 mg/L correlate with susceptibility. Please consult the microbiologist-on-call with any questions. For research use only.
There are no CLSI standards for this drug. EUCAST suggests MICs >2 mg/L correlate with resistance. Please consult the microbiologist-on-call with any questions. For research use only.

[16] Report on bone/joint samples (or when requested) without interpretation. Use comment "Levofloxacin zone size = xx mm. There are no levofloxacin CLSI breakpoints for enterococci isolated from this specimen type. CLSI suggests zone size >=17mm correlate with susceptibility for enterococci isolated from urine specimens."
(or from 14 to 16mm correlate with intermediate susceptibility)
(or <= 13mm correlate with resistance)

[17] Report on bone/joint samples (or when requested), DO NOT report on blood culture.

Note: If all antimicrobial agents are resistant, inform the Microbiologist on-call.

Blood and other Sterile Sites, excluding Spinal Fluids/VP Shunts or Brain Tissue – Gram Positive Susceptibility Reporting – 2 – *S. pneumoniae*, viridans *Streptococcus*, *Streptococcus bovis*, *S. anginosus* group, Group A, B, C, G *Streptococcus*, *Listeria* species, *Corynebacterim* species, *Bacillus* species

Antimicrobial Agent	*S. pneumoniae*	viridans *Strep.* *Strep. Bovis group*	*S. anginosus* group	Group A, B, C,G *Streptococcus*
Ceftriaxone		X[5]	X[5]	
Ceftriaxone-meningitis	X			
Ceftriaxone-non-meningitis	X			
Clindamycin	**X[3,]**	**X[3,]**	**X[3,]**	X[3]
Doxycycline		X[10]	**X[10]**	**X[10]**
Erythromycin	X			X[3]
Levofloxacin	X[1, 8]	X[9]	**X[9]**	**X[9]**
Moxifloxacin	X[1, 7]			
Penicillin-IV meningitis	X	X[4]	X[4]	X
Penicillin-IV non-meningitis	X			
Penicillin-oral	X			
Vancomycin	X[5]	X[5]	X[5]	X[5]

[1] Adults only (>18 y)
[2]
[3] Report as R if D-zone is present.
[4] For viridans S*treptococcus, S. bovis* and *S. anginosus*, report MIC value as Isolate Comment **only** when from a Blood Culture or heart tissue specimen (eg. Heart valve, vegetation, pericardial fluid).
[5] Report only if Pen I or R
[6]
[7] Report on MSH and UHN patients only.
[8] DO NOT report on MSH and UHN patients.
[9] Report on bone/joint samples and sterile specimens or when requested DO NOT report on blood culture
[10] Report on bone/joint samples (or when requested) based on tetracycline result. DO NOT report on blood culture.
If doxycycline is I/R, include comment "Doxycycline results are based on testing tetracycline which may overcall doxycycline resistance."

Note: ***Listeria* species** – DO NOT report susceptibility result. Report with ISOLATE comment –"Routine in vitro susceptibility testing of this organism is unreliable. *Listeria* spp. should be considered resistant to all cephalosporins. The recommended regimen for therapy is ampicillin. If additional advice on antimicrobial therapy is required, please contact the Medical Microbiologist."
***Corynebacterium* species, *Bacillus* species**. – DO NOT report susceptibility result. Report with ISOLATE comment "In vitro susceptibility testing for this organism is not routinely performed and/or is unreliable. If advice on antimicrobial therapy is required, please contact the Medical Microbiologist".
If all antimicrobial agents are resistant, inform the Microbiologist on-call.

Blood and other Sterile Sites, excluding Spinal Fluids/VP Shunts or Brain Tissue – Gram Negative Susceptibility Reporting -1 – Enterobacterales and *Acinetobacter* spp., *Salmonella* species including *S. typhi*, *Shigella* species

Antimicrobial Agent	Enterobacterales excluding *Salmonella* species and *Shigella* species	*Acinetobacter* spp.	*Salmonella* spp. Including *S. typhi*	*Shigella* species	*Aeromonas* species[17]
Amikacin	X[14]	X[14]			X[14]
Ampicillin	X[1, 10, 11]		X	X	
Amoxicillin/ Clavulanate	X				
Azithromycin			X[19] (S.Typhi ONLY)	X	
Aztreonam	X[9, 23]				
Cefazolin	X[2, 10, 11, 16]				
Cefiderocol	X[23]				
Cefepime	X[23]				
Ceftazidime		X[5, 8, 11]			
Ceftazidime-avibactam	X[23]				
Ceftazidime-avibactam+Aztreonam combination	X[21, 22]				
Ceftolozane/ Tazobactam	X[9, 23]				
Ceftriaxone	X[10, 11, 12]	X[11]	X	X	X
Chloramphenicol	X[9, 23]				X[13]
Ciprofloxacin	X[4]	X[4]	X[4]	X[4]	X[4]
Colistin	X[23]	X[15] (A. baumanii complex only)			
Doxycycline	X[9,23]		X[20]	X[20]	
Ertapenem	X[7]		X[20]	X[20]	
Fosfomycin	X[23]				
Gentamicin	X	X			X
Imipenem-Relebactam	X[13]				
Meropenem	X[7]	X	X[20]	X[20]	X[5]
Meropenem-Vaborbacam	X[13]				
Minocycline	X[9,23]				
Piperacillin/Tazobactam	X[10, 12]	X			X
Tetracycline	X[3, 9, 23]				X[3,6]
Tigecycline	X[9,23]	X[9]			
Trimethoprim/Sulfa [18]	X	X	X	X	X
Tobramycin	X	X			

[1] Always report *Klebsiella* spp., and SPICE as R.
[2] Always report SPICE as R.
[3] Report only for age>7 y
[4] Adults only (>18 y)
[5] Report only if I or R
[6] Report if I/R to **All** ciprofloxacin, amoxicillin/clavalacnic acid and trimethoprim/sulfamethoxazole
[7] Report if I/R **OR** if I/R to 3 of the 4 antimicrobial agents: amikacin, ciprofloxacin, 3rd Generation Cephalosporins, Septra **OR** if requested
[8] Always report for PMH
[9] Report with comment if I/R to **All** other Antimicrobial Agents **OR** if only aminoglycoside is S.
[10] For *E. coli*, *Klebsiella* species and *Proteus* species that are confirmed to have an ESBL of any class, report all penicillins and first, second and third generation cephalosporins and piperacillin/tazobactam as R.
[11] For *Acinetobacter* sp. And *Enterobacterales* other than *E. coli*, *Klebsiella* species and *Proteus* species where ESBL testing is not done, if any one of cefotaxime/ceftriaxone or ceftazidime=R, report all penicillins and first, second and third generation cephalosporins as R
[12] For SPICE report all penicillins and first, second and third generation cephalosporins and piperacillin/tazobactam as R; report with comment "Resistance to extended-spectrum penicillins, beta-lactam/beta-lactamase inhibitor combinations (e.g. piperacillin-tazobactam), and cephalosporins may develop during therapy. These agents should be avoided and will be reported as resistant regardless of their in vitro susceptibility results. If you have questions, please contact the medical microbiologist on call."
[13] Report if I/R to **All** other Antimicrobial Agents
[14] Report if both Gentamicin and Tobramycin are I/R
[15] Report if I/R to **All** other antimicrobial agents **OR** if only aminoglycoside is S **OR** if requested.
[16] Report from KB result ONLY. Do NOT report for *Proteus mirabilis*.
[17] Report with "Resistance to beta-lactam antimicrobials may develop in Aeromonas species during therapy. Choosing a non-beta-lactam antimicrobial and considering combination therapy is recommended for serious infections. Consultation with infectious diseases or medical microbiology is advised."
[18] Do not report on patients <2 months old
[19] Report *for Salmonella typhi* **ONLY**
[20] Report if I/R to ceftriaxone
[21] Report CZA+ATM combination result based on double disc diffusion between CZA and ATM
[22] Report if I/R to All other Antimicrobial Agents **AND** if oxa-48, NDM, VIM or IMP detected
[23] Report if I/R to all **routinely** tested antimicrobials (***disregarding*** *aminoglycosides*)

Blood and other Sterile Sites, excluding Spinal Fluids/VP Shunts or Brain Tissue – Gram Negative Susceptibility Reporting – 2 – *Pseudomonas aeruginosa, Pseudomonas* spp. (other than P. aeruginosa), fastidious gram-negative bacteria, non-fermenters, *M. catarrhalis, N. meningitidis, Stenotrophomonas maltophilia, Burkholderia cepacia, Haemophilus* species.

Antimicrobial Agent	*P. aeruginosa*	*S. maltophilia* *	*B. cepacia*	*Haemophilus* species
Amikacin	X[5]			
Ampicillin				X[1]
Aztreonam	X[9]			
Cefazolin				
Cefepime	X[9]			
Cefiderocol		X[6]		
Ceftazidime	X		X	
Ceftazidime-avibactam	X[9]			
Ceftazidime-avibactam+Aztreonam combination	X[6,12]	X[6,12]		
Ceftolozane/Tazobactam	X[9]			
Ceftriaxone				X
Ciprofloxacin	X[2]			X[2]
Colistin	X[9]	X[6]	X[6]	
Fosfomycin	X[6]			
Imipenem-relebactam	X[7]			
Levofloxacin		X[2, 8]		
Meropenem	X[4]		X	
Meropenem-vaborbactam	X[7]			
Minocycline		X[11]	X[11]	
Piperacillin/Tazobactam	X			
Tigecycline		X[6]	X[6]	
Trimethoprim/Sulfa [10]		X	X	
Tobramycin	X			

[1] Based on beta-lactamase result
[2] Adults only (>18 y)
[3] Report with comment if I/R to all other drugs base on PHL MIC result
[4] Report if I/R **OR** if I/R to 3 of the 4 antimicrobial agents: aminoglycosides, ciprofloxacin, 3rd Generation Cephalosporins, Piperacillin/tazobactam **OR** if requested
[5] Report if Tobramycin is R.
[6] Report with comment if I/R to all other drugs;
[7] Report if I/R to **All** other antimicrobial agents (disregarding aminoglycosides and colistin)
[8] Report with comment "NOTE: There are no standardized interpretive breakpoints for *Stenotrophomonas maltophilia* and moxifloxacin but in general, levofloxacin and moxifloxacin minimum inhibitory concentrations (MICs) correlate well with each other. Ref: J Chemother. 2008 Feb; 20(1):38-42."

[9] Report if I/R to **all** routinely tested antimicrobials including (disregarding aminoglycosides and colistin)
[10] Do not report on patients <2 months old
[11] Adults only (>13 y).
[12] Report CZA+ATM combination result based on double disc diffusion between CZA and ATM

Note: *Pseudomonas* species (other than *P. aeruginosa*), fastidious Gram-negative bacteria, non–fermenters report susceptibilities as per PHOL.

For ***N. meningitidis*** – DO NOT report susceptibility result. Report with ISOLATE comment "In vitro susceptibility testing for this organism is not routinely performed and/or is unreliable. If advice on antimicrobial therapy is required, please contact the Medical Microbiologist".

For ***M. catarrhalis*** – DO NOT report susceptibility result. Report with ISOLATE comment: "The majority of *Moraxella catarrhalis* are resistant to ampicillin. In vitro susceptibility testing for this organism is not routinely performed and/or is unreliable. If advice on antimicrobial therapy is required, please contact the Medical Microbiologist".

*For *Stenotrophomonas maltophilia* isolated from all sites and susceptibility result is reported, add comment "If treatment is deemed clinically warranted for *S. maltophilia*, combination therapy with two effective antimicrobials is recommended until clinical improvement is observed.

If all antimicrobial agents are resistant, inform the Microbiologist on-call.

Antimicrobial Related LIS Canned Messages

I. Introduction

Antimicrobial related canned messages are built into the Laboratory Information System to provide uniform reporting phrases to be used when certain pre-described conditions are met.

II. Procedure

E. Automatic Canned Messages:

The lists below are automatic canned messages that will appear when set conditions are met. The message will appear in a warning box when entering or before exiting an order.

1. When the message code appears, press F12 to save.
2. Continue with another F12 to save the order.
3. View the report.
4. If the same message has been saved previously (i.e. appeared more than once), go to the Isolate Comment window and delete the duplicate comment using CTRL L.
5. Re-status as required.
6. Press F12 to save the order.

LIS messages are sort below by type:

General GPC GPB GNB

General:

Ear and Eye specimens with susceptibility results

LIS Isolate Canned Message Code: **&eye**; attached to Organism classes A and B with procedures EYE and COR and with source EAR and drugs am, betalac, cc, peng, sxt.

"These susceptibility testing results are based on guidelines for systemic antimicrobial agents and may not accurately represent activity of topical agents.

For **MSH MRO's**

LIS Isolate Canned Message Code: **MRES**, attached to drug – tax **"MULTIPLE ANTIBIOTIC RESISTANT ORGANISM. THIS PATIENT MUST BE ON "CONTACT PRECAUTIONS" UNTIL FURTHER NOTICE FROM INFECTION CONTROL."**

For isolates that **susceptibility testing is not routinely performed and/or is unreliable**:

LIS Isolate Canned Message Code: **NSEN**; attached to organisms and Isolate Comment keypad.
"In vitro susceptibility testing for this organism is not routinely performed and/or is unreliable. If advice on antimicrobial therapy is required, please contact the Medical Microbiologist".

GPC:

For **MSH MRSA's**
LIS Isolate Canned Message Code: **ICPR**, attached to organism staamr **"THIS PATIENT MUST BE ON "CONTACT PRECAUTIONS" UNTIL FURTHER NOTICE."**

MRSA isolated from MRSA Screen Susceptibility Result Comment
LIS Isolate Canned Message Code: **MRSS**; linked to organism staamr, Dox=R
"Susceptibility results are provided for infection control purposes only."

- **MRSA PBP2a=negative, Oxacillin Screen=negative, Oxacillin =>4mcg/L**, isolate is a **BORSA**; report as *S. aureus* with LIS Isolate Canned Message Code: **BORS** "*This organism is a borderline-oxacillin resistant Staphylococcus aureus (BORSA) which is resistant to cloxacillin and cefazolin by a mechanism different from that in typical MRSA. Consultation with a Microbiologist or Infectious Disease physician is advised."*

For *S. aureus* vancomycin MIC=2.0 mg/L, result with ISOLATE comment:
Vva=2.0 **~\va2**
"This isolate has a vancomycin MIC of 2 mg/L which is associated with an increased risk of vancomycin treatment failures. Consultation with infectious diseases or medical microbiology is advised."

LIS Isolate Canned Message Code: **MUPz**; for KB zone size ≥19mm, linked to drug code – mup
"Mupirocin zone size = xx mm
There are no standards to interpret this result as susceptible or resistant. Published literatures suggest MICs <2 mg/L and zones of inhibition ≥19mm may correlate with susceptibility. For help with interpretation, please consult the microbiologist-on-call. (Refs: J Clin Micro 1990;28(3); 608-609; AMMI Canada 2006 abstract P2.27)."

LIS Isolate Canned Message Code: **MUP**; for MIC result, linked to drug code – mup
"Mupirocin MIC = xx mg/L
There are no standards to interpret this result as susceptible or resistant. Published literatures suggest MICs <2 mg/L and zones of inhibition ≥19mm may correlate with susceptibility. For help with interpretation, please consult the microbiologist-on-call. (Refs: J Clin Micro 1990;28(3); 608-609; AMMI Canada 2006 abstract P2.27)."

LIS Isolate Canned Message Code: \icr-;
For MRSA isolated from non-sterile sites: ICR-neg/clindamycin=S/erythromycin=R.

Report with comment: "If clindamycin susceptibility testing is required, please contact the microbiology laboratory within 48 hours."

For *S. aureus* reporting Tigercycline – messages link to Organism classes D, code **\TIGD**

"Tigecycline MIC is xx mg/L.

There are no CLSI standards for this drug. EUCAST suggests MICs <=0.5 mg/L correlate with susceptibility. Result for tigecycline is based on Etest gradient strips (bioMérieux) which have been validated with well-characterized laboratory (ATCC) strains. Verification on clinical isolates against a gold standard method has been limited. Please take this into consideration when interpreting these results. Please consult the microbiologist-on-call with any questions."

Coagulase-negative staphylococci, not *S. lugdunensis* from Blood Cultures:
LIS Isolate Canned Message Code: **\cnst**; linked to organism code – staneg
"Coagulase-negative staphylococci may be blood culture contaminants; clinical correlation is needed to determine the significance of this result. The vast majority of coagulase-negative staphylococci are susceptible to vancomycin; susceptibility testing will only be completed if requested."

***S. lugdunen*sis from Blood Cultures**:
LIS Isolate Canned Message Code: **\slug**; linked to organism code – stalug
"*S. lugdunensis* is a virulent coagulase-negative staphylococcus that is associated with abscesses, native valve endocarditis, and other serious infections. Consultation with infectious diseases is recommended."

***Staphylococcus saprophyticus* and CNST from urine**
LIS Isolate Canned Message Code: **\ssap**; attached to organism code stasap.
"Susceptibility testing of this organism is not routinely done because infections respond to concentrations achieved in urine of antimicrobial agents commonly used to treat acute, uncomplicated urinary tract infections e.g. nitrofurantoin, trimethoprim/sulfa or fluoroquinolones. Suggest repeat specimen with request for susceptibility testing if patient does not respond to empiric therapy."

***Staphylococcus aureus* or MRSA where doxycycline is reported as R**
LIS Isolate Canned Message Code: **\doxyR**; attached to organism code staaur and staamr
"Doxycycline results are based on testing tetracycline which may overcall doxycycline resistance. If you wish this isolate to be tested with doxycycline directly, please contact the microbiology laboratory."
For **MRSA** if **Ceftobiprole** is requested:
MIC <=2 mg/L – code **/BPRS**

There are no CLSI standards for this drug. EUCAST suggests MICs <=2 mg/L correlate with susceptibility. Please consult the microbiologist-on-call with any questions. For research use only.
MIC >2 mg/L – code **/BPRR**
There are no CLSI standards for this drug. EUCAST suggests MICs >2 mg/L correlate with resistance. Please consult the microbiologist-on-call with any questions. For research use only.

***ß-haemolytic Streptococcus* Groups A, B, C and G**
LIS Isolate Canned Message Code: **\GBS**; attached to organism straga, strpyo, strgrc, strgrg.
"This organism is intrinsically susceptible to penicillin. If treatment is required AND this patient cannot be treated with penicillin, please contact the Microbiology Department within 48 hours to request sensitivity testing."

***Streptococcus anginosus* group on Non-Sterile Sites excluding Urines**
LIS Isolate Canned Message Code: **\Mill**; attached to Organism Class u.
"*Streptococcus anginosus* group are generally susceptible to penicillin, clindamycin, and levofloxacin. If susceptibility testing for this organism is required, please contact the microbiology laboratory within 48 hours."

***Streptococcus anginosus* group on Urines**
LIS Isolate Canned Message Code: **\MilU**; attached to Organism Class u.
"Streptococcus anginosus group are generally susceptible to penicillin and levofloxacin. If susceptibility testing for this organism is required, please contact the microbiology laboratory within 48 hours."

For *Enterococcoccus* reporting Tigercycline – messages link to Organism classes E, code **\TIGE**

"Tigecycline MIC is xx mg/L.
There are no CLSI standards for this drug. EUCAST suggests MICs <=0.25 mg/L correlate with susceptibility. Result for tigecycline is based on Etest gradient strips (bioMérieux) which have been validated with well-characterized laboratory (ATCC) strains. Verification on clinical isolates against a gold standard method has been limited. Please take this into consideration when interpreting these results. Please consult the microbiologist-on-call with any questions."

For **MSH VRE's**
LIS Isolate Canned Message Code: **\ICPR**, attached to organisms Trimethoprim/Sulfa **"THIS PATIENT MUST BE ON "CONTACT PRECAUTIONS" UNTIL FURTHER NOTICE."**

Vancomycin for ***E. gallinarum, and E. casseliflavus*** report as **R** with the statement
LIS Isolate Canned Message Code: **\EntV**; attached to organisms – entgal and entcas.
"This organism always has intrinsic non-transmissible resistance to vancomycin. The patient does not require isolation."

VRE isolated from VRE Screen Susceptibility Result Comment
LIS Isolate Canned Message Code: **\ICSN**; linked to organism Trimethoprim/Sulfa, Dlinezo=S or R. "Susceptibility results are provided for infection control purposes only."
For ***Listeria* species**:
LIS Isolate Canned Message Code: **\LIST**; attached to organism codes lismoc and lismon.
"In vitro susceptibility testing of this organism is not routinely performed. *Listeria* spp. Should be considered resistant to all cephalosporins. The recommended regimen for therapy is ampicillin. If additional advice on antimicrobial therapy is required, please contact the Medical Microbiologist."

***Enterococcus faecium* vanA gene positive but vancomycin susceptible**
LIS Isolate code: "entvaa" linked to Canned Message 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."

GPBs:

For ***Corynebacterium spp*., not *C. jeikeium* or *Bacillus* spp., not *B .anthracis* isolated from Blood Cultures**:
LIS Isolate Canned Message Code: **\cors**; linked to organism class – h
LIS Isolate Canned Message Code: **\bacs**; linked to organism class – j
"'*Corynebacterium* spp.' OR '*Bacillus* spp.' Are frequent blood culture contaminants. Clinical correlation is needed to determine the significance of this result. Susceptibility testing for this (these) organism(s) can be completed at a reference laboratory if requested."

For ***Propionibacterium* spp., Cutibacterium acnes and *Micrococcus* spp. Isolated from Blood Cultures:**
LIS Isolate Canned Message Code: **\pros**; linked to organism class – i
LIS Isolate Canned Message Code: **\mics**; linked to organism class – k
"'*Propionibacterium* spp.' Or 'Cutibacterium acnes' Or '*Micrococcus* spp.' Are frequent blood culture contaminants. Clinical correlation is needed to determine the significance of this result. Susceptibility testing for this(these) organism(s) is(are) unreliable. If advice on antimicrobial therapy is required, please contact the Medical Microbiologist."

For ***C difficile*** and ***C difficile* Detected**:
LIS Isolate Canned Message Code: **\Cdif** linked to organism **clodif** and **clodip**

GNBs:

For **SPICE**
LIS Isolate Canned Message Code: **&spc** attached to Organisms ceddav, cedlap, cedspp, prepen, provul, provp, Classes d, e, H, L and S.

"Resistance to extended-spectrum penicillins, beta-lactam/beta-lactamase inhibitor combinations (e.g. piperacillin-tazobactam), and cephalosporins may develop during therapy. These agents should be avoided and will be reported as resistant regardless of their in vitro susceptibility results. If you have questions, please contact the medical microbiologist on call."

For ***Aeromonas*** spp
LIS Isolate Canned Message Code: **\aero** attached to organism "f"
"Resistance to beta-lactam antimicrobials may develop in *Aeromonas* species during therapy. Choosing a non-beta-lactam antimicrobial and considering combination therapy is recommended for serious infections. Consultation with infectious diseases or medical microbiology is advised."

For **MSH *E. coli, Klebsiella* species, *Proteus*** Class A ESBL, Infection Control message:
Isolate canned message code **&taz** linked to organisms *E. coli*, Class J and Class T:
"MULTIPLY ANTIBIOTIC RESISTANT ORGANISM. THIS PATIENT MUST BE ON "CONTACT PRECAUTIONS" UNTIL FURTHER NOTICE FROM INFECTION CONTROL."

ESBL Comments
Attached to organisms esccol, Class J and Class T
Desbinh=Y Dfox=S **~&cla** "The susceptibility pattern suggests that this organism contains a class A extended spectrum beta-lactamase (ESBL)."
Dtaz=R Desbinh=N Dfox=R or I **~&claC** "The susceptibility pattern suggests that this organism contains a class C extended spectrum beta-lactamase (ESBL)."
Dtaz=R or I Desbinh=N Dfox=R or I Ddzone=Y**~&clIC** "The susceptibility pattern suggests that this organism contains an inducible class C extended spectrum beta-lactamase (ESBL)."
Desbinh=Y Dfox=R or I **~&clAC** "The susceptibility pattern suggests that this organism contains class A and C extended spectrum beta-lactamases (ESBL)."
Dtaz=R Desbinh=N Dfox=S **~&esbl** "The susceptibility pattern suggests that this organism contains an extended spectrum beta-lactamase (ESBL) other than class A or C."

ESBL or other Resistant Gram-Negative-Bacilli isolated from ESBL Screen, Resistant Pseudomonas Screen or Resistant Gram-Negative-Bacilli Screen – Susceptibility Result Comment
LIS Isolate Canned Message Code: **\ICSN**; linked to organism Class B, Dctr=R. "Susceptibility results are provided for infection control purposes only."

Positive *B*LACTA test result, link to organism Class 1 and drug blacta=Y **(\BLTA):**
"~Presumptive resistance to extended-spectrum penicillins,
~beta-lactam/beta-lactamase inhibitor combinations
~**(**e.g. piperacillin-tazobactam), and cephalosporins
~has been detected.
~Confirmation and further susceptibilities to follow. **"**

Previous Positive ESBL, LIS isolate comment code: \ESBP

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"."

For **MSH CRE's**
LIS Isolate Canned Message Code: **\ICPR**, attached to organism Class B **"THIS PATIENT MUST BE ON "CONTACT PRECAUTIONS" UNTIL FURTHER NOTICE."**

Previous Positive CRE, isolate comment code \CREP
"Phenotypic testing suggests this organism is carbapenemase POSITIVE as previously confirmed on "yyyy.mm.dd"."

For **Resistant Enterobacterales Colistin MIC Reporting**

MIC <=2 mg/L, LIS code **\CO<2**
"Colistin MIC = xx mg/L.
There are no CLSI standards for this drug. EUCAST suggests MICs <=2 mg/L correlate with susceptibility. Please consult the microbiologist-on-call with any questions."

MIC >2 mg/L, LIS code **\CO>2**
"Colistin MIC = xx mg/L.
There are no CLSI standards for this drug. EUCAST suggests MICs >2 mg/L correlate with resistance. Please consult the microbiologist-on-call with any questions."

For **Enterobacterales (other than *Proteus* spp. *Providencia* spp., *Morganella* spp.) and *S. maltophilia reporting* tigecycline** – pick from keypad:

For Susceptible results code **\TIGS**:
"Tigecycline MIC = mg/L
There are no CLSI standards for this drug. EUCAST suggests MICs <=1 mg/L correlate with susceptibility. Result for tigecycline is based on Etest gradient strips (bioMérieux) which have been validated with well-characterized laboratory (ATCC) strains. Verification on clinical isolates against a gold
standard method has been limited. Please take this into consideration when interpreting these results.
Please consult the microbiologist-on-call with any questions."

For Intermediate results code **\TIGI**:
"Tigecycline MIC = 2 mg/L

There are no CLSI standards for this drug. EUCAST suggests MICs =2 mg/L correlate with intermediate susceptibility. Result for tigecycline is based on Etest gradient strips (bioMérieux) which have been validated with well-characterized laboratory (ATCC) strains. Verification on clinical isolates against a gold
standard method has been limited. Please take this into consideration when interpreting these results. Please consult the microbiologist-on-call with any questions."

For Resistant results code **TIGR**:
"Tigecycline MIC = mg/L
There are no CLSI standards for this drug. EUCAST suggests MICs >2 mg/L correlate with resistance. Result for tigecycline is based on Etest gradient strips (bioMérieux) which have been validated with well-characterized laboratory (ATCC) strains. Verification on clinical isolates against a gold standard method has been limited. Please take this into consideration when interpreting these results. Please consult the microbiologist-on-call with any questions."

For *Morganella, Proteus, Providencia* Resistant results code **TIGN**:
"Tigecycline MIC = mg/L
There are no Clinical and Laboratory Standards Institute (CLSI) interpretive standards for this drug. For help with interpretation, please consult the microbiologist-on-call. (Ref: Pfizer Canada Inc. Product Monograph[Pr] Tygacil® Tigecycline for Injection. Kirkland, PQ: Pfizer Canada Inc., November 11, 2010)"

For **Enterobacterales carbapenemase reporting**

Preliminary Report based on ertapenem result, if ertapenem is I or R or =>1 mg/L code **MHT**
"~Screening tests suggest this organism may produce a carbapenemase. Further report to follow. If you have any questions, please contact the Medical Microbiologist on call."

For **Carbapenemase** Comments on ***Enterobacterales***:

Preliminary Report when Rosco disks and potentiation is available:

Mero & DPA (MRDP) >= 5 mm compared with Rosco meropenem (MRP10), code **MRDP**:
"Additional testing suggests this organism produces a metallo-beta-lactamase carbapenemase (e.g. NDM-1). Confirmation by PCR to follow."

Mero & Boronic Acid (MRBO) >=5 mm compared to Rosco meropenem (MRP10), code **MRBO**: "Additional testing suggests this organism produces a class A carbapenemase (e.g. KPC). Confirmation by PCR to follow."

Both Mero & DPA and Mero & Boronic Acid < 5 mm compared with Rosco meropenem (MRP10):

If **mero S**, code **MR-S**: "Additional testing indicates that this organism does NOT produce a carbapenemase."

Final CRE Reports:

Final Report – **NEGATIVE PCR** results from NML:

For **clinical specimens**, code **KPCN** – "The previous reported carbapenemase test for ....(isolate name)..was NOT confirmed. This organism is NEGATIVE by PCR for carbapenemase genes; as reported by the National Microbiology Laboratory 1015 Arlington St. Winnipeg, MB. Canada, R3E 3R2. If you have any questions, please contact the Medical Microbiologist on call."

For **Infection Control Screens**:
If the isolate is to be reported as ESBL, code **KPCN** – "The previous carbapenemase test for ....(isolate name)……..was NOT confirmed. This organism is NEGATIVE by PCR for carbapenemase genes; as reported by the National Microbiology Laboratory 1015 Arlington St. Winnipeg, MB. Canada, R3E 3R2. If you have any questions, please contact the Medical Microbiologist on call."

If the isolate is not generally reported (e.g. *Enterobacter* in ESBL screens),

- Change isolate to an alpha isolate.
- Report at the TEST Window with TEST COMMENT code **}KPCN** – " The previous carbapenemase test for ....(isolate name)……was NOT confirmed. This organism is NEGATIVE by PCR for carbapenemase genes; as reported by the National Microbiology Laboratory 1015 Arlington St. Winnipeg, MB. Canada, R3E 3R2. If you have any questions, please contact the Medical Microbiologist on call."

Final Report – **POSITIVE PCR** results from NML:
Report with ISOLATE COMMENT code **KPCP** – "This organism is POSITIVE for ___ carbapenemase (add specific carbapenemase that is confirmed) based on PCR; as reported by the National Microbiology Laboratory 1015 Arlington St. Winnipeg, MB. Canada, R3E 3R2. If you have any questions, please contact the Medical Microbiologist on call."

For ***Haemophilus* species** from Respiratory and Miscellaneous Sites – LIS Isolate Canned Message Code, attached to organism Class X:
If beta-lactamase is negative, **Bla-** "beta-lactamase negative result suggests susceptible to ampicillin."
If beta-lactamase is positive, **Bla+** "beta-lactamase positive result suggests resistance to ampicillin but generally susceptible to amoxicillin-clavulanic acid and cefuroxime."

For ***M. catarrhalis*** – LIS Isolate Canned Message Code: **mcat**; attached to Organism: "The majority of *Moraxella catarrhalis* are resistant to ampicillin. In vitro susceptibility testing for this

organism is not routinely performed and/or is unreliable. If advice on antimicrobial therapy is required, please contact the Medical Microbiologist".

For **Shigella spp** from Enteric sites with susceptibilities performed – LIS Isolate Canned Message Code **Shig:** "This isolate has a ciprofloxacin MIC of mg/L. There is the risk of ciprofloxacin treatment failures in infections caused by ciprofloxacin-susceptible Shigella with ciprofloxacin MICs between 0.125 and 1mg/L. Consultation with medical microbiology or infectious diseases is advised."

For ***S. maltophilia*** **reporting levofloxacin** – code **\sten:**
"NOTE: There are no CLSI interpretive standards for moxifloxacin and Stenotrophomonas maltophilia but levofloxacin and moxifloxacin minimum inhibitory concentrations (MICs) generally correlate well with each other. Ref: J Chemother. 2008 Feb;20(1):38-42."

For reporting **Cetolozane-Tazobactam** – code **\C/T**:
"Result for ceftolozane/tazobactam is based on Liofilmchem gradient strips (Alere) which have been validated with well-characterized laboratory (ATCC) strains. Verification with clinical isolates against a gold standard method has been limited. Please take this into consideration when interpreting these results."

B. Canned Messages to be selected from the Isolate Comment keypad:

The listed below are canned messages to be selected from the Isolate Comment keypad when needed.

1. At the LIS Isolate Comment Window, type the appropriate number on the keypad.
2. Press F12 to save.
3. Continue using F12 to save the order.
4. View the report.
5. Status the report as required.

For **Isoniazid** (**INH**) reporting if 0.1 mg/L=R and 0.4mg/L=S:
LIS Isolate Canned Message Code: **\INHr**; select from keypad
"This isolate has low-level resistance to isoniazid (INH). Patients infected with strains exhibiting this level of INH resistance may benefit from continuing therapy with INH. Consultation with a specialist experienced in the treatment of tuberculosis is recommended."

BORSA (PBP2a/*mec*A-negative *S. aureus* with oxacillin MIC>=4mg/L)
LIS Isolate Canned Message Code: **\BORS**; select from Isolate Keypad
This organism is a borderline-oxacillin resistant Staphylococcus aureus (BORSA) which is resistant to cloxacillin and cefazolin by a mechanism different from that in typical MRSA. Consultation with a Microbiologist or Infectious Disease physician is advised."

If susceptibility is done on request for ***ß-haemolytic Streptococcus*** **Groups A, B, C & G**

Do not remove original canned message. Add message from Isolate Comment keypad code "Susceptibility\done" – "Susceptibility completed as requested."

Enterococcus from Blood and Sterile Sites:
If high level gentamicin is **susceptible** (regardless of streptomycin result), select from Isolate Comment Keypad **\EGMS**: "Serious enterococcal infections may require an aminoglycoside for synergy. Please contact the Medical Microbiologist for treatment advice".
If high level gentamicin is **resistant** (regardless of streptomycin result), select from Isolate Comment Keypad **\EGMR**: "This organism is high level aminoglycoside resistant. Please contact the Medical Microbiologist for treatment advice".

Positive *B*LACTA test result and
If ESBL is confirmed, report with isolate comment (\ESBC):
"Resistance to extended-spectrum penicillins, beta-lactam, beta-lactamase inhibitor combinations (e.g. piperacillin-tazobactam), and cephalosporins has been confirmed."

OR

If ESBL is NOT confirmed e.g. in *K. oxytoca*, report with isolate comment (\ESBN):
"The previously reported presumptive resistance to extended-spectrum penicillins, beta-lactam, beta-lactamase inhibitor combinations (e.g. piperacillin-tazobactam), and cephalosporins was NOT confirmed."

Comment Template for Infrequently tested Antibiotics

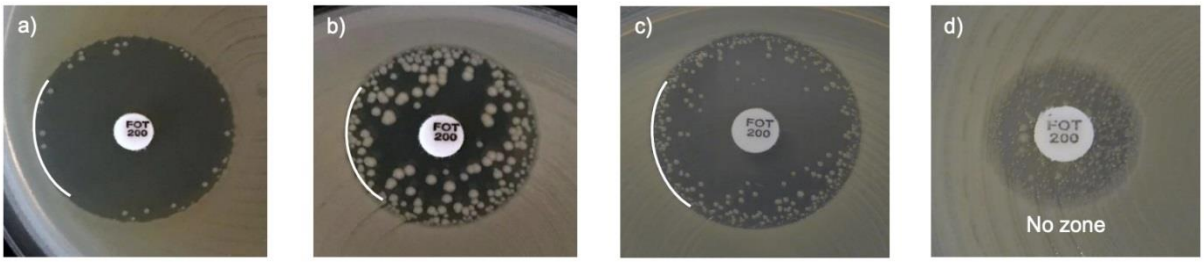
- For reporting of susceptibility results of infrequently tested antibiotics.
- Setup and read KB zone size/Etest strip.
- Select the appropriate option to append based on the test result.

FOSFOMYCIN

Non-urinary E coli:
Fosfomycin zone of inhibition is __ mm. There are no CLSI breakpoints for this specimen type.
Add **one** of the following options:
a) CLSI recommends zones of inhibition ≥ 16mm correlate with susceptibility for urinary E coli.
b) CLSI recommends zones of inhibition 13-15mm correlate with intermediate susceptibility for urinary E coli.
c) CLSI recommends zones of inhibition ≤ 12mm correlate with resistance for urinary E coli.

***AND** add one of the following options:*
a) EUCAST recommends zones of inhibition ≥ 24 mm correlate with susceptibility for E. coli from all specimen types.
b) EUCAST recommends zones of inhibition < 24 mm correlate with resistance for E. coli from all specimen types.

NOTE: FOR EUCAST INTERPRETATIONS testing of **KB fosfomycin on E. coli** isolates, IGNORE GROWTH IN INNER ZONE WHERE THERE IS STILL DISTINCT OUTER ZONE:



Examples of inhibition zones for *Escherichia coli* with fosfomycin.
a-c) Ignore all colonies and read the outer zone edge.
d) Record as no inhibition zone.

Enterobacterales and other Gram-negatives upon microbiologist approval:
Fosfomycin zone of inhibition is __ mm. There are no CLSI breakpoints for this drug-organism combination.
Add **one** of the following options:
a) CLSI recommends zones of inhibition ≥ 16mm correlate with susceptibility for urinary E coli.
b) CLSI recommends zones of inhibition 13-15mm correlate with intermediate susceptibility for urinary E coli.
c) CLSI recommends zones of inhibition ≤ 12mm correlate with resistance for urinary E coli.
***AND** add one of the following options:*
a) EUCAST recommends zones of inhibition ≥ 24 mm correlate with susceptibility for E. coli from all specimen types.
b) EUCAST recommends zones of inhibition < 24 mm correlate with resistance for E. coli from all specimen types.

All Enterobacterales:
AGAR DILUTION
Fosfomycin MIC = _____ mg/L. There are no CLSI breakpoints for this drug-organism combination.
Add **one** of the following options:

a. EUCAST recommends MIC ≤ 32 mg/L correlate with susceptibility for Enterobacterales from all specimen types.
b. EUCAST recommends MIC > 32 mg/L correlate with resistance for Enterobacterales from all specimen types.

Non-Enterobacterales:
AGAR DILUTION
Fosfomycin MIC is _____ mg/L. There are no CLSI or EUCAST standards for this drug-organism combination.
Add **one** of the following options:

a. EUCAST recommends MIC ≤ 32 mg/L correlate with susceptibility for Enterobacterales from all specimen types.
b. EUCAST recommends MIC > 32 mg/L correlate with resistance for Enterobacterales from all specimen types.

CEFIDEROCOL

Enterobacterales:
Cefiderocol zone of inhibition is ___mm.
Add **one** of the following options:

a) CLSI recommends zone size of ≥16 mm correlates with susceptibility. Accuracy and reproducibility of cefiderocol testing is low. False-resistant and false-susceptible results may occur, especially when results are close to the breakpoint.
b) CLSI recommends zone size of 9-15 mm correlates with intermediate susceptibility. Accuracy and reproducibility of cefiderocol testing is low. False-resistant and false-susceptible results may occur, especially when results are close to the breakpoint.
c) CLSI recommends zone size of ≤8 mm correlates with resistance Accuracy and reproducibility of cefiderocol testing is low. False-resistant and false-susceptible results may occur, especially when results are close to the breakpoint.

Pseudomonas aeruginosa:
Cefiderocol zone of inhibition is ___mm.
Add **one** of the following options:

a) CLSI recommends zone size of ≥18mm correlates with susceptibility. Accuracy and reproducibility of cefiderocol testing is low. False-resistant and false-susceptible results may occur, especially when results are close to the breakpoint.
b) CLSI recommends zone size of 13-17mm correlates with intermediate susceptibility Accuracy and reproducibility of cefiderocol testing is low. False-resistant and false-susceptible results may occur, especially when results are close to the breakpoint.
c) CLSI recommends zone size of ≤12mm correlates with resistance Accuracy and reproducibility of cefiderocol testing is low. False-resistant and false-susceptible results may occur, especially when results are close to the breakpoint.

Stenotrophomonas maltophilia:
Cefiderocol zone of inhibition is ___mm.
Add **one** of the following options:

a) CLSI recommends zone size of ≥15 mm correlates with susceptibility. Accuracy and reproducibility of cefiderocol testing is low. False-resistant and false-susceptible results may occur, especially when results are close to the breakpoint.

Acinetobacter baumanii only :
Cefiderocol zone of inhibition is ___mm.
Add **one** of the following options:

a) CLSI recommends zone size of ≥15 mm correlates with susceptibility. Accuracy and reproducibility of cefiderocol testing is low. False-resistant and false-susceptible results may occur, especially when results are close to the breakpoint.

Enterobacteriales/Pseudomonas aeruginosa/Stenotrophomonas maltophilia/Acinetobacter:
Cefiderocol MIC is ___ mg/L.
Add **one** of the following options:

a) CLSI recommends MIC of ≤4 mg/L correlates with susceptibility. CLSI breakpoints are based on dosage of 2g iv q8h infused over 3h and are considered under investigational status.
b) CLSI recommends MIC of 8 mg/L correlates with intermediate susceptibility. CLSI breakpoints are based on dosage of 2g iv q8h infused over 3h and are considered under investigational status.
c) CLSI recommends MIC of ≥16 mg/L correlates with resistance. CLSI breakpoints are based on dosage of 2g iv q8h infused over 3h and are considered under investigational status.

Non-fermenters and Burkholderia:
There are no CLSI or EUCAST breakpoints for this drug-organism combination.
Cefiderocol MIC is ___ mg/L.
Add **one** of the following options:

a) CLSI recommends MIC of ≤4 mg/L correlate with susceptibility for Enterobacterales, Pseudomonas aeruginosa, Stenotrophomonas maltophilia, and Acinetobacter. CLSI breakpoints are based on dosage of 2g iv q8h infused over 3h and are considered under investigational status.
b) Cefidercol MIC is 8 mg/L. CLSI recommends MIC of 8 mg/L correlate with intermediate susceptibility for Enterobacterales, Pseudomonas aeruginosa, Stenotrophomonas maltophilia, and Acinetobacter. CLSI breakpoints are based on dosage of 2g iv q8h infused over 3h and are considered under investigational status.
c) Cefiderocol MIC is __mg/L. CLSI recommends MIC of ≥16 mg/L correlates with resistance for Enterobacterales, Pseudomonas aeruginosa, Stenotrophomonas maltophilia, and Acinetobacter. CLSI breakpoints are based on dosage of 2g iv q8h infused over 3h and are considered under investigational status.

PLAZOMICIN

Enterobacterales, Pseudomonas aeruginosa/Acinetobacter/Stenotrophomonas

maltophilia/Burkholderia cepacia/Non-fermenters
Plazomicin MIC is __ mg/L. There are no CLSI or EUCAST breakpoints for this drug-organism combination.
Add **one** of the following options:
a) The FDA-identified interpretable criteria suggests MIC of ≤2 mg/L correlate with susceptibility for Enterobacterales.
b) The FDA-identified interpretable criteria suggests MIC of 4 mg/L correlate with susceptibility for Enterobacterales.
c) The FDA-identified interpretable criteria suggests MIC of ≥8 mg/L correlate with susceptibility for Enterobacterales.

CEFTAZIDIME-AVIBACTAM

BUILD INTO LIS:
Enterobacterales and Pseudomonas
CLSI recommends MIC of ≤8 mg/L correlates with susceptibility
CLSI recommends MIC of ≥16 mg/L correlates with resistance

Stenotrophomonas maltophilia/Acinetobacter/Burkholderia cepacia:
There are no CLSI breakpoints for this drug-organism combination.
Ceftazidime-avibactam MIC is _____ mg/L.
Add **one** of the following options:
a) CLSI suggests an MIC of ≤8 mg/L correlate with susceptibility for Enterobacterales and Pseudomonas aeruginosa.
b) CLSI suggests MIC ≥16 mg/L correlate with resistance for Enterobacterales and Pseudomonas aeruginosa.

MEROPENEM-VABORBACTAM

BUILD INTO LIS:
Enterobacterales
CLSI recommends MIC of ≤4 mg/L correlates with susceptibility.
CLSI recommends MIC of 8 mg/L correlates with intermediate susceptibility.
CLSI recommends MIC of ≥16 mg/L correlates with resistance.

Pseudomonas
Meropenem-vaborbactam MIC is __ mg/L. There are no CLSI breakpoints for this drug-organism combination.
Add **one** of the following options:
a) EUCAST suggests MIC of ≤8 mg/L correlate with susceptibility.
b) EUCAST suggests MIC of ≥16 mg/L correlate with resistance.

Stenotrophomonas maltophilia/Acinetobacter/Burkholderia cepacia/other Gram-negatives
Meropenem-vaborbactam MIC is __ mg/L. There are no CLSI breakpoints for this drug-organism combination.
Add **one** of the following options:
a) CLSI recommends MIC of ≤4 mg/L correlates with susceptibility for Enterobacterales
b) CLSI recommends MIC of 8 mg/L correlates with intermediate susceptibility for Enterobacterales
c) CLSI recommends MIC of ≥16 mg/L correlates with resistance for Enterobacterales.

IMIPENEM-RELEBACTAM

BUILD INTO LIS:
Enterobacterales, Pseudomonas aeruginosa

Acinetobacter
Imipenem-relebactam MIC is __ mg/L. There are no CLSI breakpoints for this drug-organism combination.
Add **one** of the following options:
a) EUCAST suggests MIC of ≤2 mg/L correlate with susceptibility for Acinetobacter.
b) EUCAST suggests MIC of ≥4 mg/L correlate with resistance for Acinetobacter.

Stenotrophomonas maltophilia/Burkholderia cepacia/Non-fermenters
Imipenem-relebactam MIC is __ mg/L. There are no CLSI breakpoints for this drug-organism combination.
Add **one** of the following options:
a) CLSI suggests MIC of ≤2 mg/L correlate with susceptibility for Pseudomonas aeruginosa.
b) CLSI suggests MIC of ≥4 mg/L correlate with resistance for Pseudomonas aeruginosa.
c) CLSI suggests MIC of ≥8 mg/L correlate with resistance for Pseudomonas aeruginosa.

APPENDIX A. VERIFICATION OF UNUSUAL RESULTS

Verification of Antimicrobial Susceptibility Test Results and Confirmation of Organism Identification

I. Introduction

This section describes the occasions where the drugs tested against isolates showed phenotype that:
1. have never been documented
2. are uncommon, and/or
3. represent results that could easily occur from technical errors which may have significant clinical consequences.

II. Reagents/Materials/Media

Analytical Process - Bacteriology Reagents_Materials_Media List QPCMI10001

III. Procedure

When any of the listed results in the TABLE 1 below occurs, verify the result as follows:
1. Check purity plate.
2. Check previous reports on the patient.
3. Confirm the identification of the isolate from the original isolation medium.
4. Repeat susceptibility test to confirm result. Use an alternative method if applicable.
5. For isolates that show results other than susceptible for those antimicrobial agents for which only susceptible interpretive criteria are provided by CLSI guidelines M100-S23 (listed as “not S” above) and for staphylococci with vancomycin – I or R results:
   i. Confirm the organism identification
   ii. Confirm the antimicrobial susceptibility test results
   iii. Freeze the isolate
   iv. Send the isolate to PHL for confirmation.
6. If the result is confirmed, notify the Charge Technologist.
7. The Charge Technologist confirms the result and notifies the Microbiologist.
8. The Microbiologist further confirms the result and notifies the Infection Control Practitioner.

For results marked with *****, LIS reflex rules will automatically report these as R; repeat susceptibility testing is not required if the purity and organism identification is confirmed.

Organism or Group	**Uncommon results**
Any organism	Resistant to all agents routinely tested
Gram-negative organisms	
Any gram-negative organisms	Piperacillin – S **and** Piperacillin/tazobactam – R
Enterobacterales	Imipenem, Meropenem – I or R Carbapenem – I or R Amikacin, gentamicin, and tobramycin – R
Enterobacterales	Imipenem, Meropenem – I or R **and** Ertapenem = S
Citrobacter freundii *Enterobacter* species *Serratia marcescens*	Ampicillin, Cefazolin – S*****
Klebsiella species *Proteus vulgaris* *Providencia* species	Ampicillin – S*****
Escherichia coli, *Klebsiella* species, *Proteus* species	Cefpodoxime – Vitek=I or R; KB=S
Escherichia coli, *Klebsiella* species, *Proteus mirabilis*	Extended-spectrum cephalosporin (III or IV) – I or R
Escherichia coli, *Klebsiella* species, *Proteus* species	KB-ESBL panel with reduction in zone of inhibition instead of potentiation or no change.
Salmonella and *Shigella* spp.	Cephalosporin III – I or R Fluoroquinolone – I or R
Pseudomonas aeruginosa	Colistin/polymyxin – I or R Amikacin, and tobramycin – R Carbapenem – I or R
Acinetobacter baumannii	Colistin/polymyxin – R Carbapenem – I or R
Stenotrophomonas maltophilia	Imipenem, Meropenem – S Trimethoprim-sulfamethoxazole – I or R
Haemophilus influenzae	Amoxicillin-clavulanate – R Ampicillin – R and β-lactamase negative Aztreonam – not S Imipenem, Meropenem – not S 3rd generation cephalosporin – not S Extended-spectrum cephalosporin (III or IV) – not S Ceftaroline – not S Carbapenem – not S

Organism or Group	**Uncommon results**
	Fluoroqinolone – not S
Neisseria gonorrhoeae	3rd generation cephalosporin – R Extended-spectrum cephalosporin (III or IV) – not S Fluoroquinolone – I or R
Neisseria meningitides	Ampicillin or Penicillin – R Extended-spectrum cephalosporin (III or IV) – not S Meropenem – not S Ampicillin or Penicillin – I Azithromycin – not S Chloramphenicol – I or R Fluoroquinolone – I or R Minocycline – not S Rifampin – I or R
Gram-positive organisms	
Enterococcus spp.	Daptomycin – not S Linezolid – R Vancomycin – R High-level aminoglycoside – R
Enterococcus faecalis	Ampicillin or Penicillin – R Daptomycin – not S Quinupristin-Dalfopristin – S Linezolid – I or R
Enterococcus faecium	Ampicillin – S Daptomycin – not S Linezolid – I or R
Staphylococcus aureus	Daptomycin – not S Linezolid – R Quinupristin-Dalfopristin – I or R Oxacillin – R Vancomycin – I or R Vancomycin MIC = 4 ug/mL Vancomycin MIC ≥8 ug/mL Clindamycin=R and Erythromycin=S Ceftaroline – R
Coagulase-negative *Staphylococcus*	Daptomycin – not S Linezolid – R

Organism or Group	**Uncommon results**
	Vancomycin – I or R Clindamycin=R and Erythromycin=S Quinupristin-Dalfopristin – I or R

Organism or Group	**Uncommon results**
Gram-positive organisms (cont'd)	
Streptococcus pneumoniae	3rd generation cephalosporin – R Fluoroquinolone – I or R Linezolid – not S Vancomycin – not S Clindamycin=R and Erythromycin=S Ceftaroline – not S Imipenem or meropenem – I or R Quinupristin-dalfopristin – I or R Rifampin – I or R <u>Using nonmeningitis breakpoints:</u> Amoxicillin or penicillin – R Extended-spectrum cephalosporin (III or IV) – R Oxacillin=S & Penicillin etest R
beta-haemolytic *Streptococcus*	Ampicillin or Penicillin – not S 3rd generation cephalosporin – not S Daptomycin – not S Linezolid – not S Vancomycin – not S Clindamycin=R and Erythromycin=S Quinupristin-dalfopristin – I or R Ceftaroline – not S Ertapenem or meropenem – not S Extended-spectrum cephalosporin (III or IV) – not S
viridans *Streptococcus* (including anginosus group)	Daptomycin – not S Ertapenem or meropenem – not S Linezolid – not S Quinupristin-dalfopristin – I or R Vancomycin – not S Clindamycin=R and Erythromycin=S

IV. Reference

Suggestions for Verification of Antimicrobial susceptibility Test Results and Confirmation of Organism identification in Table 8 of Clinical and Laboratory Standards Institute (CLSI) Document – Performance Standards for Antimicrobial Susceptibility Testing M100-S25 appendix A.

APPENDIX B. AGENTS NEVER TO BE REPORTED BY SITE

The antimicrobial agents listed in the table below should never be used for any isolate reported from the specified site.

URINE SPECIMENS	CSF/VP SHUNT & BRAIN SPECIMENS	BAL/ETT/sputum/ lung biopsy	PEDEATRIC PATIENTS	PREGNANCY

• Clindamycin
• Macrolides:
  • Erythromycin
  • Clarithromycin
  • Azithromycin
• Minocycline
• Tigecycline
• Chloramphenicol
• Moxifloxacin
	• Agents given orally only
	• 1st & 2nd generation cephalosporins
	• Clindamycin
	• Macrolides
	• Tetracyclines
	  - Doxycycline
	  - Minocycline
	  - Tigecycline
	• Fluoroquinolones
	  - Ciprofloxacin
	  - Levofloxacin
	  - Moxifloxacin
	• Aminoglycosides
	  - Gentamicin
	  - Tobramycin
	  - Amikacin
	• Doripenem, Ertapenem, Imipenem
	• Lefamulin
		Daptomycin	• Ceftriaxone (< 1 month old)
			• Chloramphenicol (< 1 month old)
			• Nitrofurantoin (< 1 month old)
			• TMP-SMX (< 2 months old)
			• Tetracyclines(< 8 year old)
			• Fosfomycm (< 12 yearsold)
			• Daptomycin (< 18 years old)
			• Fluoroquinolones• (< 18 years old)
			• Linezolid (< 18 yearsold)
				• Clarithromycin
				• Nitrofurantoin

References:

Clinical and Laboratory Standards Institute (CLSI) Document – Performance Standards for Antimicrobial Disk Susceptibility Testing M2-Disk Diffusion M100-S27, 2017.

Institute for Quality Management in Healthcare (IQMH) Consensus Practice Recommedations – Antimicrobial Susceptibility Testing and Reporting on Bacteriology Specimens. Revision 2016.07.06.

APPENDIX C. DETECTION OF ANTIMICROBIAL RESISTANT ORGANISMS

Antibiotic Resistant Organisms Detection

A. How to Detect MRSA/BORSA:

Routine Bench

Screening tests:

- Oxacillin screen positive (and/or)
- Vitek cefoxitin screen positive (and/or)
- Vitek oxacillin MIC =>4 mg/L

Confirmatory testing (to be done sequentially if any of the screening tests are positive)

- PBP2a
- Induced PBP2a with cefoxitin KB (if PBP2a negative)
- If both negative, Send to PHL for PCR (*mecA* and *mecC*) & Oxacillin MIC

Previously positive tests:

Report as MRSA based on:

- <3days: Oxacillin screen only
- 3days-3months: Oxacillin screen with vitek susceptibilities (no PBP2a)
- >3months: Oxacillin screen with vitek susceptibilities plus PBP2a
  (since last MRSA positive on routine or screening bench)

Reporting:

- If PBP2a test positive, then **finalize** as methicillin-resistant *Staphylococcus aureus* (MRSA).

- If PBP2a induced test negative, while waiting for mecA and mecC PCR / Oxacillin MIC:

  - Send **prelim** report of *Staphylococcus aureus* with susceptibilities. Supress beta-lactams with the following comment added in the isolate comment:
    "Screening tests suggest this isolate may be resistant to cloxacillin and cefazolin. Confirmation to follow. If you have any questions, please contact the microbiologist-on-call."

When mecA and mecC PCR / Oxacillin MIC results are available:

- If all confirmatory tests are negative but oxacillin <4 mg/L, then finalize as oxacillin susceptible *Staphylococcus aureus.*

- If *mecA or mecC* are positive then finalize as methicillin-resistant *Staphylococcus aureus* (MRSA).

- If all confirmatory tests are negative but oxacillin =>4mg/L, then report *Staphylococcus aureus* with the following BORSA comment: *"This organism is a borderline-oxacillin resistant Staphylococcus aureus (BORSA) which is resistant to cloxacillin and cefazolin by a mechanism different from that in typical MRSA. Consultation with a Microbiologist or Infectious Disease physician is advised."*

Screening Bench

Screen:

- Denim blue colonies

Initial Confirmatory test:

- PBP2a – if positive, then prelim as methicillin-resistant Staphylococcus aureus (MRSA) and continue with workup.

Additional Confirmatory testing if PBP2a negative:

- Induced PBP2a with cefoxitin KB
- Oxacillin screen positive
- Vitek cefoxitin screen positive
- Vitek oxacillin MIC =>4 mg/L

Previously positive tests:

- <3months: Report as MRSA based on denim blue colonies on screen plate and MALDI confirms S. aureus ID.
  ≥3months: Full work-up as above (since last MRSA positive on routine or screening bench)

Reporting:

- If all additional confirmatory tests are positive, then **finalize** as methicillin-resistant Staphylococcus aureus (MRSA).

- When conflicting results arise, please consult senior/charge technologist for further advice.

Results should be held back (no isolate reported) but calls made to infection control as per senior/charge technologist's advice

How to Detect VISA/hVISA/VRSA:

Routine Bench

Screen:

- Vanco screen plate positive
  or
- VITEK Vanco MIC > 1 mg/L

Confirmatory testing:

- Vanco Etest to confirm MIC
- Macro Etest to detect hVISA

Previously positive tests:

Report as VISA/hVISA/VRSA based on:

- <3days: Vancomycin screen only
- 3days-3months: Vancomycin screen with vitek susceptibilities
- >3months: Vancomycin screen with vitek susceptibilities and Vanco Etest & Macro Etest.
  (since last VISA/ hVISA/VRSA positive on routine bench or screening bench culture)

Report:

If VITEK vanco MIC =2 mg/L:

- If vanco Etest (rounded up to 2 fold dilution vanco) is <2 mg/L and macro Etest is negative, then report isolate as MSSA or MRSA.
- If vanco Etest = 2mg/L and macro Etest is negative, then report as MSSA or MRSA with MIC and with the following comment: "This isolate has a vancomycin MIC of 2 mg/L which is associated with an increased risk of vancomycin treatment failrues. Consultation with infectious diseases or medical microbiology is advised."
- If vanco Etest <4mg/L and macro Etest is POSITIVE, then report as MSSA or MRSA with the hVISA comment as follows: "Presumptive vancomycin hetero-intermediate S. aureus (hVISA). Confirmation to follow".

If VITEK vanco &/or Etest MIC 4-8 mg/L, regardless of the macro Etest result:

- Then report as MSSA or MRSA with the VISA comment as follows: "Presumptive vancomycin-intermediate S. aureus (VISA). Confirmation to follow".

If VITEK vanco &/or Etest MIC >8 mg/L, regardless of the macro Etest result:

- Then report as MSSA or MRSA with the VRSA comment as follows: "Presumptive vancomycin-resistant S. aureus (VRSA). Confirmation to follow."

B. How to detect VRE:

Routine Bench

Screen:

- Vancomycin screen pos

Confirmatory testing:

- Vancomycin and teicoplanin macro Etests (all benches)
- Cepheid PCR (all benches, including vancomycin susceptible *E. faecium* from blood culture)

Previously positive tests:

Report as VRE based on:

- <3days: Vancomycin screen
- 3days-3 months: Vancomycin screen plus Etests (no Cepheid PCR)
- >3 months: Vancomycin screen, Etests plus Cepheid PCR (since last full VRE workup)

Report:

- If macro Etests are positive, then VRE
- If Cepheid is positive, then VRE or VanS VRE
- Otherwise, no VRE

Screening Bench

Screen:

- Purple/blue colonies on Brilliance agar

Confirmatory testing:

- Cepheid PCR
- Vancomycin and teicoplanin macro Etests
- Vancomycin screen

Previously positive tests:

Report as VRE based on:

- <3 months: ID and Vancomycin screen
- ≥3 months: ID, Vancomycin screen, Etests plus Cepheid PCR (since last full VRE workup)

Report:

- If macro Etests are positive, then VRE
- If Cepheid is positive, then VRE or VanS VRE
- Otherwise, no VRE

C. How to detect ESBL:

Routine Bench

Screen:
Cefpodoxime (Vitek) I/R (MIC > 1 mg/L) – for *E coli, Klebsiella pneumonia, K. oxytoca* and *Proteus mirabilis*

Confirmatory testing:
ESBL Double disk

1. Routine Figure 1. KB-ESBL Template

Previously positive tests:
Report as ESBL based on:

- <3days: Reported ID with referral to previous isolate
- >3days: Reported ID with vitek and ESBL Double disk
  (since last full ESBL workup)

Report:
Report with susceptibilities based on vitek
Confirm with ESBL double disk and record result on back of card only – issue a corrected report if discrepancy found

Screening Bench

Screen:
MCPOD plate – oxidase negative LF / NLF are considered screen positive

Confirmatory testing:
ESBL Double disk

A. Infection Control Figure 2. Infection Control KB-ESBL Template
   (only for Mother/Infant ward and special requests)

Previously positive tests:
Report as ESBL based on:

- <3months: growth on McPOD, ID with referral to previous isolate
- ≥3months: growth on McPOD, ID with ESBL Double disk
  (since last full ESBL workup)

Report:
Report positive ESBL Double disks to Mother/Infant wards only

NOTE: An isolate with cefpodoxime S and ceftriaxone or ceftazidime I/R is an UNUSUAL RESULT. Check for purity and redo the susceptibility.

D. How to detect CPO:

Routine Bench

Screen:

- Erta=I/R and Mero mic ≤0.25
- Mero=I/R or Mero=S mic ≥0.5
- KB Mero Screen Test = R

Confirmatory testing:

- βCARBA
- CARB-R Cepheid PCR

Additional Confirmatory testing:

- ROSCO with Temocillin (if βCARBA= negative **OR** βCARBA = positive & CARB-R Cepheid PCR neg)
- PCR – send to NML (if CARB-R Cepheid PCR neg OR ROSCO with Temocillin=R/potentiation)

Notify as per Isolate Notification and Freezing Table QPCMI15003

Previously positive tests:

Report as CRE based on:

- <3days: ID with meropenem screen results
- 3days-6 months: ID with Vitek, βCARBA
- >6 months: ID with Vitek , βCARBA, CARB-R Cepheid PCR, ROSCO with Temocillin (if βCARBA =neg) NML PCR(if CARB-R Cepheid PCR neg OR ROSCO with Temocillin=R/potentiation)
  (since last full CPO workup)

Report:

- See Carbapenemase Testing Reporting

Screening Bench

Screen:

- KB MERO Screen=R

Confirmatory testing:

- βCARBA
- CARB-R Cepheid PCR

Additional Confirmatory testing:

- ROSCO with Temocillin (if βCARBA= negative **OR** βCARBA = positive & CARB-R Cepheid PCR neg)
- PCR – send to NML (if CARB-R Cepheid PCR neg **OR** if Rosco with Temocillin=R/potentiation)

- Notify as per Isolate Notification and Freezing Table QPCMI15003

Previously positive tests:

Report as CPO based on:

- <6 months: βCARBA test
- ≥6 months: βCARBA, CARBR Cepheid PCR, (ROSCO with Temocillin (if βCARBA =neg) PCR NML (if CARB-R Cepheid PCR =neg or ROSCO with Temocillin=R/potentiation) (since last full CPO workup)

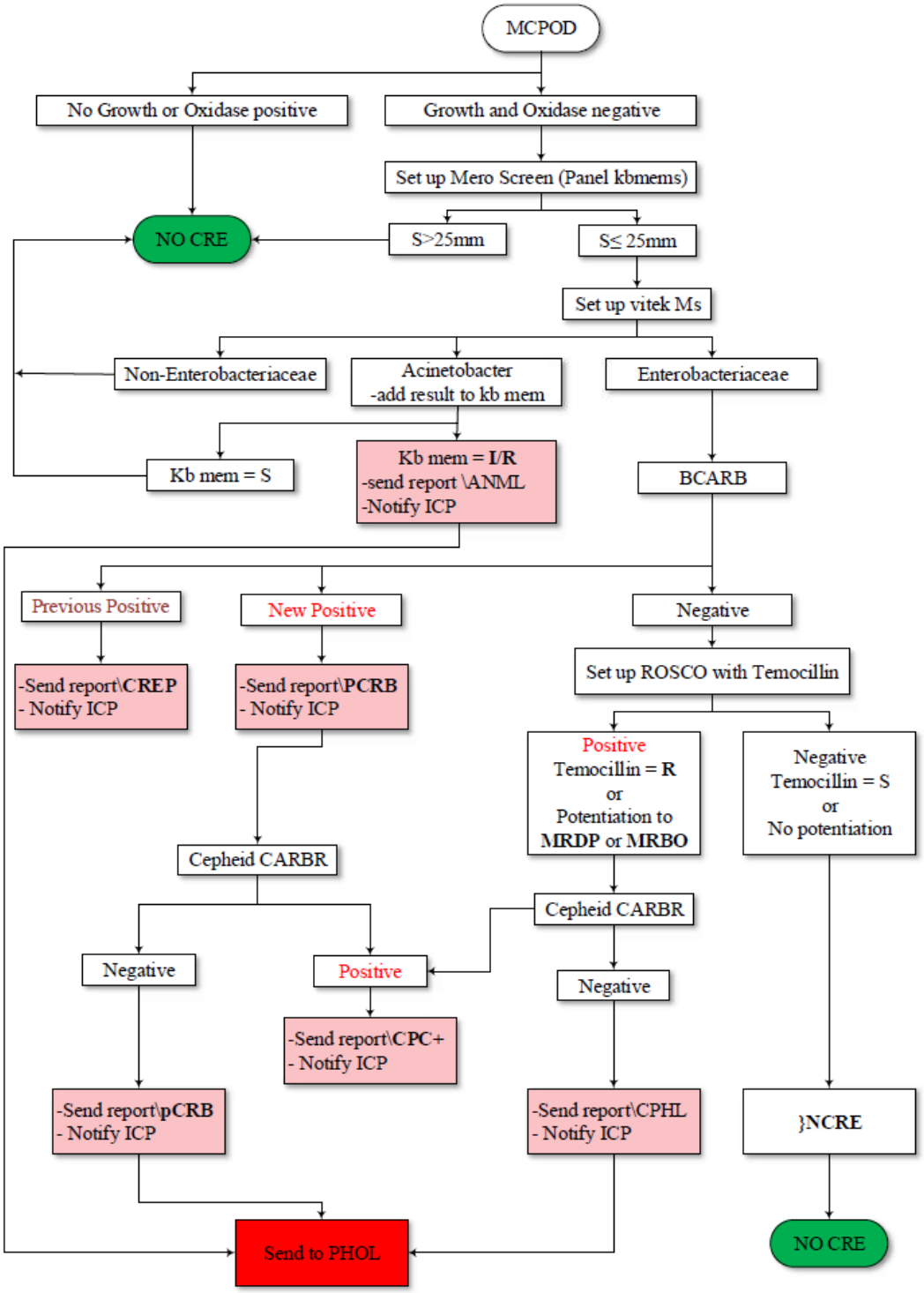
Report:

- See Carbapenemase Testing Reporting

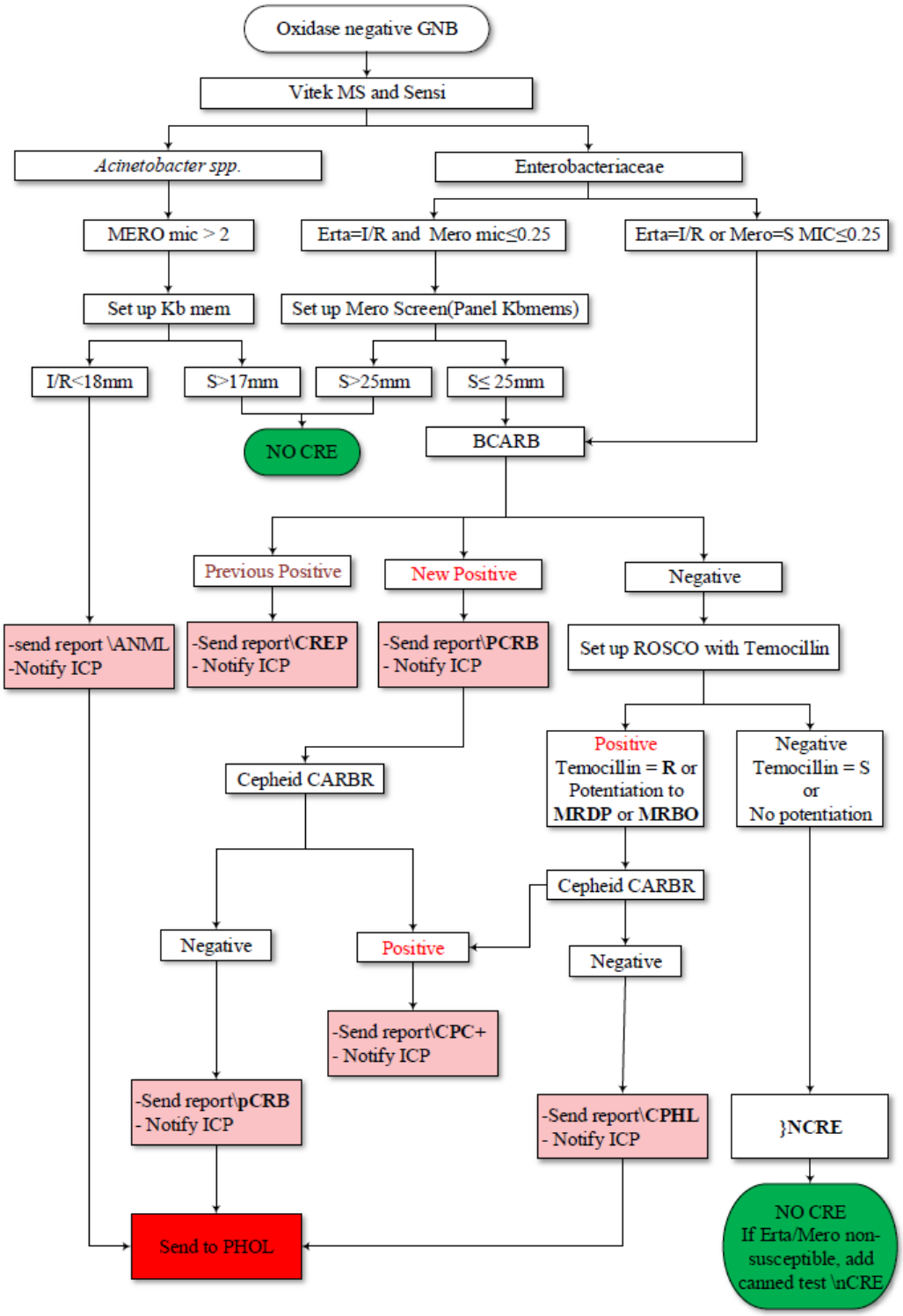
NOTE: An isolate with erta S and mero I/R is an UNUSUAL RESULT. Check for purity and repeat the susceptibility.

CARBAPENEMASE TESTING FLOWCHART

Infection Control CRE Screen Flowchart



Identification of Carbapenemase Producing isolates from Clinical Samples Flowchart



Carbapenemase Testing Reporting

Direct Specimen PCR Reporting

	LIS Code	Test Comment
Negative Cepheid CARBA-R	}CAR-	"Negative – No carbapenemase genes detected by Cepheid Xpert CARBA-R Assay (for research use only). This assay is able to detect NDM, KPC, OXA48, OXA181, OXA232, OXA244, IMP-1, and VIM carbapenemase genes."
Positive Cepheid CARBA-R	}CAR+	"_____ gene DETECTED by Cepheid Xpert CARBA-R Assay (for research use only). This assay is able to detect NDM, KPC, OXA48, OXA181, OXA232, OXA244, IMP-1, and VIM carbapenemase genes."

For IC Screen & Clinical Culture Reporting – *Acinetobacter spp*

	Test Comment	Isolate Comment	Report Status	Notification to ICP/Ward	Other Instructions
KB mem I/R	For IC Screen: \ANML	For Clinical Cultures: \ANML	Prelim	Yes	
Negative NML Report	For IC Screen: "UPATED REPORT" }NCRE	For Clinical Cultures: \ACCN	Final	Yes	For IC Screen: Suppress previously reported Isolate
Positive NML Report	For IC Screen: "UPDATED REPORT" "POSITIVE Carbapenemase Screen"	For IC & Clinical Cultures: \ACCP	Final	Yes	

For IC Screen & Clinical Culture Reporting

	Test Comment	Isolate Comment	Report Status	Notification to ICP/Ward	Other Instructions
Negative βCARBA					
Negative βCARBA/ Negative ROSCO	For IC Screen: "UPDATED REPORT" }NCRE	For Clinical Cultures: report susceptibility results as per susceptibility;	Final	Yes	

	Test Comment	**Isolate Comment**	**Report Status**	**Notification to ICP/Ward**	**Other Instructions**
		with comment \nCRE			
Negative βCARBA/ Positive ROSCO	For IC Screen: "UPDATED REPORT" "POSITIVE Carbapenemase Screen"	For Clinical Cultures: \CNML	Final	Yes	
Positive βCARBA					
Previous positives ≤6 months	For IC Screen: "POSITIVE Carbapenemase Screen."	For IC & Clinical Cultures: \CREP	Final	Yes	
New positive	For IC Screen: "POSITIVE Carbapenemase Screen"	For IC & Clinical Cultures: \PCRB	Prelim	Yes	
Positive βCARBA and Negative Cepheid CARBA-R PCR (CARBR)	For IC Screen: "UPDATED REPORT" "POSITIVE Carbapenemase Screen" For Clinical Cultures: "UPDATED REPORT"	For IC & Clinical Cultures: \pCRB	Prelim	Yes	Remove the original Isolate comment and replace with new Isolate comment.
Positive βCARBA and Positive Cepheid CARBA-R PCR (CARBR)	For IC Screen: "UPDATED REPORT" "POSITIVE Carbapenemase Screen" For Clinical Cultures:	For IC & Clinical Cultures: \CPC+	Final	Yes	

	Test Comment	**Isolate Comment**	**Report Status**	**Notification to ICP/Ward**	**Other Instructions**
	"UPDATED REPORT"				
Negative NML Report	UPDATED REPORT }KPCN		Final	Yes	For IC Screen: Suppress previously reported Isolate
Positive NML Report	For IC Screen: "UPDATED REPORT" "POSITIVE Carbapenemase Screen" For Clinical Cultures: "UPDATED REPORT"	For IC & Clinical Cultures: \KPCP	Final	Yes	Enter report for genes in kpcros panel

CPO Reporting Canned Messages

A. *Acinetobacter spp*. Reporting Messages

LIS Code	Canned Message
TEST COMMENTS	
}NCRE	Negative – No carbapenemase-producing organism (CRE) isolated
ISOLATE COMMENTS	
\ANML	~This organism is meropenem non-susceptible. ~Further characterization from the National Microbiology ~Laboratory to follow.
\ACCN	This organism is NEGATIVE for carbapenemase genes by PCR; as reported by the National Microbiology Laboratory (NML) 1015 Arlington St. Winnipeg, MB. Canada, R3E 3R2. NML Specimen No.
\ACCP	This organism is POSITIVE for ___ carbapenemase (add specific carbapenemase that is confirmed) based on PCR; as reported by the National Microbiology Laboratory (NML) 1015 Arlington St. Winnipeg, MB. Canada, R3E 3R2. NML Specimen No. The NML assay is able to detect NDM, KPC, OXA-48, OXA-181, OXA-232, OXA-244, IMP-1, VIM, NMC, and IMI as well as OXA-58, OXA-51, OXA-23, OXA-24, OXA-235, and OXA-143 carbapenemases. If you have any questions, please contact the Medical Microbiologist on call.

B. Enterobacterales Reporting Messages

TEST COMMENTS

LIS Code	Canned Message
}NCRE	Negative – No carbapenemase-producing organism (CRE) isolated
}KPCN	The previously reported positive carbapenemase result for was NOT confirmed. This organism is NEGATIVE for carbapenemase genes by PCR; as reported by the National Microbiology Laboratory (NML) 1015 Arlington St. Winnipeg, MB. Canada, R3E 3R2. The NML assay is able to detect NDM, KPC, OXA48, OXA181, OXA232, OXA244, IMP-1, VIM, NMC, IMI, and SME carbapenemases. If you have any questions, please contact the Medical

	Microbiologist on call.

ISOLATE COMMENTS

LIS Code	Canned Message
\nCRE	Additional testing indicates that this organism does NOT produce a carbapenemase
\CPHL	~Phenotypic testing cannot rule out carbapenemase ~production. Genotypic confirmation from ~Public Health Ontario Laboratory to follow.
\CREP	Phenotypic testing suggests this organism is carbapenemase POSITIVE as previously confirmed on yyyy.mm.dd.
\PCRB	~This organism is phenotypically carbapenemase POSITIVE ~by the βCARBA test (Bio-Rad). ~Genotypic confirmation to follow.
\pCRB	~This organism is phenotypically carbapenemase POSITIVE ~by the βCARBA test (Bio-Rad). No carbapenemase genes ~were detected by the Cepheid Xpert CARBA-R Assay ~(for research use only). ~This assay is able to detect NDM, KPC, OXA48, OXA181, ~OXA232, OXA244, IMP-1, and VIM carbapenemase genes. ~Additional genotypic testing from the National ~Microbiology Laboratory to follow.
\CPC+	_____ 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.
\KPCP	This organism is POSITIVE for ___ carbapenemase (add specific carbapenemase that is confirmed) based on PCR; as reported by the National Microbiology Laboratory (NML) 1015 Arlington St. Winnipeg, MB. Canada, R3E 3R2. The NML assay is able to detect NDM, KPC, OXA48, OXA181, OXA232, OXA244, IMP-1, VIM, NMC, IMI, and SME carbapenemases. If you have any questions, please contact the Medical Microbiologist on call

APPENDIX D. SUSCEPTIBILITY TESTING METHODOLOGIES:

I – Disk Diffusion

I. Introduction

The disk diffusion method of susceptibility testing (also known as the Kirby-Bauer (KB) method) has been standardized primarily for testing of rapidly growing bacteria. To perform the test, filter paper disks impregnated with a specific amount of antimicrobial agent are applied to the surface of an agar medium that has been inoculated with a known amount of the test organism. The drug in the disk diffuses through the agar. As the distance from the disk increases, the concentration of the antimicrobial agent decreases creating a gradient of drug concentrations in the agar medium. Concomitant with diffusion of the drug, the bacteria that were inoculated and that are not inhibited by the concentration of the antimicrobial agent continue to multiply until a lawn of growth is visible. In areas where the concentration of drug is inhibitory, no growth occurs, forming a zone of inhibition around each disk. Criteria currently recommended for interpreting zone diameters and MIC results for commonly used antimicrobial agents are published by CLSI. Results are reported categorically as Susceptible (S), Intermediate (I), or Resistant I.
For *E. coli, Klebsiella* species and *Proteus* species, instead of using standard cutoffs to determine S, I or R, screening test cutoffs are used and interpretations as R and S are reported if zone size is < or > of these screening breakpoints.

p. **Materials**

Antimicrobial disks (store frozen with a desiccant)
Mueller Hinton Agar (MH)
Mueller Hinton Blood Agar (MHB)
Haemophilus Test Media (HTM)
Trypticase Soy Broth (TSB) (3 mL)
VITEK colorimeter
Sterile saline
Sterile swabs

p. **Procedure**

a. Allow disks to come to room temperature before opening the container.
b. Using the Vitek colorimeter, prepare a suspension of the test organism in sterile saline equivalent to a 0.5 McFarland standard using isolated colonies. If there is not enough growth, inoculate the organism into TSB, and incubate at 35^{o}C for 2-4 hours or until it reaches the turbidity of a 0.5 McFarland standard (with the colorimeter adjusted for TSB).
c. Using a sterile cotton swab, inoculate the organism onto an appropriate agar plate, streaking in 3 directions over the entire agar surface. For organisms that grow rapidly use MH agar. For

Haemophilus species use HTM and for *S. pneumoniae*, beta-haemolytic *streptococcus* and viridans *streptococcus* use MHB. For other organisms that do not grow on MH, use MHB.

d. Using forceps or a disk dispenser, apply the appropriate Antimicrobial disks onto the agar. Place the disks with an equal distance apart from each other and put no more than 6 disks on a 100mm diameter plate.

e. Incubate plates as follows:

Campylobacter species – microaerophilically at 35°C x 18 hours
Haemophilus species – CO_2, 35°C x 18 hours
S. pneumoniae – CO_2, 35°C x 20 to 24 hours
Beta-haemolytic *streptococcus* – CO_2, 35°C x 20 to 24 hours
viridans *streptococcus* - CO_2, 35°C x 20 to 24 hours
S. aureus and *Enterococcus* species for Methicillin and Vancomycin – O_2, 35°C x 24 hours
Others – O_2, 35°C x 18 hours

p. **Interpretation**

After incubation, measure the diameters of the zone of complete inhibition (as judged by the unaided eye) with 81ipro81ti.

For MH and HTM agar (except for *Staphylococcus* spp. –oxacillin, vancomycin):

1. Measure from the back of the plate.
2. Hold the petri dish a few inches above a black, nonreflecting background illuminated with **reflected light**.
3. The zone margin should be considered the area showing no obvious, visible growth that can be detected with the unaided eye. Ignore faint growth of tiny colonies that can be detected only with a magnifying lens at the edge of the zone of inhibited growth.
4. Strains of *Proteus* spp. May swarm into areas of inhibited growth around certain antimicrobial agents. With *Proteus* spp., ignore the thin veil of swarming growth in an otherwise obvious zone of growth inhibition.
5. With trimethoprim and the sulfonamides, antagonists in the medium may allow some slight growth; therefore, disregard slight growth (20% or less of the lawn of growth) and measure the more obvious margin to determine the zone diameter.

For *Staphylococcus* spp. – oxacillin, vancomycin:

1. Measure from the back of the plate.
2. Use **transmitted light** (plate held up to light source).
3. The zone margin should be considered the area showing no obvious, visible growth that can be detected with the unaided eye.
4. Any discernable growth within the zone of inhibition is indicative of resistant.

For MHB agar:

1. Measure the zones from the upper surface of the agar illuminated with **reflected light** and with the cover removed.
2. The zone margin should be considered the area showing no obvious, visible growth that can be detected with the unaided eye. Ignore faint growth of tiny colonies that can be detected only with a magnifying lens at the edge of the zone of inhibited growth.

Refer to CLSI Document M100-S23 for the zone size interpretations. Report susceptible, resistant and intermediate as appropriate.

p. **Quality Control**

Check for pure culture before recording test results. Retest if disk diffusion plate appears to be of mixed culture.

Test the following organisms each time a new batch of MH agar is prepared and once weekly. Subculture the organisms from the BHI slant (stored refrigerated) to BA the day before setting up the QC.

For weekly QC on MH:
S. aureus ATCC 25923
E. coli ATCC 25922
P. aeruginosa ATCC 27853

For weekly QC on HTM:
Haemophilus 82ipro82tib ATCC 49247
Haemophilus 82ipro82tib ATCC 10211 (test for growth)

For weekly QC on MHB:
Streptococcus pneumonieae ATCC 49619

For each new batch of MH:
S. aureus ATCC 25923
E. coli ATCC 25922
P. aeruginosa ATCC 27853
S. faecalis ATCC 29212

For each new batch of HTM:
Haemophilus 82ipro82tib ATCC 49247
Haemophilus 82ipro82tib ATCC 10211 (test for growth)

See CLSI Document M100-S26 Table 3 for acceptable QC results.
For troubleshooting out-of range QC results, see CLSI Document M100-S26 Table 3C.

p. **Reference**

Clinical and Laboratory Standards Institute (CLSI) Document – Performance Standards for Antimicrobial Disk Susceptibility Testing M02-A11, 2012

Clinical and Laboratory Standards Institute (CLSI) Document – Performance Standards for Antimicrobial Disk Susceptibility Testing M100-S23, 2013

Toma, E., Barriault, D. Antimicrobial Activity of Fusidic Acid and Disk diffusion susceptibility Testing Criteria for Gram-Positive Cocci *J Clin Microbiol* 1995; 33:1712-1715

Finelay, J.E., Miller, A., Poupard, J.A. Interpretive Criteria for Testing Susceptibility of Staphylococci to Mupirocin *J Clin Microbiol* 1997; 41:1137-1139

Fuchs, P.C., Jones, R.N., Barry, A.L. Interpretive Criteria for Disk Diffusion Susceptibility Testing of Mupirocin, a Topical Antibiotic *J Clin Microbiol* 1990; 28:608-609

II – Double Disk Diffusion for Erythromycin and Clindamycin on *Staphylococcus* species, ß-haemolytic *Streptococci* Groups A, B, C, G and *Streptococcus pneumoniae*

I. Introduction

Macrolide (erythromycin) resistant *Staphylococcus* species, ß-haemolytic *Streptococci* and *Streptococcus pneumoniae* isolates may have constitutive or inducible resistance to lincosamides (clindamycin). The mechanisms of resistance include:

- Ribosomal modification encoded by an *erm* gene; also refer to as MLS_B (macrolide, lincosamide and type B streptogramin) resistance.
- Efflux of the antibiotic encoded by a *mef* gene; resistant only to macrolide
- Drug inactivation

Inducible clindamycin resistance can be detected using a disk approximation test with a clindamycin disk placed 12 mm from an erythromycin disk as part of the normal disk diffusion test.

II. Materials

Antimicrobial disks – clindamycin (DA, 2 µg) and erythromycin (E, 15 µg)
Mueller Hinton Agar (MH) – for *Staphylococcus* species
Mueller Hinton Blood Agar (MHB) – for *Streptococcus* species
VITEK colorimeter
Sterile saline
Sterile swabs

III. Procedure

p. Allow disks to come to room temperature before opening the container.

p. Using the Vitek colorimeter, prepare a suspension of the test organism in sterile saline equivalent to a 0.5 McFarland standard using isolated colonies.

3. Using a sterile cotton swab, inoculate the standardized organism onto a MH or MHB agar plate, streak in three directions over the entire agar surface.

4. Place plate on disk template (Figure 1.)

5. Using forceps or a disk dispenser, apply the clindamycin and erythromycin disks onto the agar, 15 mm to 26 mm away for staphylococci or 12 mm away for streptococci, from edge to edge using template below (Figure 1). Other antimicrobial disks can be placed on the same agar plate if needed.

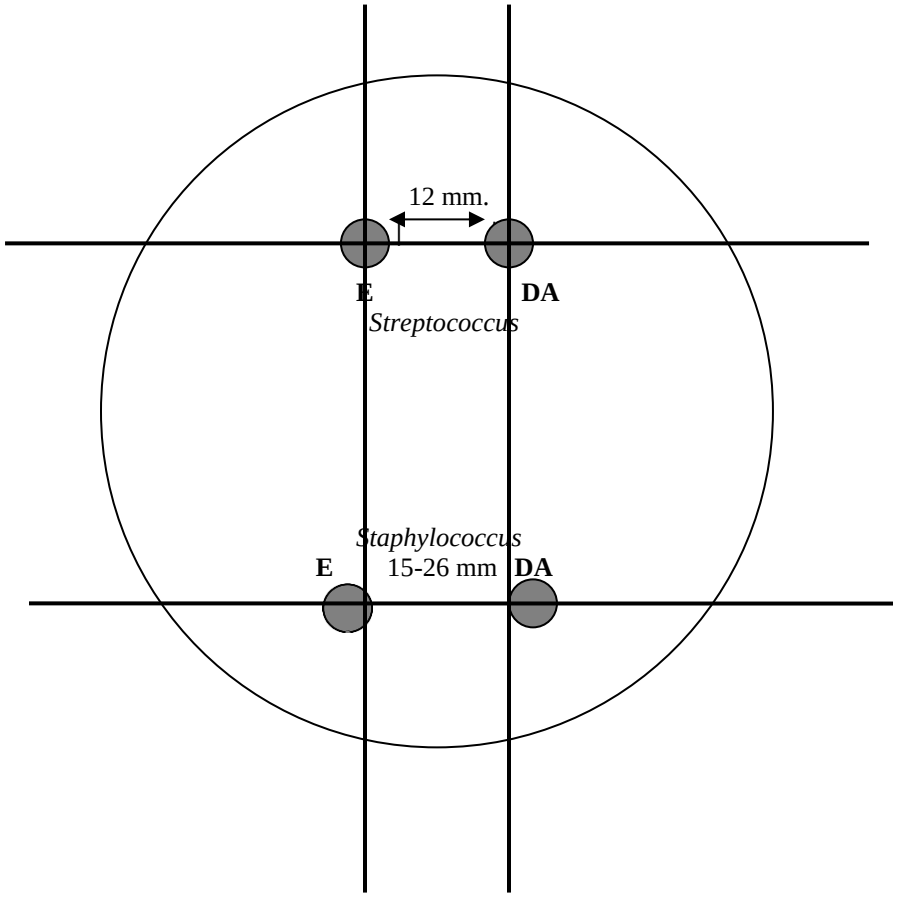


Figure1. Template for Clindamycin and Erythromycin disks placement

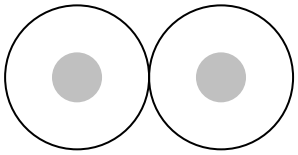
p. For *Staphylococcus*, incubate plates in O_2 at 35°C for 20 to 24 hours. For *Streptococcus*, incubate plates in CO_2 at 35°C for 20 to 24 hours

IV. Interpretation

1. After incubation, measure the diameters of the zone of complete inhibition with callipers/ruler. Measure at the narrowest side of the zone. Refer to Clinical and Laboratory Standards Institute (CLSI) Document – M100 for the zone size interpretations.
2. Enter zone size measurements into the LIS.
3. Organisms that show flattening of the clindamycin zone adjacent to the erythromycin disk in the shape of the letter D (referred to as a "D" zone) have inducible clindamycin resistance. Enter into the LIS under LIS drug "D zone" the presence or absence of "D" zone as "Y" or "N". Isolates that

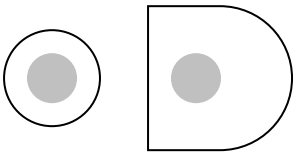
show the presence of D zone will be automatically reflexed in the LIS to report as "clindamycin resistant".

Examples of Zone of Inhibition Patterns and their Interpretation:



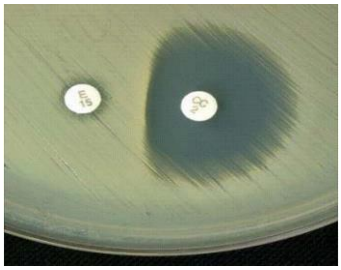
E **DA**

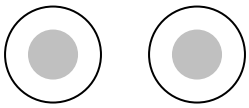
Both E and DA are Susceptible.
Report both E and DA as S



E **DA**

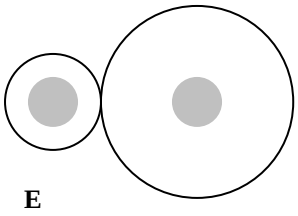
Both E and DA (measured at the narrowest side) are I or R; "D" zone is positive – Inducible MLS_B; presumed genotype: *erm*
Report both E and DA as R





E DA

Both E and DA I or R – Inducible or constitutive MLS_B; presumed genotype: *erm*
Report both E and DA as R



E

DA

E is I or R and DA is S – M phenotype; presumed genotype: *mef.*
Report E as I or R and DA as S.

V. Quality Control

See Clinical and Laboratory Standards Institute (CLSI) Document – M100-S23 Table 3 for acceptable QC results.

VI. References

Clinical and Laboratory Standards Institute (CLSI) Document – Performance Standards for Antimicrobial Disk Susceptibility Testing M2-A10, 2009.

Clinical and Laboratory Standards Institute (CLSI) Document – Performance Standards for Antimicrobial Disk Susceptibility Testing Information Supplement Table 2H M2-Disk Diffusion M100-S23, 2013.

Quality Management Program-Laboratory Services (QMP-LS) Committee Comments BACT-020, Vol. 3, 2.2:721-724.

Streptococci and *Staphylococcus* (overview of macrolides and lincosamide resistance)
Leclercq CID 2002; 34:482-92

Streptococcus pneumoniae
Descheemaeker et al JAC 2000 45:167-173

Beta-haemolytic streptococcus (Groups A, B, C, G)
GAS Descheemaeker et al. JAC 2000 45:167-173
GBS de Azavedo et al. AAC 1001;45:3504-3508
GCS & GGS Kataja et al. AAC 1998;42:1493-1494

III – Double Disk Test for ESBL Confirmation

Introduction

Cefpodoxime*, third generation cephalosporins, and aztreonam are all extremely susceptible to ESBLs and can be used as screening agents to test for the presence of ESBLs. CLSI suggests using screening MIC and disk diffusion zones breakpoints for these antibacterials that are distinct from treatment breakpoints to screen for ESBLs.

When *E. coli*, *Klebsiella* species or *Proteus* species are cefpodoxime resistant by Vitek OR either cefpodoxime or any 3rd generation cephalosporin or aztreonam are tested "resistant" by disk diffusion and screening breakpoints are used, confirmation of the presence of ESBL can be determined by the double disk test.

*Cefpodoxime alone can be used to screen for the presence of ESBL.UHN/MSH data from isolates in 2000 to 2006 did not reveal any *E. coli*, *Klebsiella* species or *Proteus* species that are cefpodoxime susceptible but 3rd generation cephalosporin or aztreonam resistant.

p. **Materials**

Mueller-Hinton (MH) agar (150) mm
20/10 mg amoxicillin-clavulanate disk
30 mg ceftazidime disk
30 mg ceftriaxone or cefotaxime disk
30 mg aztreonam disk
10 mg cefpodoxime disk (optional)
30 mg cefoxitin disk
30 mg cefepime disk
5 mg ciprofloxacin disk (for Infection Control Screen orders)
10 mg ertapenem disk (for Infection Control Screen orders or if Vitek susceptibility has not been done)
10 mg gentamicin disk (for Infection Control Screen orders)
10 mg meropenem disk (for Infection Control Screen orders)
110 mg piperacillin/tazobactam disk
Quality control strain: *E. coli* ATCC 35218

p. **Procedure**

1. Prepare a bacterial suspension of the organism to be tested that has a turbidity equivalent to a 0.5 McFarland standard.

2. Inoculate a Mueller-Hinton agar plate with this suspension in accordance with CLSI M100-S23 guidelines for disk diffusion testing.

3. Place the amoxicillin-clavulanic acid disk on the plate so that ceftriazone, ceftazidime, aztreoman and cefpodoxime disks may be placed around it with 15 mm between disk edges (See Figure 1. KB-ESBL Template). Add cefoxitin, cefepime and piperacillin/tazobactem disks on other parts of the plate. If Vitek susceptibility has not been done, add ertapenem disk.

4. For Infection Control screen orders, add ciprofloxacin, ertapenem and gentamicin disks. (See Figure 2. Infection Control KB-ESBL Template)

5. Incubate 35^{o}C, in O_2 x 18-24 hours and record the zone diameters for the all cephalosporins as per CLSI guidelines.

6. For *E. coli*, *Klebsiella* species and *Proteus* species, instead of using standard cutoffs to determine S, I or R, ESBL screening test cutoffs are used and interpretations as R and S are reported if zone size is < or > of these screening breakpoints.

Figure 1. KB-ESBL Template

To be used for ESBL Screen isolates where Vitek card has been set up.

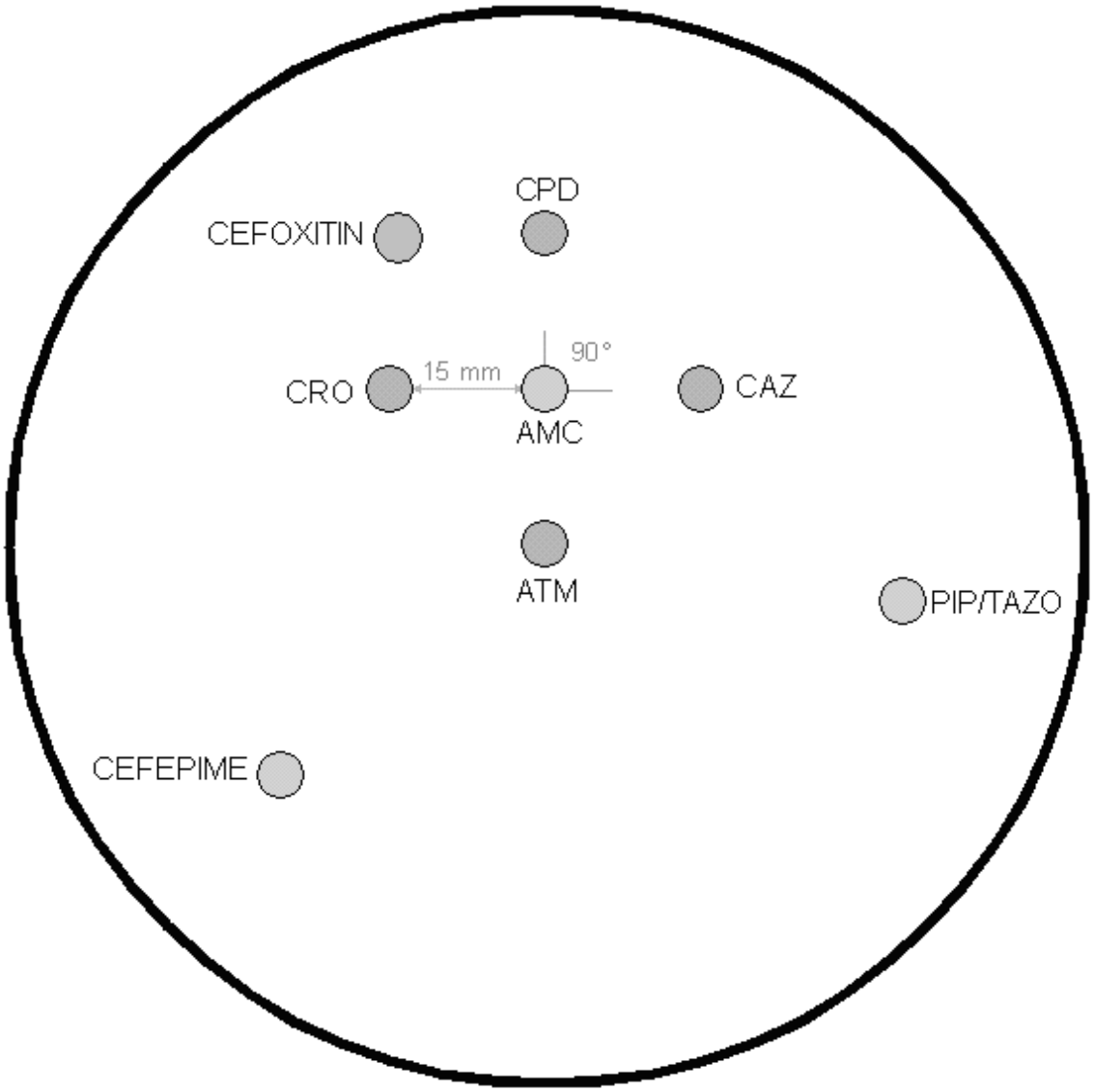
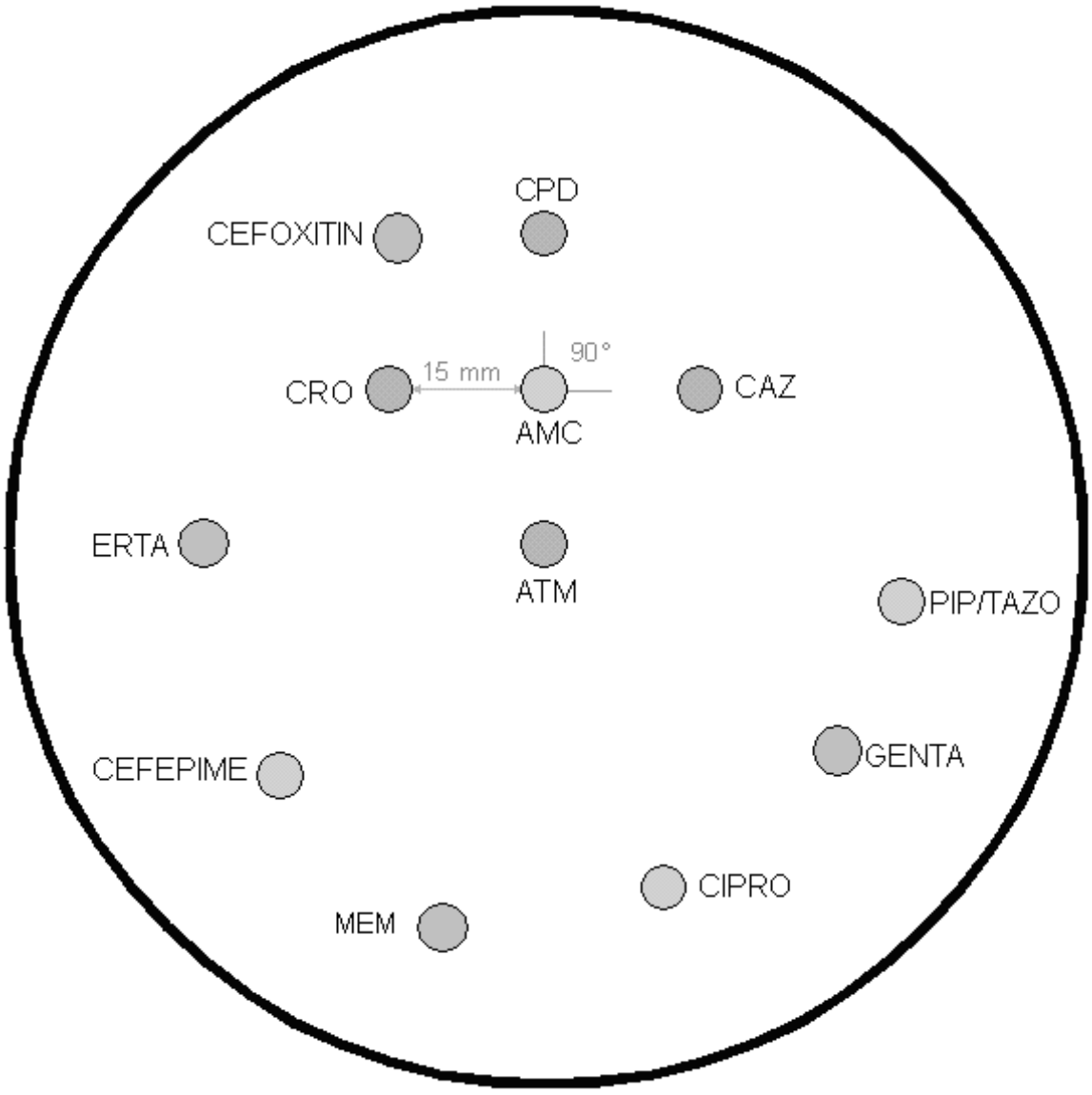


Figure 2. Infection Control KB-ESBL Template

To be used for Infection Control ESBL Screen isolates where Vitek card has NOT been set up.



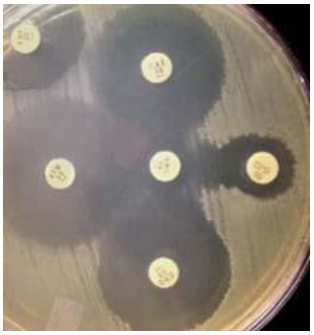
p. **Interpretation**

Note: The following applies to cefpodoxime-nonsusceptible *E. coli*, *Klebsiella* species and *Proteus* species only.

1. After incubation, measure the diameters of the zone of complete inhibition with 92ipro92ti/ruler. Measure at the narrowest side of the zone.
2. Document zone size for all antibiotics into the LIS.
3. Observe for **potentiation** of the inhibition zone (i.e. **increase** in the inhibition zone) of any one of cefpodoxime, ceftazidime, ceftriaxone or aztreonam when combined with clavulanic acid (enter **Y**es or **N**o to the "drug" named "Potentiation" in the LIS).
4. If a **reduction** of zone of inhibition of any one of cefpodoxime, ceftazidime, ceftriaxone or aztreonam when combined with clavulanic acid is observed (i.e. a D zone formation), enter **Y**es or **N**o to the "drug" named "D zone" in the LIS. Recheck the identification of the isolate and repeat testing if the identification is questionable.
5. If a reduction of zone of inhibition of either cefpodoxime or ceftriaxone when combined with cefoxitin is observed (ie. A D zone formation), enter **Y**es or **N**o to the "drug" named "D zone" in the LIS.

Class A ESBL present:

i) Potentiation of the inhibition zone of any one of cefpodoxime, ceftazidime, ceftriaxone or aztreonam when combined with clavulanic acid (see below for examples of different patterns of potentiation that can be seen with organisms that contain Class A ESBLs)
ii) Susceptibile to cefoxitin.
iii) Susceptibile, Intermediate or Resistant to any one of ceftazidime, ceftriaxone or aztreonam


Class A and Class C ESBL present:

i) Potentiation of the inhibition zone of any one of cefpodoxime, ceftazidime, ceftriaxone or aztreonam when combined with clavulanic acid
ii) Resistant or Intermediate to cefoxitin.
iii) Susceptibile, Intermediate or resistant to any one of ceftazidime, ceftriaxone or aztreonam

Class C-ESBL present:

i) No potentiation with clavulanic acid
ii) Resistant or Intermediate to cefoxitin
iii) Resistant to any one of ceftazidime, ceftriaxone or aztreonam.

Inducible Class C-ESBL present:
i) No potentiation with clavulanic acid
ii) Resistant or Intermediate to cefoxitin
iii) Susceptible, Intermediate or Resistant to any one of ceftazidime, ceftriaxone or aztreonam.
iv) D zone with clavulanic acid against ceftazidime, ceftriaxone, aztreonam or cefpodoxime
v) D zone with cefoxitin against ceftriaxone or cefpodoxime

ESBL not Class A or Class C present:
i) No potentiation with clavulanic acid
ii) Susceptibile to cefoxitin
iii) Resistant to any one of ceftazidime, ceftriaxone or aztreonam

ESBL absent:
i) No potentiation with clavulanic acid
ii) Susceptibile, Intermediate or resistant to cefoxitin
iii) Susceptibile to all of ceftazidime, ceftriaxone or aztreonam

V. Reporting

Reporting Comment	**Potentiation** of the inhibition zone of any one of cefpodoxime, ceftazidime, ceftriaxone or aztreonam when combined with clavulanic acid (enter **Y** or **N** to the "drug" "Potentiation" in the LIS)	**Cefoxitin**	**Ceftazidime, ceftriaxone or aztreonam**	**D zone** (enter **Y** or **N** to the "drug" "D zone" in the LIS)
The susceptibility pattern suggests that this organism contains a class A extended spectrum beta-lactamase (ESBL).	**Y**es	S	S/I/R	N
The susceptibility pattern suggests that this organism contains class A and C extended spectrum beta-lactamases (ESBL).	**Y**es	I/R	S/I/R	N
The susceptibility pattern suggests that this organism	**Y**es	I/R	S/I/R	Y

Reporting Comment	**Potentiation** of the inhibition zone of any one of cefpodoxime, ceftazidime, ceftriaxone or aztreonam when combined with clavulanic acid (enter **Y** or **N** to the "drug" "Potentiation" in the LIS)	**Cefoxitin**	**Ceftazidime, ceftriaxone or aztreonam**	**D zone** (enter **Y** or **N** to the "drug" "D zone" in the LIS)
contains class A and an inducible class C extended spectrum beta-lactamases (ESBL).				

Reporting Comment	**Potentiation** of the inhibition zone	**Cefoxitin**	**Ceftazidime, ceftriaxone or aztreonam**	**D zone**
The susceptibility pattern suggests that this organism contains a class C extended spectrum beta-lactamase (ESBL).	**N**o	I/R	R	N
The susceptibility pattern suggests that this organism contains an inducible class C extended spectrum beta-lactamase (ESBL).	**N**o	I/R	S/I/R	Y
The susceptibility pattern suggests that this organism contains an extended spectrum beta-lactamase (ESBL) other than class A or C.	**N**o	S	R	N
Not ESBL – no reporting comment	**N**o	S/R	S	N

For *E. coli*, *Klebsiella* species and *Proteus* species that are confirmed to have an ESBL of any class, report all pencillins and first, second and third generation cephalosporins as R; for Class A, also report fourth generation cephalosporins (i.e. cefepime) as R.

For carbapenemase reporting, see **Carbapenemase Reporting** section.

p. **References**

p. Cormican MG, Marshall SA, Jones RN. 1996. Detection of extended-spectrum beta-lactamase (ESBL)-producing strains by the Etest ESBL screen. J. Clin. Microbiol. 8:1880-1884.

p. Jacoby GA, Han P. 1996. Detection of extended-spectrum beta-lactamases in clinical isolates of *Klebsiella pneumoniae* and *Escherichia coli*. J. Clin. Microbiol. 34:908-911.

3. Jacoby GA. 1994. Genetics of extended-spectrum beta-lactamases. Eur. J. Clin. Microbiol. Infect. Dis. Suppl 1,13:2-11.

4. Livermore DM. 1995. Beta-lactamases in laboratory and clinical resistance. Clin. Microbiol. Rev.8:557-584.

p. Moland ES, Sanders CC, Thomson KS. 1998. Can results obtained with commercially available MicroScan microdilution panels serve as an indictor of beta-lactamase (ESBL)-production among *Escherichia coli* and *Klebsiella* isolates with hidden resistance to expanded spectrum cephalosporins and aztreonam? J. Clin. Microbiol. 36:2575-2579.

p. CLSI. Performance standards for antimicrobial susceptibility testing; Eighth informational supplement. CLSI document M100-S23 [ISBN 1-26238-337-X]. CLSI, 940 West Valley Road, Suite 1400, Wayne, Pennsylvania 19087-1898 USA, 2013)

p. Sanders CC, Barry AL, Washington JA, Shubert C, Moland ES, Traczewski MM, Knapp C, Mulder R. 1996. Detection of extended-spectrum beta-lactamase-producing members of the family Enterobacterales with the Vitek ESBL test. J. Clin. Microbiol. 34:2997-3001.

p. Swenson JM, Ferraro MJ, Jorgensen JH, Tenover FC. 1996. Can different breakpoints be used to detect extended-spectrum beta-lactamase (ESBL)-producing organisms? Abstr D-22, ICAAC meeting, New Orleans, Louisianna, Sept 15 -18, 1996.

p. Vercauteren E, Descheemaeker P, Ieven M, Sanders CC, Goossens H. 1997. Comparison of screening methods for detection of extended-spectrum beta-lactamases and their prevalence among blood isolates of *Escherichia coli* and *Klebsiella* spp. In a Belgian teaching hospital. J. Clin. Microbiol. 35:2191-2197.

IV Ceftazidime-Avibactam and Aztreonam Combination Test

Introduction:
Aztreonam plus ceftazidime-avibactam is a preferred antimicrobial regimen for the treatment of metallo-β-lactamase (MBL) expressing Enterobacterales, or *Stenotrophomonas maltophilia* isolates that are resistant to all other first line agents (1). Rarely, MBL-expressing *Pseudomonas aeruginosa* may also be susceptible to this combination. This section describes the testing of aztreonam-avibactam synergy by double-disk diffusion method (2,3) for these organism types.

Reagents/Materials/Media:
Mueller Hinton (MH) agar (150 mm)
30-µg aztreonam disks
30/20-µg ceftazidime-avibactam disks
Sterile saline
Sterile swabs

Quality Control Organisms:
The following quality control organisms should be run with each batch of clinical isolates.

- *Klebsiella pneumoniae* ATCC 2146
  - Expected result: Resistant to aztreonam and ceftazidime-avibactam in isolation, susceptible to combination. Expected zone of potentiation of ≥21mm
- Clinical Isolate ENT18
  - Expected result: Resistant to aztreonam and ceftazidime-avibactam in isolation, as well as in combination. No zone of potentiation expected.

Procedure:

1. Preparation of inoculum
   a. A standard inoculum should be made as described in APPENDIX D. SUSCEPTIBILITY TESTING METHODOLOGIES
   b. If performing testing on a previously frozen isolate, maintain antibiotic pressure by placing a meropenem disk in the main inoculum, and collecting bacterial growth from around this disk for testing of aztreonam-avibactam synergy.

2. Preparation and inoculation of plate
   a. Using a sterile cotton swab, inoculate the organism onto a Mueller-Hinton agar plate, streaking in 3 directions over the entire agar surface.
   b. Using forceps, apply aztreonam and ceftazidime-avibactam disks onto the agar, aligned 20mm from center to center, using the template in **Figure 1.**

3. Incubation
    a. Incubate plates at 35ºC, ambient air x 18 hours

4. Interpretation
    a. Visually assess plates for zones of potentiation, which should meet the following criteria (See **Illustrations A** and **B** for examples):
        i. Zone of potentiation must arise from the aztreonam disk.
        ii. There must be no detectable bacterial growth between edge of aztreonam disk and zone of potentiation.
    b. Measure zone of potentiation as depicted in **Illustrations C** and multiply by 2 to obtain a zone diameter.

5. Troubleshooting
    a. Perform quality control assessment as outlined in APPENDIX D. SUSCEPTIBILITY TESTING METHODOLOGIES.
    b. Large zones of clearance around either aztreonam or ceftazidime-avibactam disk (in the range of susceptible zone breakpoint measurements) may indicate loss of ESBL/MBL-expressing plasmid. If this is found, repeat testing with a fresh sub-culture with meropenem disk placed in main inoculum to maintain antibiotic pressure.

Reporting:

- Use CLSI M100 aztreonam zone measurement breakpoints for Enterobacterales and *P. aeruginosa* to determine whether the organism is susceptible to the combination of aztreonam and ceftazidime/avibactam. For *S. maltophilia*, use CLSI *Pseudomonas aeruginosa* breakpoints
    - Interpret susceptibility or non-susceptibility based on zone sizes ≥ or < the susceptible breakpoint, respectively.
- Report visual appearance (i.e. potentiation, "yes" or "no") and zone size of QC organisms and the clinical isolate.
- If **NO** potentiation drop KB panel "kbceta-" and enter result.
- If **YES** for potentiation drop KB panel "kbceta+" and enter result and zone size.
- Based on zone size interpretation populate with appropriate isolate comment:
    - If zone measurement is within susceptible range for the appropriate organism type, report with ISOLATE COMMENT code\atas: *"Aztreonam and Ceftazidime-Avibactam Combination Testing: Susceptibility to the combination of aztreonam and ceftazidime-avibactam was demonstrated using the double-disk diffusion method. Please contact the microbiologist-on-call with any questions."*
    - If zone measurement is NOT within susceptible range, report with ISOLATE COMMENT code \atar: *"Aztreonam and Ceftazidime-Avibactam Combination Testing: NO susceptibility to the combination of aztreonam and ceftazidime-avibactam was demonstrated using the double-disk diffusion method. Please contact the microbiologist-on-call with any questions."*

- Use the following template to set up the Aztreonam and Ceftazidime-Avibactam disksensuring a **14 mm** distance between the inner edges of the two disks..
  Note:
  1. Add **separated** Aztreonam and Ceftazidime-Avibactam disks for Stenotrophomonas maltophilia ONLY
  2. Record the zone size of Aztreonam and Ceftazidime-Avibactam in the workcard

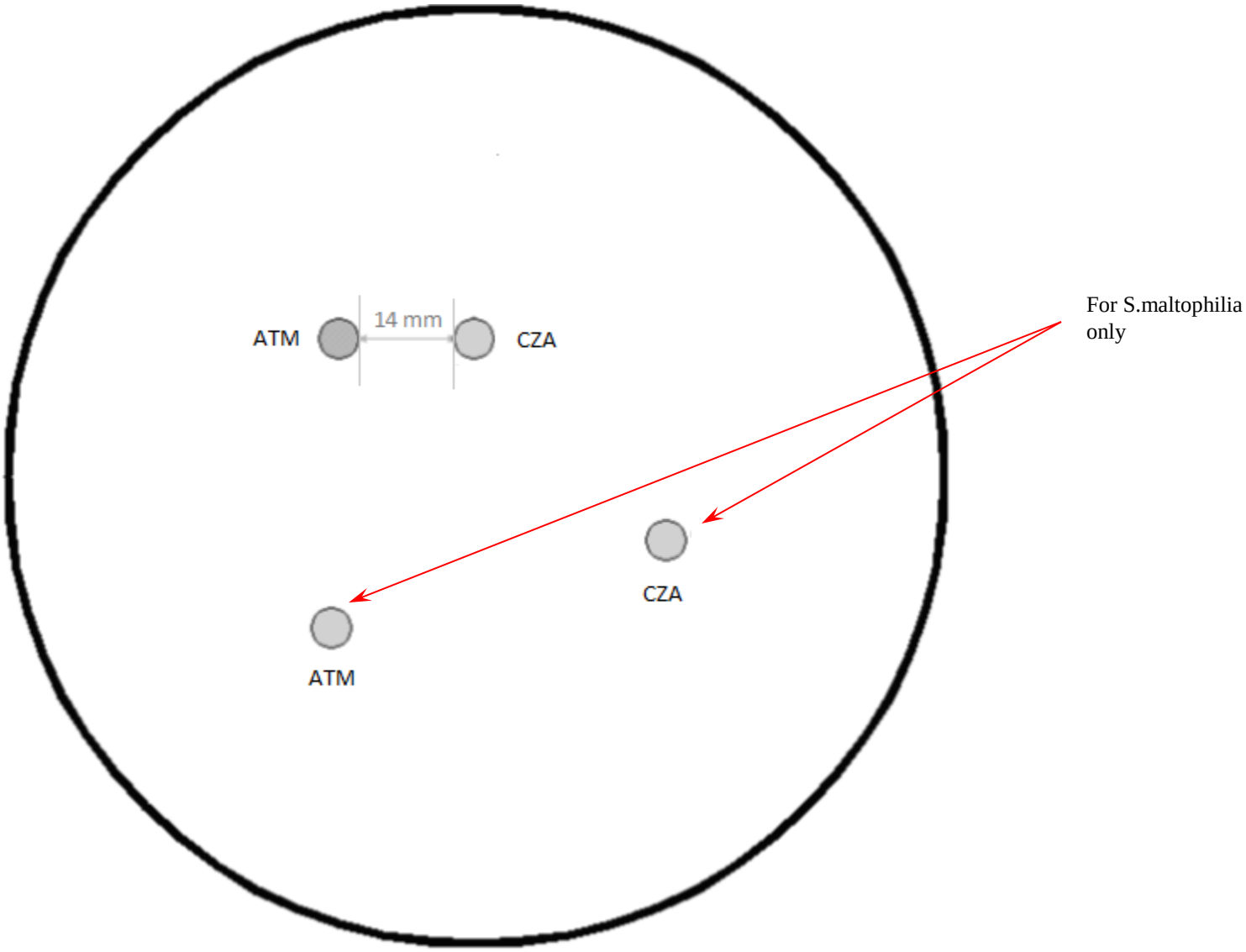
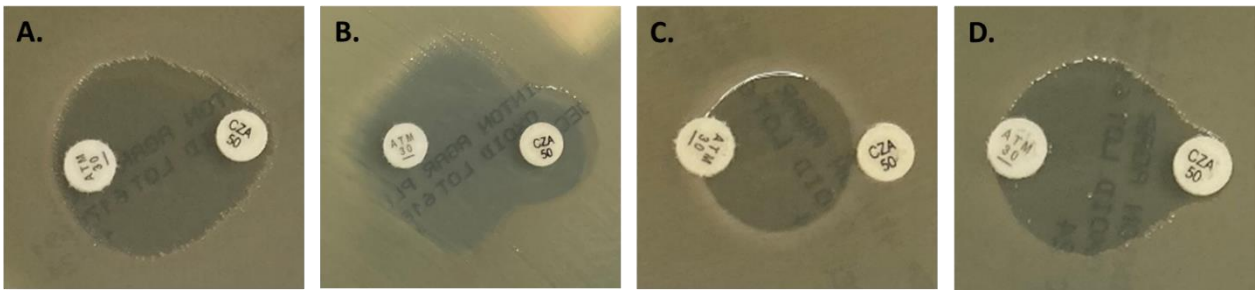


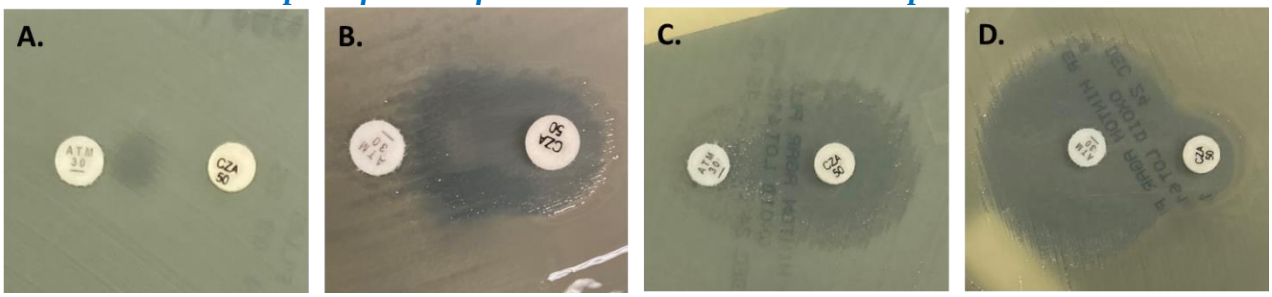
Figure 1: Plate template for aztreonam and ceftazidime-avibactam disk placement.

Illustrations:

Illustrations A: Examples of zones of clearance CONSISTENT with potentiation



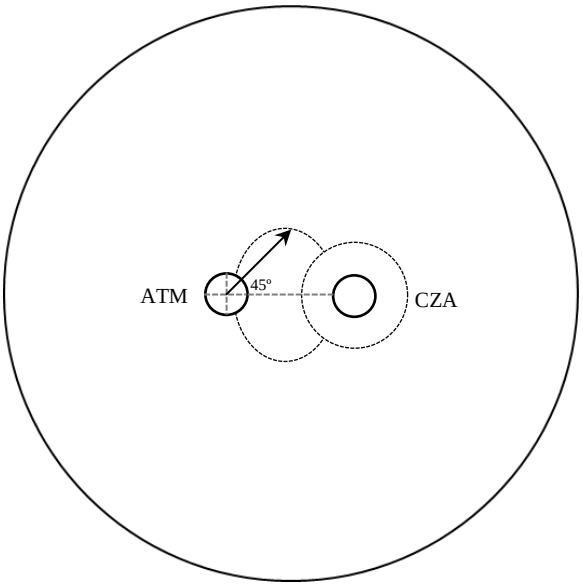
Illustrations B: Examples of zones of clearance NOT consistent with potentiation

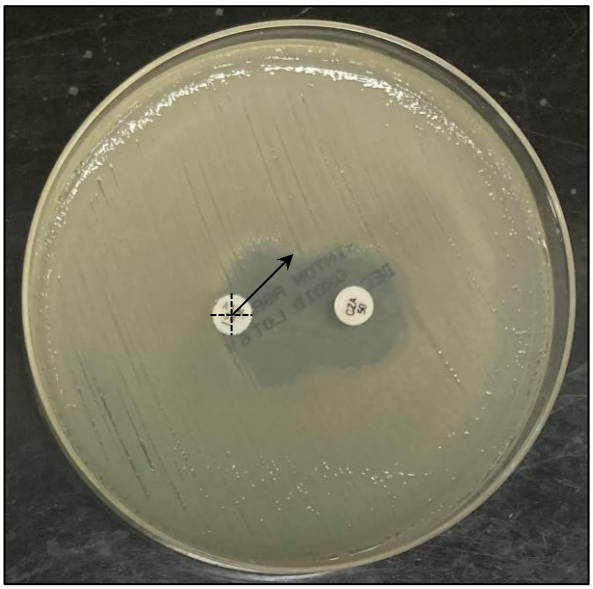


- In **panels A-C**, there is growth surrounding the entire aztreonam disk;
- In **panel D,** the zone around the aztreonam disk connects with that of the ceftazidime-avibactam disk; however, there is no true potentiation (i.e. increase in zone radius) between the two disks.

Illustrations C: Measurement of zone size

- *Only read/measure the potentiation zone if both Aztreonam (ATM) and Ceftazidime-Avibactam (CZA) show resistance, otherwise consult with seniors.*
- *Measure the zone of potentiation starting from the center of the ATM disk, at a 45° from the line connecting the centers of the ATM and CZA disks.*
- *Multiply the measured radius by **2** to obtain the zone diameter.*





References:

1. Tamma PD et al. Infectious Diseases Society of America 2024 Guidance on the Treatment of Antimicrobial-Resistant Gram-Negative Infections. Clin Infect Dis. 2024 Aug 7:ciae403. Doi: 10.1093/cid/ciae403. Epub ahead of print. PMID: 39108079.
2. Falcone M et al. Efficacy of Ceftazidime-avibactam Plus Aztreonam in Patients With Bloodstream Infections Caused by Metallo-β-lactamase–Producing Enterobacterales. Clin Infect Dis. 2020;72(11):1871–8.
3. Verschelden G et al. Aztreonam-avibactam synergy, a validation and comparison of diagnostic tools. Front Microbiol. 2023 Nov 29;14:1322180. Doi: 10.3389/fmicb.2023.1322180. PMID: 38094627; PMCID: PMC10716278.
4. Clinical and Laboratory Standards Institute (CLSI) Document – Performance Standards for Antimicrobial Susceptibility Testing M100, 2024.

V – Beta-Lactamase Testing

Introduction

Cefinase disks are intended for use in rapid testing of isolated colonies of *Neisseria gonorrhoeae, M. catarrhalis, Staphylococcus* species, *Enterococcus* species, *Haemophilus 102ipro102tib* and anaerobic bacteria for the production of beta-lactamase. Refer to CRITERIA FOR SUSCEPTIBILITY TESTING for appropriate beta-lactamase testing.

The Cefinase disk is impregnated with the chromogenic cephalosporin, Nitrocefin. This compound exhibits a very rapid colour change from yellow to red as the amide bond in the beta-lactam ring is 102ipro102tibi by beta-lactamase. When a bacterium produces this enzyme in significant quantities, the yellow-coloured disk turns red in the area where the isolate is smeared.

Although other penicillins and cephalosporins may be used as substrates for specific enzymes, Nitrocefin has the wide spectrum of susceptibility and sensitivity of the commercially available beta lactams. It is not known to react with other microbial enzymes.

Materials

Cefinase disks (BBL) (store refrigerated)
Sterile distilled water
Microscope slides
Sterile Pasteur pipettes

Procedure

1. Using forceps remove the required number of disks from the dispenser and place on a microscope slide. Use 1 disk per organism.

2. Using a sterile Pasteur pipette, moisten each disk with a drop of sterile water.

3. With a sterile loop or applicator stick, pick several similar colonies from the agar plate and smear onto the surface of the disk.

4. Observe the disk for up to 5 minutes for a colour change. For staphylococci, observe the disks for up to 60 minutes.

Interpretation

Positive: yellow to red colour change on the area where the culture was applied. Note: colour change does not usually develop over the entire disk. A negative result will show no colour change on the disk.

Negative: no colour change

For most bacterial strains a positive result will develop within 5 minutes. However, positive reactions for some staphylococci may take up to 1 hour to develop.

Organisms	Result	Approx. Reaction Time	Interpretation
Staphylococcus aureus	Positive	1 hr	Resistant to penicillin, ampicillin, carbenicillin. Probably susceptible to cephalothin, methicillin, oxacillin, naficillin and other penicillinase-resistant penicillins.
Enterococcus faecalis	Positive	5 min	Resistant to penicillin and ampicillin.
Haemophilus influenzae	Positive	1 min	Resistant to ampicillin Susceptible to cephalosporins.
Neisseria gonorrhoeae and *Branhamella catarrhalis*	Positive	1 min	Resistant to penicillin.
Anaerobic bacteria	Positive	30 mins	Probable identification is *Bacteroides* species. Probably resistant to penicillin and may be resistant to cephalosporins including cefotaxime and rarely cefoxitin.

Quality Controls

Set up positive and negative controls whenever a test is performed.

Haemophilus 103ipro103tib ATCC 35056: Positive
Haemophilus 103ipro103tib ATCC 10211: Negative

Reference

- Murray P.A., et al. Manual of Clinical Microbiology, 7th ed. 1999.
- BBL paper Disks for the Detection of β-Lactamase Enzymes, Cefinase Disks, package insert 2004/06.

VI – Oxacillin Screen Plate

Introduction

This is an agar dilution method using a single concentration of oxacillin incorporated into Mueller Hinton (MH) agar to screen for resistant strains of *S. aureus*.

Materials

Control plate (MH with 4% NaCl)
Screen plate (MH with 4% NaCl and 6 µg/mL Oxacillin)
VITEK colourimeter
Sterile saline
Sterile swabs

Procedure

1. Using the VITEK colourimeter, prepare a suspension with isolated colonies of the test organism (from solid medium after overnight culture) in sterile saline equivalent to a 0.5 McFarland standard (inoculum prepared for VITEK can be used).

2. Retrieve OXA, NACL, BHI with casein and VISA plates from fridge and their corresponding registration label (Lot number and expiry date). Affix the appropriate label to the reverse side of the worksheet OXACILLIN AND VANCOMYCIN SCREEN RECORDING SHEET FOR *S. aureus*. Write the date of testing on the sheet

3. Using a sterile swab, spot inoculate the suspension onto the screen and control plates. Numerous organisms can be tested on one plate (use grid TEMPLATE).

4. After the inocula have dried, incubate the plate at 35^{o}C, O_2 for a total of 24 hours.

5. All resistant isolates on the screen plate must be checked for purity (e.g. Gram stain, *S. aureus* tube coagulase or slide agglutination and sub-culture). The resistance must be confirmed by PBP2a MRSA Screen. Send a preliminary report as ISOLATE: Methicillin-resistant *S. aureus* and report to infection control.

Interpretation

Growth on the screen plate indicates that the organism is methicillin resistant and therefore is considered resistant to all beta-lactam Antimicrobials (eg. Penicillin, oxacillin, cephalosporins). Note: test is valid only for organisms which grow on the control plate.

Quality Control

Controls must be tested each day. The organisms are to be sub-cultured from the TSA slant (in refrigerator) to Blood Agar each day.

Sensitive : *S. aureus* ATCC 29213

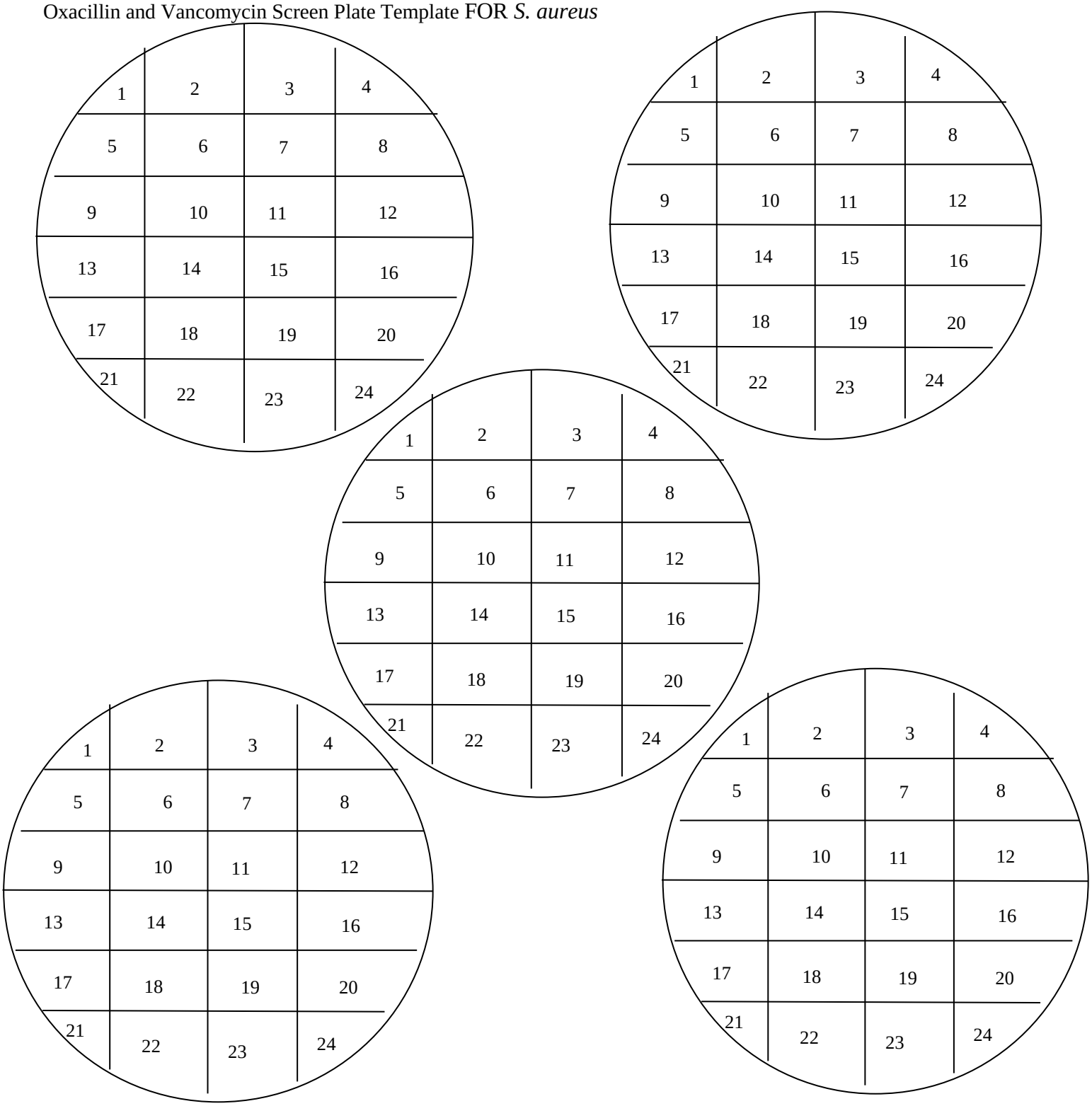
Sensitive/
Haze : *S. aureus* ATCC 43387

Resistant : *S. aureus* LPTP 8610-1
S. aureus ATCC 43300

Reference

Clinical and Laboratory Standards Institute (CLSI) Document – Methods for Dilution Antimicrobial Susceptibility Testing M7-A9, 2012.

Oxacillin and Vancomycin Screen Plate Template FOR *S. aureus*



OXACILLIN AND VANCOMYCIN SCREEN RECORDING SHEET FOR *S. aureus* page 1

Enter Lot number of OXA, NACL, VISA, BHI+casein on the back of this worksheet.

Date: **Set up by:**

18h OXA + VISA read by:

24h OXA + VISA read by:

48h VISA read by:

No. / Bench	ATCC Control OR Lab No.	OXA 18 h	OXA 24h	VISA 18 h	VISA 24h	VISA 48h	NACL Gr/NG	BHIA+ Casein Gr/NG	No. / Bench	Lab No.	OXA 18 h	OXA 24h	VISA 18 h	VISA 24h	VISA 48h	NACL Gr/NG	BHIA+ Casein Gr/NG
1	*S. aureus* **LPTP 8610**	R		S					13								
2	*S .aureus* **ATCC 43300**	R		S					14								
3	*S. aureus* **ATCC 29213**	S		S					15								
4	*S. aureus* **ATCC43387**	S							16								
	E.gallinarum **ATCC 49573**			R													
5									17								
6									18								
7									19								
8									20								
9									21								
10									22								
11									23								
12									24								

OXACILLIN AND VANCOMYCIN SCREEN RECORDING SHEET FOR *S. aureus* page 2

Policy #MI\ANTI\04\05b\v05

Lot number and expiry date labels:

OXA_____

NACL _____

VISA _____

BHIC_____

VII – PBP2 MRSA Screen

Principle

To be used as a screening test for the detection of Methicillin Resistant *S. aureus* (MRSA) from isolated colonies.

Reagents

PBP2a SA Culture Colony Test Kit (Alere)
Oxoid Needle
Vortex

Safety Precautions

Wear eye protection, protective clothing, protective gloves.

Method

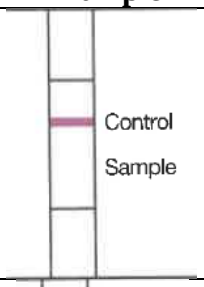
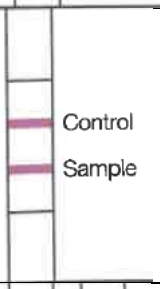
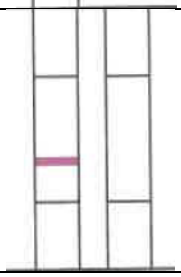
1. Holding the dropper bottle vertically, add two drops of Reagent 1 to an assay tube.
2. Take 3 well-grown-isolated colonies on the culture plate, place into the tube and thoroughly mix.
3. Holding the dropper bottle vertically, add two drops of Reagent 2 to the tube.
4. Vortex briefly. The blue solution must turn a clear color (if the color does not change, add one more drop of Reagent 2 and mix until the sample turns clear).
5. Insert the test strip into the tube with arrow pointed downward.
6. At five minutes, withdraw the strip from the tube and read the result.

   The control area is read at the top half of the strip

   The test area is read at the bottom half of the test strip

Interpretation

A positive result in interpreted by a pink/purple control line on any intensity (faint or strong appearance).

Test Area	Control Area	Example	Result
-	+		Negative – Not MRSA
+	+		Positive – MRSA
- or +	-		Invalid

Repeat any invalid test results with a new strip

Induced PBP2 MRSA Screen

For discrepant *S.aureus* results perform an **Induced** PBP2 MRSA Screen:

1. Prepare a 0.5 mcFarland standard using isolated colonies.
2. Using a sterile cotton swab, inoculate the organism onto MH agar with a cefoxitin disk. Incubate in O2 at 35^{o}C overnight.
3. Measure and document the zone size around the cefoxitin disk.
4. Repeat PBP2 MRSA Screen using growth closest to the cefoxitin disk.
5. If Induced PBP2 MRSA Screen is positive, it is a confirmed *S. aureus* report as MRSA.
   If cefoxitin-induced PBP2a is negative, check the Vitek Oxacillin MIC and Cefoxitin screen and Oxacillin screen result. Refer to How to Detect MRSA/BORSA section for futher testing and reporting.

Quality Control

Positive and negative controls must be set up once per week.

1. Positive: S.aureus ATCC 43300
2. Negative: S.aureus ATCC 29213

Reference

Alere Scarborough, Inc., Maine, USA, PBP2a SA Culture Colony Test insert 2017

VIII – Serum Bactericidal and Bactericidal Titres

I. Introduction

In the treatment of bacterial endocarditis or osteomyelitis, it maybe important to know whether the prescribed dosage of antimicrobials are achieving blood levels sufficiently high enough to kill the causative organism.

The bacteriostatic level is the dilution of serum that inhibits visible bacterial growth; the bactericidal level is the serum dilution that kills 99.9% of the initial inoculum.

NOTE: This test is to be performed only with the approval of a microbiologist.

II. Specimen Collection

The dose, the time the dose was given, and the time of collection must be recorded on the requisition. Pre- and post-dose blood specimens are obtained in serum separator tubes. The pre-dose blood specimen is drawn immediately before administering the next dose of antimicrobial in order to evaluate the pre (trough) level. Blood for the post-dose (peak) level should be drawn 1 hour after an intravenous infusion has been started, 1 hour after an intramuscular dose and 1 to 2 hours after an oral dose.

Reagents/Materials/Media

Mueller Hinton Broth (MHB) (100 mL)
Blood Agar (BA)
Sterile 13 x 100 mm glass tubes
Sterile 1.0 mL pipettes
Sterile yellow pipette tips
Test tube racks
Pipetter
Precision pipette to deliver 20 µL

IV. Procedure

p. Processing of Specimens
Upon arrival in the laboratory, centrifuge the blood and aseptically transfer the serum into a sterile tube.

p. Preparation of bacterial suspension:
Inoculate several colonies of a pure culture of the patient's organism (overnight sub-culture) into 5 mL MHB. Incubate on a shaker at 36°C for a minimum of 3 hours or until it achieves turbidity greater than the 0.5 McFarland standard (approximately 1.3 x 10^8 CFU/mL).

p. Serum dilution:
a. Place 12 sterile test tubes in a rack for each serum sample to be diluted.
b. Number the tubes 1 to 12.
c. Aseptically pipette 1.0 mL of patient's serum into tubes 1 and 2.
d. Aseptically pipette 1.0 mL of MHB into tubes 2 -12.
e. With a new 1.0 mL sterile pipette transfer 1.0 mL of serum from tube 2 to tube 3. Mix well.
f. Serially dilute the serum by sequentially transferring 1.0 mL of the mixture through to tube 10. Diskard 1.0 mL of the mixture from tube 10. No serum is to be added to tube 11 (positive inoculum control) or to tube 12 (broth sterility control). The final dilution of serum in tube 10 is 1:512 and final volume in all tubes should be 1.0 mL.

p. Inoculating Broth
a. Using the Vitek colourimeter, dilute the bacterial suspension to the turbidity of a 0.5 McFarland standard using MHB.
b. Prepare a 1:4 dilution of the standardized inoculum by adding 1.0 mL of inoculum to 3.0 mL MHB. Mix well.
c. Using a precision pipette, dispense 20 µL (0.02 mL) of diluted inoculum into tubes 1 through 11. Inserting the pipette tip well under the surface of the antimicrobial containing serum broth mixture. AVOID ANY CONTACT BETWEEN THE TIP AND THE WALLS OF THE TUBE – to prevent transfer of organisms to the inside of tube above the meniscus. Mix by flushing 2 or 3 times without creating air bubbles or splashing. Use a new tip for each tube.
d. Incubate all tubes at 37°C for 20 hours in a CO_2-free incubator.
e. From the 1:4 dilution of the standardized inoculum, dilute 1:250 in MHB (0.1 mL in 24.9 mL MHB) to achieve an inoculum of 10^5 CFU/mL.
f. Perform a colony count to confirm the bacterial count in the final inoculum. Transfer 0.001 mL of diluted inoculum to BA using a urine loop and distribute evenly on the surface of a BA plate.
g. Incubate the BA plate overnight at 35°C.

<u>Determination of serum bacteriostatic titres</u>

p. After incubation, tube 12 (broth sterility control) should be clear while tube 11 (positive inoculum control) should be turbid.

p. Record the colony count. The colony count plate should have 75-150 colonies. If the colony count is <75 or >150 consult the charge technologist before reading the tubes.

p. The highest dilution of serum that completely inhibits visible growth represents the bacteriostatic titre.

Determination of serum bactericidal titre

p. Vortex all tubes without visible growth for 15 seconds.

p. Use a urine loop to subculture all of the clear tubes onto ¼ BA. Incubate at 37°C for 18 hours.

p. After incubation, read the plates and record the colony count.

p. The first dilution showing 99.9% killing activity (ie. No growth on sub-culture) is reported as the serum bactericidal titre.

V. Reporting Results

Telephone all results when available. Report as follows and give a copy of the report to the microbiologist:

Pre-dose serum bacteriostatic titre –
Pre-dose serum bactericidal titre –

Post-dose serum bacteriostatic titre –
Post-dose serum bactericidal titre –

VI. Reference

National Committee for Clinical Laboratory Standards. Methodology for the Serum Bactericidal Test, CLSI Document M21-P,Vol. 7, No. 1, 1987.

IX – Broth Macrodilution and Agar Dilution

p. **Introduction**

These tests are not routinely done and will only be performed following consultation with a microbiologist. Refer to the CLSI standard M7-A9, 2012 for methodology.

X – Broth Microdilution MIC

I. Introduction

Dilution susceptibility testing methods are used to determine the minimal concentration of an antimicrobial agent required to inhibit or kill a microorganism. Antimicrobial agents are usually tested at $\log_2$ (twofold) serial dilutions, and the lowest concentration that inhibits visible growth of an organism is regarded as the MIC. The concentration range used may vary with the drug, the organism tested, and the site of infection. The method and principles of the microdilution method is essentially the same as the macrodilution method except that the antimicrobial dilutions are in 0.1 mL volumes contained in wells of a microdilution tray (usually 96 well trays). Results obtained may be reported as the actual MIC or categorically as Susceptible (S), Intermediate (I), or Resistant I. Interpretive categories are published and up-dated regularly by CLSI.

II. Materials

Sterile saline
Transfer pipettes
Sterile distilled water
Vitek colorimeter
MIC microtitre panel
Inoculator (tray and lid)

III. Procedure

1. Remove the desired MIC panel from the -70^0C 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 order number.

3. Prepare a suspension of the test organism in sterile saline equivalent to a 0.5 McFarland standard using isolated colonies.

4. Transfer l.5 mL of the suspension into the inoculation tray and add approximately 30 mL of sterile distilled water.

5. Aseptically replace the transfer lid into the inoculating tray making sure no bubbles are under the prongs.

6. Lift the transfer lid and center it over the previously thawed MIC panel.

7. Align the left side (lettered) of the panel with the left side (lettered) of the inoculator.

8. Lower the transfer lid into the panel so the prongs enter all wells.

9. Remove transfer lid and cover the panel with a dummy MIC panel.5.

10. Record the date and time of panel set-up on the lid of the panel.

11. Using a transfer pipette, transfer 1 drop of suspension from the inoculation tray to a blood agar plate and streak for isolated colonies.

12. Pour the suspension into a sharps container containing hypochloride and 117ipro117t the inoculator into a sharps disposal box.

13. Incubate for in O_2 at 35^{o}C.

    For Staphylococcus and Enterococcus, read panel after 16-20 hours incubation. Reincubate the panel and read the oxacillin and vancomycin at 24 hours.

IV. Interpretation

The highest dilution of the antimicrobial that completely inhibits visible growth represents the minimum inhibitory concentration (MIC).

V. Quality Control

Panels are Quality Controlled with the appropriate ATCC control organisms.

For troubleshooting out-of range QC results, see CLSI Document M100-S23 Table 3F (page 158).

Reference

Clinical and Laboratory Standards Institute (CLSI) Document – Methods for Dilution Antimicrobial Susceptibility Testing M7-A9, 2012.

XI – Etest

I. Introduction

The Etest (also known as the Gradient Diffusion Method) is based on the same principle as the disk diffusion method. It is an *in vitro* method for quantitative antimicrobial susceptibility testing whereby a preformed antimicrobial gradient from a plastic-coated strip diffuses into an agar medium inoculated with the test organism. The MIC values are read directly from the scale on the top of the strip, typically at the point where the ellipse of organism growth inhibition intercepts the strip, but this may vary slightly between drug and organism types and should be checked.

II. Materials

Etest strips (AB BIODISK, bioMerieux, store at -20C and with desiccant when opened)
Brain Heart Infusion Agar with Casein (BBL BHIA or Oxoid BHIA plus Casein)
Mueller-Hinton Agar (MHA)
BBL-Mueller-Hinton Agar (BD BBL-MHA)
Mueller-Hinton Blood Agar (MHBA)
BBL-Mueller-Hinton Blood Agar (BD BBL-MHBA)
Haemophilus Test Media (HTM)
RPMI 1460 (contains L-glutamine) + 2% Glucose + MOPS + 1.5% Bacto Agar (Oxoid)
Trypticase Soy Broth (TSB) (3 mL)
Mueller-Hinton Broth (MHB) (3mL)
Sterile saline (3-5mL)
VITEK colourimeter
Sterile wooden sticks
Sterile swabs

III. Procedures

p. Allow Etest strips to come to room temperature before opening the container.

2. Use the Vitek colourimeter to prepare a suspension of the test organism in sterile saline unless otherwise specified. If there is not enough growth, inoculate the organism into TSB, and incubate at 35^{o}C for 2-4h or until it reaches sufficient turbidity to prepare the required McFarland standard. Use pure culture for testing ONLY

    a) For testing non-fastidious organisms to most antibiotic combinations, prepare a bacterial suspension equivalent to a 0.5 McFarland standard using isolated colonies.

    b) For very mucoid organisms, adjust the suspension to a 1.0 McFarland standard.

c) For detecting reduced susceptibility to glycopeptides in *Staphylococcus aureus* (VISA or hVISA) or acquired glycopeptide resistance in *Enterococcus* species (VRE) using the MacroEtest Method, prepare a bacterial suspension equivalent to a 2.0 McFarland standard using isolated colonies and inoculate to a BBL BHIA or Oxoid's BHI plus Casein Agar for testing both teicoplanin and vancomycin (For Etest Macromethod procedure for *S. aureus*, see EAS-003 Staphylococci, and for enterococci, see EAS 006 Enterococci from http://www.abbiodisk.com/bd_litt_eas.html).

d) For determining standard glycopeptide MIC by Etest in coagulase-negative staphylococci, use a 0.5 MacFarland equivalent suspension prepared in saline, plate to MHA and test vancomycin only (See EAS 003 Staphylococci from http://www.abbiodisk.com/bd_litt_eas.html).

e) For determining MIC by Etest to *Streptococcus pneumoniae* or *Haemophilus 120ipro120tib*, prepare in Mueller-Hinton broth a 0.5 MacFarland equivalent bacterial suspension (if non-mucoid) or a 1.0 MacFarland (if mucoid) using a blank tube of MHB to adjust the Vitek colourimeter instead of a blank saline. (See EAS 010 and CIS 004 for *Streptococcus pneumoniae*, and EAS 005 for *Heamophilus* spp. From or http://www.abbiodisk.com/bd_litt_eas.html). Plate suspension to the appropriate agar without delay to prevent loss of cell viability within the suspensions as this will negatively affect interpretation of results.

f) For determining MIC for yeasts by Etest (i.e. caspofungin) prepare organism suspensions in saline equivalent to a 0.5 MacFarland for *Candida* spp. And to a 1.0 MacFarland for *Cryptococcus* spp. For these organisms, ensure to "double dip" the swab when inoculating the RPMI: i.e. after inoculating the plate the first time, soak the swab again and repeat the process a second time (See EAS 006 and CIS 005 Media for Antifungal testing from http://www.abbiodisk.com/bd_litt_eas.html).

3. Use a sterile cotton swab to inoculate the organism onto an appropriate agar plate, streaking in 3 directions over the entire agar surface.

a) For non-fastidious, rapid-growing organisms (excluding those below) use MHA.

b) For VRE detection in *Enterococcus* and VISA/hVISA detection in *Staphylococcus aureus*, test both vancomycin AND teicoplanin, evenly applying the heavy suspension to BBL BHIA or Oxoid "BHIA with Casein" (See EAS 009 from http://www.abbiodisk.com/bd_litt_eas.html).

c) For testing daptomycin in *Staphylococcus aureus*, coagulase-negative staphylococci or enterococci, use BD BBL MHA (or confirm Ca++ concentration in another brand is between 25-40mg/L) (See CIS 014 from http://www.abbiodisk.com/bd_litt_eas.html).

d) For MIC to agents other than glycopeptides in *S. aureus* and enterococci, use MHA inoculated with 0.5 MacFarland equivalent bacterial suspensions.

e) For *Haemophilus* spp. Use HTM (See EAS 005 from http://www.abbiodisk.com/bd_litt_eas.html)

f) For *S. pneumoniae* or viridans streptococci use BD BBL-MHBA (See CIS 004 from http://www.abbiodisk.com/bd_litt_eas.html).

g) For organisms that do not grow on MHA without blood, use MHBA.

5. Use sterile forceps to apply the appropriate Etest strip, MIC scale facing upwards, onto the appropriate agar making sure no bubbles remain trapped under the strip.

   a) Apply the strip ONLY AFTER the suspension has been allowed to dry thoroughly for at least 15 minutes. If strips are applied when plates are still wet, a ridge of growth running up the base of the strip will occur that will make interpretation difficult – if this happens, this growth should always be ignored.

   b) A maximum of two Etest strips may be placed on a small plate and a maximum of six strips may be placed on a large plate. When placing multiple strips on one plate, always ensure that the expected elliptical diffusion zones of adjacent drugs are not close enough to overlap.

p. Incubate plates as follows:

p) Non-fastidious organisms (except those specified below) – O_2, 35°C x 18h

p) *Haemophilus* species – CO_2, 35°C x 18h

p) *Streptococcus pneumoniae* or viridans streptococci – CO_2, 35°C x 20-24h

p) For MacroEtest with vancomycin and teicoplanin against *Staphylococcus aureus* and *Enterococcus* spp. – O_2, 35°C x 24h and 48h

p) For *Candida* spp. (plates in plastic bag) O_2, 35°C x 24-48h (when testing *C. glabrata* and *C. tropicalis*, MIC must always be confirmed at 48h)

p) For *Cryptococcus* spp. (plates in plastic bag) O_2, 35°C x 48-72h

ETEST Procedure Summary:

Staphylococcus aureus set up all of the following if Vitek MIC =>2 mg/L OR Growth on Vancomycin Screen:

Antimicrobial	Inoculum	Media	Incubation	Reading
Vancomycin-macro + Teicoplanin-macro	2.0 McFarland Std.	BHI with Casein Agar (OXOID)	35°C in O_2	At 24 hours and 48 hours
Vancomycin + Teicoplanin	0.5 McFarland Std.	Mueller Hinton Agar (OXOID)	35°C in O_2	At 24 hours

Coagulase- negative-Staphylococcus or Enterococcus set up if Vitek vancomycin = I or R OR Growth on Vancomycin Screen:

Organism	Antimicrobial	Inoculum	Media	Incubation	Reading
Coagulase-negative-*Staphylococcus*	Vancomycin	0.5 McFarland Std.	Mueller Hinton Agar (OXOID)	35°C in O_2	At 24 hours
Enterococcus species	Vancomycin + Teicoplanin	2.0 McFarland Std.	BHI with Casein Agar (OXOID)	35°C in O_2	At 24 hours and 48 hours

Other organisms:

Organism	Inoculum	Media	Incubation	Reading
Non-fastidious organisms	0.5 McFarland Std.	Mueller Hinton Agar (OXOID) (Mueller Hinton with Blood if organism fails to grow)	35°C in O_2	At 18 hours
Mucoid Organisms	1.0 McFarland Std.	Media appropriate for the organism	35°C in O_2	At 18 hours
Haemophilus species	0.5 McFarland Std.	Hemophilus Test Medium (HTM)	35°C in CO_2	At 18 hours
Streptococcus pneumoniae or viridans streptococci	0.5 McFarland Std.	BD BBL-MHBA	35°C in CO_2	At 20 to 24 hours
Candida species	0.5 McFarland Std.	RPMI 1460 using "double dip" yeast inoculation technique	35°C in O_2 in plastic bag	At 24 to 48 hours
Cryptococcus species	1.0 McFarland Std.	RPMI 1460 using "double dip" yeast inoculation technique	35°C in O_2 in plastic bag	At 48 to 72 hours

Interpretation

After the appropriate incubation, read the MIC value as per AB BIODISK instructions. Note: reading and interpretations are often drug-organism specific. Therefore check specific instructions first for each drug-organism combination.
ETEST Reading Guide
Antimicrobial effect: Cidal or Static
Etest Use – Reading powerpoint.pdf

a. For bactericidal drugs, the MIC is typically read at the point of complete inhibition where the zone edge intersects the Etest strip, whereas for bacteriostatic agents, the MIC is read at 80% inhibition when trailing is seen (See CIS 006 from ETEST Reading Guide for mode of antibiotic action to determine which are read at complete versus 80% inhibition).

b. Since Etest comprises a continuous gradient, MIC values in between two-fold dilutions may be obtained. In most cases, these values may be rounded up to the next two-fold dilution before interpretation but DO NOT round up for ALL organism types without checking as exceptions exist (i.e. for vancomycin and teicoplanin with possible VISA) (See EAS 003 Staphylococci and CIS 002 Endpoints for Glycopeptides from ETEST Reading Guide).

c. **Polymyxin B** is prone to hazy endpoints, so to avoid these, stay on the lighter side of the 0.5MacFarland standard when preparing the bacterial suspension, and inoculate the MHA using a swab that has been squeezed of all excess fluid. Do not exceed the recommended 18h incubation time. When reading the MIC, read at the point of complete inhibition, and if there is a dip, read at the base of the dip. (See Etest Polymyxin B Reading CIS 007 and CIS 012 from ETEST Reading Guide).

d. For testing **vancomycin and teicoplanin** against *Staphylococcus aureus* use the Etest macromethod procedure to detect VISA or hVISA (See EAS 003 Staphylococci and CIS 002 Endpoints for Glycopeptides from ETEST Reading Guide):

e. A VISA is defined as a *S. aureus* (MRSA or MSSA) with MIC to vancomycin AND teicoplanin of 8mg/L or greater, *OR* a teicoplanin MIC of 12mg/L or greater with a vancomycin MIC of <8mg/L (hence BOTH drugs MUST be tested simultaneously)

f. To determine endpoints, it may be necessary to use a magnifying glass, oblique light and to tilt the plate
g. Read at complete inhibition, looking for hazes, micro-colonies and isolated colonies within the zone of inhibition

h. DO NOT ROUND UP the MIC to the next two-fold value as with other organisms, especially if the *S. aureus* zone ellipse intersects the strip at 6mg/L, as this may result in a major error. Results of <8mg/L are interpreted as susceptible for vancomycin unless teicoplanin is 12mg/L or greater.

6. For **daptomycin** testing (See CIS 014 from ETEST Reading Guide , and Etest Daptomycin communiqué), it is necessary to:
   a) only use media that has a Ca++ concentration of 25-40mg/L (BD BBL MHA for staphylococci or enterococci, BD BBL MHBA for *S. pneumoniae*; awaiting information regarding the Ca++ concentration of Oxoid MHA and MHBA – apparently they corrected the CA++ a couple of years ago to comply with dapto testing requirements)
   b) only use bacterial suspensions that do not exceed the mid-range mark on the VITEK turbidometer for the 0.5 MacFarland Standard, as heavier suspensions may result in falsely elevated MIC
   c) squeeze out any excess liquid from the swab prior to inoculating the plate
   d) ensure that the plate dries for ~10-15min before applying the daptomycin Etest strip, as wet plates may result in growth up the side of the Etest strip, making results difficult to interpret
   e) confirm any daptomycin-resistant isolate by broth microdilution MIC testing

IV. Reporting

Report MIC values rounded up to the next two-fold dilution where indicated. DO NOT round up when reporting of macro-etest vancomycin and teicoplanin for *S. aureus*.

Reporting vancomycin and teicoplanin for *Staphylococcus aureus*:

	Vancomycin	**Teicoplanin**	**Report**
Macro-etest	<8 mg/L	<12 mg/L	Negative
Macro-etest	≥8 mg/L	≥8 mg/L	Positive
Macro-etest	<8 mg/L	≥12 mg/L	Positive

Reporting vancomycin and teicoplanin for *Enterococcus*:

	Vancomycin	**Teicoplanin**	**Phenotype**	**Species**
Macro-etest	≥32 mg/L I	≥16 mg/L (I – R)	vanA	*E. faecalis*, *E. faecium*
Macro-etest	≥8 – 256 mg/L (I – R)	≤4 mg/L (S)	vanB	*E. faecalis*, *E. faecium*
Macro-etest	4 – 16 mg/L (S – I)	≤4 mg/L (S)	vanC1	*E. gallinarum*
Macro-etest	4 – 16 mg/L (S – I)	≤4 mg/L (S)	vanC2	*E. casseliflavus*, *E.flavescens*
Macro-etest	64 mg/L I	≤4 mg/L (S)	vanD	*E. faecium*
Macro-etest	16 mg/L (I)	≤4 mg/L (S)	vanE	*E. faecalis*

V. Quality Control

p. The following four e-test strips (penicillin, ceftazidime, ceftriaxone and cefotaxime) are tested weekly with *S. aureus* ATCC 29213. The organism is sub-cultured from the TSA slant to BA the day before setting up the QC.

Expected Results*:

	MIC
Penicillin	0.25-2.0 mg/L
Ceftazidime	4.0-16.0 mg/L
Ceftriaxone	1.0-8.0 mg/L
Cefotaxime	1.0-4.0 mg/L

* As per CLSI document M100-S23, 2013, Table 3.

VI. Reference

AB BIODISK, Sohna, Sweden, Etest package insert.
http://www.ilexmedical.com/files/ETEST_RG.pdf
https://kaldur.landspitali.is/gaeda/gnhsykla.nsf/5e27f2e5a88c898e00256500003c98c2/2030bf44cbec6e0e00256f23003f2169/$FILE/Fine-tuning%20Etest%20Use%20-%20Reading.pdf

XII – Vancomycin & High Level Aminoglycoside Testing for *Enterococcus*

I. Introduction

Synergy between ampicillin, pencillin or vancomycin and an aminoglycoside for *Enterococcus* species can be predicted by high level aminoglycoside (HLA – gentamicin and streptomycin) screening test. Vancomycin resistance of *Enterococcus* species can be detected by BHI vancomycin agar screen plate containing 6mg/L of vancomycin.

II. Materials

Control plate (Brain Heart Infusion Agar)
Entero HLA and Vancomycin Screen plates
VITEK colorimeter
Sterile saline
Sterile swab

III. Procedure

1. Using the VITEK colourimeter, prepare a 0.5 McFarland suspension in sterile saline (inoculum from VITEK can be used).

2. Retrieve BHI, Vanco, and Hi-level Gent/Strept plates from fridge and their corresponding registration label (Lot number and expiry date) Affix the appropriate label to the reverse side of the worksheet. Write the date of testing on the sheet QUAD Screen Recording Sheet for *Enterococcus*

3. Using a sterile swab, spot inoculate the suspension onto each of the test and control plates (use grid TEMPLATE).

4. After the inocula have dried, incubate the plate at 35^{o}C for up to 48 hours.

IV. Interpretation

Check the control plate for adequate growth. Then check the drug plates for absence or presence of growth; any growth is considered significant. Read plates at 24 hours and record results. If there is no growth on the streptomycin plate, re-incubate plate for an additional 24 hours.

Growth on Vancomycin Screen plate must be comfirmed by checking the purity of the control plate, vancomysin E-test and repeat 129ipro129tibi screen testing.

V. Quality Control

Control strains are tested with each plate.

Controal Strains	Expected results of each quadrant			
	C	**G**	**S**	**V**
E. faecalis (ATCC 49532)	+	+	-	-
E. gallinarum (ATCC 49573)	+	-	-	+
E. faecalis (ATCC 49533)	+	-	+	-

C = Growth Control; G = Gentamicin; S = Streptomycin; V = Vancomycin

VI. Reporting Results

Blood cultures when ampicilline and / or vancomycin is susceptible:

- If high level gentamicin is **susceptible** (regardless of streptomycin result) report as: "**Serious enterococcal infections may require an aminoglycoside for synergy. Please contact the Medical Microbiologist for treatment advice".**

- If high level gentamicin is **resistant** (regardless of streptomycin result) report as: **"This organism is high level aminoglycoside resistant. Please contact the Medical Microbiologist for treatment advice".**

- Record the streptomycin result in the LIS. Report result only upon request.

Urines and other sites:

- Do not report HLA.
- Report Vancomycin result as per specimen type specific reporting tables.

Reference

PML Technical Manual data sheet No. 323, Nov. 1993.

QUAD Screen Recording Sheet for *Enterococcus* Page 1

Enter Lot numbers for BHI, HLA, VA on back page of this worksheet.

Date: **Set up by:**

18h VA + HLA (if done) read by:

24h VA + HLA (if done) read by:

48h HLA (if done) read by:

No. / Bench	ATCC Control Or Lab No.	VANC 18 h	VANC 24h	BHI 24h	GENTA 24h	STREPTO 24h	STREPTO 48h	No. / Bench	Lab No.	VANC 18 h	VANC 24h	BHI 24h	GENTA 24h	STREPTO 24h	STREPTO 48h
1	*E.faecalis* ATCC 49532	S			R	S		13							
2	*E.gallinarum* ATCC 49573	R			S	S		14							
3	*E.faecalis* ATCC 49533	S			S	R									
4								16							
5								17							
6								18							
7								19							
8								20							
9								21							
10								22							
11								23							
12								24							

QUAD Screen Recording Sheet for *Enterococcus* Page 2

Lot number and expiry date labels:

BHI_____

HLA _____

VA _____

XIII – Vancomycin-Intermediate Staphylococcus aureus Screen

I. Introduction

This spot agar dilution method uses a single concentration of vancomycin at 4mg/L incorporated into BBL Brain Heart Infusion agar (BBL BHIA) to screen for *Staphylococcus aureus* with reduced susceptibility to glycopeptides. Strains capable of growing on this medium are known as VISA or hVISA as their vancomycin and/or teicoplanin MIC are typically intermediate. This type of low-level resistance may be 133ipro133tibility133 present and so is difficult to detect, but it is important to notice as such subpopulations are implicated in vancomycin treatment failures. BBL BHI-vancomycin screen agar is commercially available from Oxoid as VISA ISOLATION AGAR (MP0243), while the control agar without vancomycin is available from Oxoid as BHIA with Casein. BBL BHIA (MP0244) is far ricore enriched than other formulations due to additional casein, and because of this, it is important not to prepare this screen agar with any other classic BHIA brand.

II. Materials

Screen plate (BD BBL BHIA with 4 mg/L vancomycin or Oxoid's VISA ISOLATION AGAR)
Control plate (BD BBL BHIA or Oxoid's BHIA with Casein)
VITEK colourimeter
24-position acetate grid for spot plate
Sterile saline
Sterile cotton swabs

III. Procedure

1. Using the VITEK colourimeter, prepare a suspension equivalent to a 0.5 McFarland standard in sterile saline (use the VITEK inoculum if already made).

2. Prepare worksheet as per OXACILLIN AND VANCOMYCIN SCREEN RECORDING SHEET FOR *S. aureus*

3. Using a sterile swab, spot inoculate the suspension onto the predetermined position on each of the screen and control agars, noting that no more than 24 isolates including controls should be spotted to each plate (use grid TEMPLATE).

4. Similarly, spot inoculate the following quality control strains, in order, to the first four positions on each agar: *E. gallinarum* ATCC 49573, *S. aureus* ATCC 43300 (MRSA), *S. aureus* LPTP 8610 (MRSA), and *S. aureus* ATCC 29213 (MSSA)

5. After the inocula have dried, incubate the plate at 35°C in O_2.

6. Perform a preliminary read for any growth after 18 hours incubation, and read again after both 24 hours and 48 hours incubation.

7. All resistant isolates on the screen plate must be checked for purity (e.g. Gram stain, tube coagulase or slide agglutination and sub-culture).

8. If the growth is:

   i. Confluent and pure, Pastorex-positive, Gram positive cocci in clusters, set up a confirmatory MacroEtest by preparing the suspension equivalent to a 2 MacFarland standard **directly from the VISA spot plate**. Inoculate to Oxoid's BHIA with Casein (BBL BHIA), allow plate to dry for 15min, apply both vancomycin and teicoplanin Etest strips, and read after both 24h and 48h incubation at 35°C for reduced suspectibility (Refer to APPENDIX X – Etest).

   ii. If the growth is spotty but pure, attempt to prepare suspension for a macroEtest and repeat screen plates **directly from the VISA spot plate**. If insufficient, use the growth on the control agar to supplement the inoculum.

   iii. If there is only a single or few colonies, subculture to a 5% sheep blood agar and perform confirmatory testing the following day. (Refer to APPENDIX X – Etest).

IV. Interpretation

1. Plates may be read initially at 18h but MUST BE REINCUBATED and read again at 24h and again at 48h, or the results are invalid.

2. At the first reading time (18h), check the control plate for adequate growth on each inoculated spot, and record these on the sheet documenting both growth and purity. Note: There must be confluent growth on the control plate for the test to be valid.

3. Then check the VISA ISOLATION plates to ensure the controls grew and/or were inhibited as appropriate (see QC table below).

4. Then check for absence or presence of growth for all test isolates; while any growth is considered significant, document confluent versus single colony growth on worksheet (see master copy below) and on the back of the specimen worksheet in the LIS.

5. Record 18h, 24h and 48h results on the worksheet. Final interpretations are made according to confirmatory testing (see above).

6. Report preliminary findings to infection control as a possible VISA if the growth on the spot plate is confluent and pure (even if colony sizes vary or growth is poor), and if the isolate is derived from blood or sterile sites.

7. However, if only a few colonies grow on the VISA screen agar, before reporting to Infection Control or Physicians, complete all confirmatory testing as there is an approximate breakthrough rate of single colonies using this method of 3%.

V. Quality Control

Control strains are tested on every plate.

Control Strains	BHIA+Casein Control plate	VISA ISOLATION AGAR
E. gallinarum ATCC 49573	**Growth**	**Growth**
S. aureus (MRSA) ATCC 43300	**Growth**	No growth

S. aureus (MRSA) LPTP 8610	**Growth**	No growth
S. aureus (MSSA) ATCC 29213	**Growth**	No growth

VI. Reference

p. **M. Willey**, N. Kreiswirth, A. Gelosia, C. Porter, T. Mazzulli , S. Pong-Porter, C. Larocque, K. Pike, B. Kreiswirth, N. Nelson, K. Wong, S. Poutanen, D. E. Low. **Screening for Vancomycin-Intermediate *Staphylococcus aureus* (VISA): Does Casein make a difference?** Abstract in proceedings of the 48th Annual ICAAC and 46th Annual IDSA Joint Meeting, Washington, DC, October 25-28, 2008

J Clin Microbiol. 2009 Jul;47(7):2013-7. Epub 2009 May 6 Accuracy of commercial and reference susceptibility testing methods for detecting vancomycin-intermediate *Staphylococcus aureus*. Swenson JM, Anderson KF, Lonsway DR, Thompson A, McAllister SK, Limbago BM, Carey RB, Tenover FC, Patel JB.

XIV – Antimicrobial Abbreviations
Antimicrobial Disks

ANTIMICROBIAL	DISK (Manufacturer)	Concentration (μg)
Amikacin	AK (Oxoid)	30
Amoxacillin/Clavulanic Acid	AMC	30
Ampicillin	AMP (Oxoid)	10
Ampicillin/Sulbactam	AMS	20
Azithromycin		15
Aztreonam	ATM	30
Cefazolin	KZ (Oxoid)	30
Cefepime	FEP	30
Cefiderocol	FDC (Liofilchem)	30
Cefixime	CFM	5
Cefotaxime	CTX	30
Cefotetan	CTT (Gen. Diag.)	30
Cefoxitin	FOX (Oxoid)	30
Ceftazidime	CAZ (Oxoid)	30
Ceftazidime-avibactam	CZA (Liofilchem)	50
Ceftriaxone	CRO (Oxoid)	30
Cefuroxime	CXM	30
Cephalothin	KF	30
Cefpodoxime	CPD	10
Chloramphenicol	C	30
Ciprofloxacin	CIP (Oxoid)	5
Clarithromycin	CLR	15
Clindamycin	DA (Oxoid)	2
Colistin	CT	10
Doxycyline	DO	30
Ertapenem	ETP (Oxoid)	10
Erythromycin	E (Oxoid)	15
Fosfomycin	FOT (Oxoid)	200
Gentamicin	CN (Oxoid)	10
Imipenem	IPM (Difco)	10
Imipenem-Relebactam	I/R (Liofilchem)	35

ANTIMICROBIAL	DISK (Manufacturer)	Concentration (μg)
Levofloxacin	LVX	5
Linezolid	LZD	30
Meropenem	MEM	10
Metronidazole	MTZ (Oxoid)	5
Minocycline	MH	30
Mupirocin	MUP	5
Nalidixic Acid	NA	30
Nitrofurantoin	F (Oxoid)	300
Norfloxacin	NOR (BBL or Difco)	10
Novobiocin	NV	5
Oxacillin	OX (Oxoid)	1
Penicillin	P (Oxoid)	10
Piperacillin	PRL (Oxoid)	100
Pipercillin/Tazobactam	TZP	110
Polymyxin B	PB	
Quinupristin-Dalfopristin (Synercid)	QD	
Rifampin	RA	5
Teicoplanin		30
Tetracycline	TE (Oxoid)	30
Ticarcillin/Clavulanate (Timentin)	TIM (Oxoid)	85
Tobramycin	TOB (Oxoid)	10
Trimethoprim/Sulfamethoxazole	SXT	25
Vancomycin	VA (Oxoid)	30

e-test Strips

ANTIMICROBIAL	ABBREVIATION
Amoxicillin	AC
Amikacin	AK
Ampicillin	AM
Azithromycin	AZ
Cefotaxime	CT
Cefotetan	CN
Cefoxitin	FX
Ceftazidime	TZ
Ceftolozane-Tazobactam	C/T
Ceftobiprole	BPR
Ceftriaxone	TX
Cefuroxime	XM
Cephalothin	CE
Chloramphenicol	CL
Ciprofloxacin	CI
Clarithromycin	CH
Clindamycin	CM
Colistin	CO
Doxycycline	DC
Daptomycin	DPC
Ertapenem	ETP
Erythromycin	EM
Gatifloxacin	GA
Gentamicin (Low Level)	GM
High Level Gentamicin	GM
Imipenem	IP
Levofloxacin	LX
Linezolid	LZ
Meropenem	MP
Metronidazole	MZ
Minocycline	MC
Moxifloxacin	MX
Mupirocin	MU
Meropenem-Vaborbacam	M/V
Oxacillin	OX
Penicillin	PG

ANTIMICROBIAL	ABBREVIATION
Piperacillin	PP
Piperacillin/Tazobactam	PTC
Polymyxin B	PO
Quinupristin-Dalfopristin (Synercid)	QDA
Rifampin	RI
Streptomycin (High Level)	SM
Teicoplanin	TP
Tetracycline	TC
Ticarcillin/Clavulanate	TLc
Tigecycline	TGC
Tobramycin	TM
Trimethoprim/sulfamethoxazole	TS
Vancomycin	VA

LIS (Soft Computer Corporation)

ANTIMICROBIAL	ABBREVIATION
Amikacin	an
Amoxicillin	amx
Amoxicillin / Clavalanic Acid	amc
Ampicillin	am
Ampicillin / Sulbactam	ama
Azithromycin	azi
Aztreonam	azm
βeta-lactamase	blac
β-lactamase	beta
Carbenicillin	cb
Cefaclor	ccl
Cefamandole	cm
Cefazolin	cz
Cefepime	cpo
Cefixime	cfm
Cefotaxime	tax
Cefotetan	cte
Cefoxitin	fox
Cefpodoxime (Vitek panel)	cpd
Cefpodoxime (Kirby-Bauer panel)	cpod
Cefpodoxime / Clavulanic Acid	cpodc
Ceftazidime	taz
Ceftizoxime	zox
Ceftolozane-Tazobactam	cfttaz
Ceftobiprole	bpr
Ceftriaxone	ctr
Ceftriaxone-meningitis	ctrm
Ceftriaxone-non meningitis	ctrnm
Cefuroxime	roxh
Cefuroxime–Axetil	roxa
Cefuroxime-sodium	rox
Cephalothin	cf
Chloramphenicol	c
Ciprofloxacin	cip
Clarifloxacin	clar
Clarithromycin	cla
Clinafloxacin	cflox
Clindamycin	cc

ANTIMICROBIAL	ABBREVIATION
Cloxacillin	clx
Colistin	ct
Dalfopristin	dalfo
Doxycycline	dx
Daptomycin	dapto
D-zone	dzone
Ertapenem	etp
Erythromycin	e
ESBL Potentiation	esbinh
Everninomycin	ever
Gatifloxacin	gat
Gentamicin	gm
Gentamicin 2000	gm2000
Gentamicin 500	gm500
Imipenem	imi
Kanamycin	k
Levofloxacin	lev
Linezolid	linezo
Meropenem	mem
Meropenem Screen	mems
Meropenem 10 – Rosco Disk	mrp10
Meropenem + DPA – Rosco Disk	mrdp
Meropenem + DPA Potentiation	mrdpp
Meropenem + Boronic acid – Rosco Disk	mrbo
Meropenem + Boronic acid Potentiation	mrbop
Meropenem + Cloxacillin – Rosco Disk	mrclx
Meropenem + Cloxacillin Potentiation	mrclxp
Metronidazole	mtz
Mezlocillin	mz
Minocycline	mn
Moxifloxacin	mox
Mupirocin	mup
Nalidixic Acid	na
Netilmicin	net
Nitrofurantoin	fd
Norfloxacin	nor
Ofloxacin	ofx
Oxacillin	ox
Penicillin	peng
Penicillin-IV-meningitis	penm

ANTIMICROBIAL	ABBREVIATION
Penicillin-IV-non-meningitis	pennm
Penicillin-Oral	penspo
Piperacillin	pip
Piperacillin / Tazobactam	pzp
Polymyxin B	pb
Pristinamycin	pris
Quinupristin-Dalfopristin (Synercid)	qda
Ramoplanin	ramo
Rifampin	rif
Streptomycin	strep
Streptomycin 2000	st2000
Sulfisoxazole	sox
Synercid	syncd
Teicoplanin	tei
Temocillin (ROSCO Disk)	tem
Tetracycline	tet
Ticarcillin	tic
Ticarcillin/Clavulanic Acid	tcc
Tigecycline	tig
Tobramycin	tob
Trimethoprim	tmp
Trimethoprim/sulfamethoxazole	sxt
Vancomycin	va

XV – Carbapenemase Testing with ROSCO Diagnostica Tablets

I. Materials

Vitek gram negative susceptibility card with ertapenem
Mueller-Hinton (MH) agar
10 mg meropenem disk (OXOID)
Rosco Diagnostica KPC + MBL Confirm ID kit tablets:

- Meropenem (MRP10)
- Meropenem+Dipicolonic acid (MR+DP)
- Meropenem+Boronic acid (MR+BO)
- Meropenem+Cloxacillin (MR+CL)
- Temocillin (TEM)

II. Procedure

For Meropenem Screen disk ≤25mm or Vitek Meropenem MIC ≥0.5mg/L & *β*CARBA (BCARB) = NEGATIVE
OR
For Meropenem Screen disk ≤25mm or Vitek Meropenem MIC ≥0.5mg/L & *β*CARBA (BCARB) = POSITIVE, CARB-R Cepheid PCR = NEGATIVE

1. Set up Rosco Diagnostica KPC + MBL with Temocillin test:

- Using the Vitek colorimeter, prepare a suspension of the test organism in sterile saline equivalent to a 0.5 McFarland standard.
- Using a sterile cotton swab, inoculate the organism onto a 150 mm (large) MH agar plate. Dispense tablets into a petri dish and use forceps to apply the 5 Rosco tablets (MRP10, MR+DP, MR+BO, MR+CX & TEM) onto the agar. Place the tablets at least 30 mm apart from each other.
- Incubate plate in O_2 at 35^{o}C x 18 hours.
- In the LIS, order Breakpoint Panel "kpcros" for drugs "mrp10", "mrdp", "mrdpp", "mrbo", "mrbop", "mrcl", "mrclp" "tem".

- Set up routine Vitek susceptibility/kbesbl as appropriate.

2. Interpretation of Rosco KPC+MBL Confirm Kit tablets:

Note: *β*CARBA=pos, CARB-R cases, record but do not report (for research purposes only)

- Record the zone size of all the tablets after incubation.
- Compare the zone size of the MRP10 tablet against the zone sizes of MRDP, MRBO and MRCL. If there is ≥5 mm difference in zone size, record "Y" for the potentiation of the drug. If there is <5 mm difference in zone size, record as "N" for the potentiation of the drug.
- Mero & Cloxacillin (MRCL) to be reported and potentiation compared to MRP10 >=5 mm to be documented in LIS.

- Measure temocillin zone size
- Refer to the table below for interpretation

Meropenem breakpoint:

By Meropenem Screen Disk (MEMS)	By ROSCO Tablet (MRP10)	Intepretation
>25 mm	>26 mm	**S**
≤25 mm	≤26 mm	**R**

Temocillin breakpoint:

By ROSCO Tablet (TEM)	Intepretation
>11 mm	**S**
≤11 mm	**R**

Rosco KPC+MBL Confirm Kit Interpretation:

	MRDP Potentiation	MRBO Potentiation	Temocillin-R	Other
Definition	MRP10 vs MRDP ≥5mm	MRP10 vs MRBO ≥5mm and MRP10 vs. MRCL <5mm	Temocillin-R (≤11 mm)	No potentiation and Temocillin-S (>11 mm)
Interpretation	**Class B carbapenemase** (metallo-β-lactamase) (e.g. NDM, VIM, IMP1)	**Class A carbapenemase** (e.g. KPC, NMC, IMI, SME, GES)	**Class D carbapenemase** (e.g. OXA48, OXA181, OXA232, OXA244) *or* **Class B carbapenemase** (metallo-β-lactamase) (e.g. NDM, VIM, IMP1)	No carbapenemase
Reporting	send to NML for PCR confirmation; order "kpcrcon" panel; see **Reporting** section for reporting phrase			**No CRE**

NML send out information (send out ASAP – Cannot send on Friday)
Attention David Boyd
Nosocomial Infect. Canadian Sciences Centre for Human and Animal Safety.
National Microbiology Laboratory, 1015 Arlington St. Winnipeg, MB Canada, R3E 3R2.

3. When PCR results are returned from NML, enter as drug kpcr=Y (if positive) or kpcr=N (if negative) with the appropriate result comments (see **Reporting** section) and call the Infection Control Practioner with the results.
   Notes:
   All screen positive isolates to be frozen.
   All screen positive to be called to infection control
   All interim and final updated results to be called to infection control.

III. Reporting

See Carbapenemase Testing Reporting

If the isolate is to be reported as ESBL, report with ISOLATE COMMENT code **\KPCN**: see Carbapenemase Testing Reporting

If the isolate is not generally reported (e.g. *Enterobacter* in ESBL screens),

a. Suppress the isolate.
b. Report at the TEST Window with TEST COMMENT code **}KPCN** - see Carbapenemase Testing Reporting

Notify as per Isolate Notification and Freezing Table QPCMI15003

IV. Reference

Rosco Diagnostica KPC + MBL Confirm Kit package insert. applicationsheet – KPC and MBL.pdf

XVI – *β*CARBA Test

p. **Introduction**

Any species within the family *Enterobacterales* may acquire genes encoding enzymes that hydrolyze carbapenem antimicrobial agents such as ertapenem, imipenem, meropenem and doripenem, and these enzymes are referred to as carbapenemases. The **Bio-Rad *β*CARBA** test provides a rapid, qualitative colorimetric procedure for detecting production of carbapenemases. It has been shown to be highly sensitive and specific, detecting all common genotypes (i.e. IMI1, KPC, NDM, OXA48-like, VIM, SME). It is performed in a micro-tube directly from colonies grown on Chromogenic agar, 5% Sheep Blood or Mueller-Hinton agar, preferably taken from around a resistant meropenem disc screen test. The test detects carbapenemase hydrolytic activity as the chromogenic carbapenem substrate changes colour from yellow (negative) to orange or red or purple (positive) within 30 minutes.

p. **Reagents**

Bio-Rad *β*CARBA Kit contains 3 reagent vials: R1 (diluent), R2 (dehydrated chromogenic substrate) and R3 (solvent for R2). The entire contents of R3 (1.1mL) is transferred to reconstitute R2 when a new kit is opened.

p. **Materials**

Required but not provided
Sterile 1µL green plastic loops
Extra sterile micro-tubes to supplement those provided
Rack with appropriate sized holes
Water bath set at 37^{o}C with thermometer
Timer

p. **Procedure**

1. The *β*CARBA test is to be done only on isolates grown preferably on Mueller-Hinton agar with meropenem disc screen resistant (inhibition zone $\leq$25mm) or on 5% Sheep Blood agar or Brilliance UTI agar according to CRE testing flowcharts.

2. In both screen and clinical cases, the isolate to be tested must already have been identified by VITEK MS PLUS as to belong to the family *Enterobacterales.*

3. When opening a new kit, homogenize reagent R1 and R3 by vortexing briefly. Ensure the lyophilized reagent is in the bottom of vial R2 prior to reconstitution. Reconstitute lyophilized R2 with full contents of R3 (1.1mL) and discard the empty R3 vial. Do not use reconstituted R2 if the colour turns red.

4. Write the date 3 months from the first opening as the new expiry date on R1 and reconstituted R2 reagents.

5. To perform one test, add 30µl each of reagents R1 and R2 into a labeled 1.5ml micro-tube.

6. Inoculate the tube using a heavy 1uL loop full of bacteria from a 5% sheep blood agar plate (clinical specimens) or from a Mueller-Hinton Plus agar plate from the inner zone of a resistant meropenem disc screen. (**Note: DO NOT test from MacConkey-based agars**).

7. Using the loop, mix the tube thoroughly to ensure the organisms are smoothly suspended in the reagents (if possible, do not vortex as the volume may be reduced).

8. Place tube in an appropriate rack into a 37^{o}C water bath and set timer for 30 minutes.

9. A positive result may be recorded as soon as a colour-change to red occurs (as early as 2 min), but a negative test (yellow) should be observed for a full 30 minutes to ensure no delayed orange (weak) positive reactions are overlooked.

10. Do not incubate longer than 30 minutes, as by 45 minutes, it is possible for a rare false-positive reaction to occur.

V. Interpretation of results:

	Result	**Interpretation**
Colour change from Yellow to Orange or Red or Purple	**Positive**	**Presence** of carbapenemase
No colour change from Yellow	**Negative**	**Absence** of carbapenemase

VI. Reporting

See Carbapenemase Testing Reporting.

VII. Quality Control

Quality control testing will be done on receipt for each new shipment by the QC bench and by the bench on opening each new kit.
Positive controls: *Klebsiella pneumoniae* ATCC 1705

Negative controls: *Klebsiella pneumoniae* ATCC 1706 or *E. coli* ATCC 25922

VIII. References

ECCMID and AMMI-CACMID abstract references.

XVIII – *B*LACTA Test

p. **Introduction:**

The BioRad *B*LACTA provides a simple, rapid qualitative procedure for detecting 3rd generation cephalosporin resistance in *Enterobacterales* without intrinsic resistance (i.e. *Escherichia coli*, any *Klebsiella* spp. Or *Proteus mirabilis*). In these species, a colour-change from yellow to red or orange indicates the enzymatic hydrolysis of a chromogenic cephalosporin due the presence of acquired cephalosporinases (i.e. ESBL or plasmidic *ampC* β-lactamases). The test is designed to enable earlier reporting of resistance, if present, to all cephalosporins, and may be done as soon as MALDI-TOF identification from short-incubation (3-6h) blood or sterile fluid cultures of the above organisms has been completed.

p. **Reagents:**

1. Reagent 1 and 2

p. **Materials:**

Provided:

1. Micro-tubes

Required but not provided:

1. Sterile 1uL plastic loops
2. Rack
3. Timer

p. **Procedure:**

1. The BLACT test is to be done only on isolates from early subcultures from blood or sterile fluid specimens that are already identified by VITEK MS as *E. coli*, *Klebsiella* spp, or *P. mirabilis*.

2. On the back of the LIS work-card, pick media "BLACT" from the test keypad menu when the test is to be set up.

3. Only proceed to the KB menu after the test has been completed.

4. To perform one test, add one drop each of reagents R1 and R2 into a labeled micro-tube.

5. Inoculate the tube using a heavy loop full of bacteria isolated on 5% sheep blood agar

   (Note: **DO NOT inoculate test from MacConkey agar**).

6. Vortex the tube thoroughly to ensure the organisms are well mixed (i.e. a smooth suspension).

7. Disregard test time and colour interpretations on the insert on inner lid of box

8. Place the tube into a rack at room temperature and set timer for 30 min

9. While the result may be recorded as soon as a colour-change to red occurs (as early as 2 min), the test should be observed for red or orange by 30 min to ensure no delayed positive reactions are overlooked

10. In the LIS at the back of the workcard adjacent to "BLACT", document the time and colour

    of the reaction by picking from the keypad (i.e. **Red within 15min, Red between 15 – 30 min, Orange at 30 min, or Yellow at 30 min**)
    POSITIVE = Red within 15min, Red between 15 – 30 min, Orange at 30 min
    NEGATIVE = Yellow at 30 min

11. If the BLACTA is **POSITIVE** proceed to the KB menu on isolate field and select the BLACTA POSITIVE panel "**kbBLAC+**". This selection will:

    (a) Enable recording of the POSITIVE BLACTA test result (enter "2" for positive)

(b) For MSH newborn:D1-M13 or MSH Female 12-50yrs, generate a "**kbESBL**" panel that must be set up right away along with the VITEK 2 AST-N213 card
(c) reflex "R" for all to extended-spectrum penicillins, beta-lactam/beta-lactamase inhibitor combinations (e.g. piperacillin-tazobactam), and cephalosporin
(d) attach a comment in the isolate window as per reporting section

12. **Do not call when we have MALDI or BLACTA results**

13. Enable recording of the NEGATIVE BLACTA test result (enter "1" for negative)

If the BLACTA is **NEGATIVE** this result **must NOT be reported** if calling the organism identification. Rather set up VITEK 2 AST-N213 card as per manual and proceed to the isolate field to select the BLACTA negative panel from the KB menu "**kbBLAC-**"
This selection WILL record the BLACTA negative result but it will NOT generate any other tests or comments.

Interpretation of results:

Color	Interpretation of results
Yellow (includes pale yellow-orange)	**Negative**
Red (at any time within 30 minutes)	**Positive**
Orange (at 30 minutes ONLY**)**	**Positive**

IX. Report:

Positive *B*LACTA, report with isolate comment (\BLTA):

"~Presumptive resistance to extended-spectrum penicillins,
~beta-lactam/beta-lactamase inhibitor combinations
~**(**e.g. piperacillin-tazobactam), and cephalosporins
~has been detected.
~Confirmation and further susceptibilities to follow. **"**

If ESBL is confirmed, report with isolate comment (\ESBC):
"Resistance to extended-spectrum penicillins, beta-lactam, beta-lactamase inhibitor combinations (e.g. piperacillin-tazobactam), and cephalosporins has been confirmed."

If ESBL is NOT confirmed e.g. in *K. oxytoca*, report with isolate comment (\ESBN):
"The previously reported presumptive resistance to extended-spectrum penicillins, beta-lactam, beta-lactamase inhibitor combinations (e.g. piperacillin-tazobactam), and cephalosporins was NOT confirmed."

Negative *B*LACTA– DO NOT REPORT

Quality Control:

Quality control testing will be done on receipt for each new shipment and weekly by the QC bench.

1. Positive control:
   Escherichia coli ATCC 51446

2. Negative control:
   Escherchia coli ATCC 35218 (type TEM-1 *B*-lactamase producing strain)

VIII. References:

1. B. M. Willey, X. Trimi, P. Lo, S. M. Poutanen. Pilot Prospective Evaluation of the *B*LACTA Test for Predicting 3rd Generation Cephalosporin Resistance in Shot-Incubation Blood Culture Isolates of *Escherichia coli*, *Klebsiella pneumoniae*, *Klebsiella oxytoca* and *Proteus mirabilis* Poster eP321 24th ECCMID Barcelona, Spain, 2014

2. B. M. Willey, Colorimetric Detection of 3rd Generation Cephalosporin Resistance in *Enterobacterales*: A Retrospective Evaluation of the Rapid Bio-Rad *B*LACTA Test. Poster eP333 24th ECCMID Barcelona, Spain, 2014.

APPENDIX E. Annual CLSI Updates Implementation

I. Introduction

This procedure outlines the process for reviewing and implementing annual updates from the Clinical and Laboratory Standards Institute (CLSI) within the laboratory. The purpose is to ensure accurate, up-to-date practices for antimicrobial susceptibility testing and reporting.

II. Procedure

1. Microbiologists, Charges, and the Laboratory Information System (LIS) Team use the CLSI change implementation checklist to review the relevant updates and plan the updates for:
    - LIS
    - Test and Reporting SOPs
    - Validation of instruments if needed (e.g., Vitek, Gradient Strips, Sensititre)
    - Dosing comments
    - Other comments
2. Microbiologists review European Committee on Antimicrobial Susceptibility Testing (EUCAST) updates to determine if LIS comments require changes.
3. Microbiologists communicate with the Charges and LIS team regarding the feasibility and timing of required changes.
4. The CLSI update plan is reviewed and discussed during the Microbiologists Meeting to get input and approval.
5. Microbiologists communicate the final update plan with the Charges and LIS team.
6. Microbiologist Antimicrobial Stewardship Programs (ASP) liaison notifies the ASP of the planned changes.
7. Microbiologists draft and distribute a Medical Staff Bulletin (MSB) summarizing the updates.
8. The laboratory aims to implement updates ideally within one month of notification, depending on the complexity and scope of changes.

III. Transition Period Guidelines

During the transition period when updated annual CLSI changes have been announced but not yet implemented in the laboratory, the following guidelines must be followed:

- Do not issue corrected reports during the transition period.
- Contact a microbiologist to review the clinical report.
- Microbiologists liaise with the clinical team to discuss the nuances of the updated guidelines and the effect on the interpretation of their patient's result
- Microbiologists issue a relevant comment to the report as deemed appropriate.

IV. References

1. Clinical and Laboratory Standards Institute (CLSI) guidelines
2. EUCAST (European Committee on Antimicrobial Susceptibility Testing) updates
3. Laboratory Information System (LIS) documentation

Vitek instructions:

Vitek Manual

2014.07.09 Vitek AES breakpoint changes:
..\..\Audits\Vitek AES Breakpoint Manual Changes\2014.07.09_2014 CLSI.pdf

TREK Sensititre instructions:

Trek Sensititer Manual

Record of Edited Revisions

Manual Section Name: ANTIMICROBIAL SUSCEPTIBILITY TESTING MANUAL

Page Number / Item	Date of Revision	Signature of Approval
Annual Review	May 2, 2001	Dr. T. Mazzulli
Annual Review	May 2, 2002	Dr. T. Mazzulli
Annual Review	May 12, 2003	Dr. T. Mazzulli
Reporting – Blood and Sterile Fluids – *S. pneumo* and viridans strep, report Ceftriaxone for UHN corrected	March 5, 2004	Dr. T. Mazzulli
Oxacillin Screen Recording Chart	April 2, 2004	Dr. T. Mazzulli
QUAD Screen Recording Chart	April 2, 2004	Dr. T. Mazzulli
Annual Review	May 26, 2004	Dr. T. Mazzulli
Add – Vancomycin Screen for *Staphylococcus* Page 3, 29	June 7, 2004	Dr. T. Mazzulli
Replace Oxacillin Screen Recording Chart with Oxa and Vanc Screen Recording Chart for *S. aureus*	June 7, 2004	Dr. T. Mazzulli
Update QUAD Screen Recording Chart	June 7, 2004	Dr. T. Mazzulli
Cefepime for PMH patients only Pages 2, 29-35	July 9, 2004	Dr. T. Mazzulli
Moxifloxcin reporting for UHN Pages 32, 35	July 9, 2004	Dr. T. Mazzulli
Group B strep testing and reporting Pages 3, 29-35	July 9, 2004	Dr. T. Mazzulli
Remove Vancomycin Screen for CNST Page 3, 39	October 12, 2004	Dr. T. Mazzulli
Reinstate Vancomycin Screen for CNST Page 3, 39	November 1, 2004	Dr. T. Mazzulli
Testing for GBS if requested Page 3, 29-35	November 9, 2004	Dr. T. Mazzulli
Nalidixic Acid for Salmonella for blood and sterile sites Page 2, 35	November 9, 2004	Dr. T. Mazzulli
Streptococcus milleri group testing Page 4, 29-35	November 9, 2004	Dr. T. Mazzulli
Positive Oxacillin Screen – set up DENKA, report if MRSA base on DENKA result Page 12	November 9, 2004	Dr. T. Mazzulli
Annual Review	April 21, 2005	Dr. T. Mazzulli
Page 2 Criteria for testing – include Aeromonas, Plesiomonas as not tested.	April 21, 2005	Dr. T. Mazzulli
Reporting – MRSA and VRE from IC screen test – do not report any susceptibility result.	April 21, 2005	Dr. T. Mazzulli
Table of Contents arrangement: - Add criteria for repeat testing table - "reporting" move up - change methodology to "Appendix" - Add Appendix – list of drug related LIS canned comments	November 21, 2005	Dr. T. Mazzulli
Criteria for repeating testing table as per CLSI M100-S15 Table 8	November 21, 2005	Dr. T. Mazzulli
Change all methodology pages to Appendix	November 21, 2005	Dr. T. Mazzulli
List of drug related LIS canned comments – Appendix XII	November 21, 2005	Dr. T. Mazzulli
Page 3 – isolates refer sensi back change to 1 day for blood cultures and sterile sites; 3 days for all other sites	November 21, 2005	Dr. T. Mazzulli
Page 3 – canned message change when refer back	November 21, 2005	Dr. T. Mazzulli
Page 3 – pod-R e coli, kleb and proteus add kb-esbl	November 21, 2005	Dr. T. Mazzulli
Page 3 – *S. maltophilia* – KB for sxt and levo	November 21, 2005	Dr. T. Mazzulli

Page Number / Item	Date of Revision	Signature of Approval
Page 3 – add *B. cepacia* – KB for sxt, Ceftazidime, meropenem	November 21, 2005	Dr. T. Mazzulli
Page 3 – change *N. gonorroheae* and *M. catarrhalis* – not tested	November 21, 2005	Dr. T. Mazzulli
Page 3 – add if isolate is resistant to all drugs – add polymyxin and colistin e-test.	November 21, 2005	Dr. T. Mazzulli
Page 4 – add MRSA Screen – MUP e-test	November 21, 2005	Dr. T. Mazzulli
Page 4 – add – if VA-R *E. facalis* and *E. faecium* add VRE MIC panel	November 21, 2005	Dr. T. Mazzulli
Page 4 – *S. pneumo* – Blood and sterile sites add KB e and cc; other sites add KB e and cc	November 21, 2005	Dr. T. Mazzulli
Page 5 – Group A, B, C, G *Streptococcus* – Blood and sterile sites – KB e, cc; p and va if e and cc are R; other sites change to KB e, cc, lvx,va; Add Vaginal/GBS screen – KB e,cc,va. Double disk for e, cc.	November 21, 2005	Dr. T. Mazzulli
Page 5 – Add *S. bovis* – blood and sterile sites – Vitek; mixed and other sites – not tested.	November 21, 2005	Dr. T. Mazzulli
Page 5 – Add *S. milleri* – blood and sterile sites – e-test CRO, P, VA; other sites – KB cc, e, p, lvx; urine – KB p, lvx	November 21, 2005	Dr. T. Mazzulli
Oxacillin Screen – add DENKA and induced DENKA to confirm 159ipro159tibi results	November 21, 2005	Dr. T. Mazzulli
Add *S. aureus* ATCC 29213 for QUAD and Vancomycin Screen Plates QC and recording charts	November 21, 2005	Dr. T. Mazzulli
Re-grouped all reporting pages	November 21, 2005	Dr. T. Mazzulli
Urine – report nitro to all sites for *Staph.* And *Entero.*	November 21, 2005	Dr. T. Mazzulli
Urine – CNST not tested	November 21, 2005	Dr. T. Mazzulli
Urine – add linezolid, synercid to enterococcus if van-R and am-R except *E. gal* and *E. cass.*	November 21, 2005	Dr. T. Mazzulli
MRSA screen – report sxt, mup, doxy, 159ipro159ti with message for re-eradication purpose; fusidic acid if mup=R	November 21, 2005	Dr. T. Mazzulli
Urine – beta strep – add report cc, e	November 21, 2005	Dr. T. Mazzulli
Urine – *S. milleri* – p, lvx	November 21, 2005	Dr. T. Mazzulli
all reporting pages, add call microbiologist if R to all drugs	November 21, 2005	Dr. T. Mazzulli
Reporting tables (Urine, Resp and other sites and blood culture) changes for *S. maltophilia* sxt and levo	November 21, 2005	Dr. T. Mazzulli
Reporting tables (Urine, Resp and other sites and blood culture) changes for *B. cepacia* for sxt, taz, mero	November 21, 2005	Dr. T. Mazzulli
Urine – imipenem – add report if R or R to all other drugs or if only aminoglycoside is S	November 21, 2005	Dr. T. Mazzulli
Resp& Misc – add cc to *S. pneumo*	November 21, 2005	Dr. T. Mazzulli
Resp & Misc – change GBS – delete am, kz,p, tet; add va with foot note-do not report for GBS screen or vag swab; uniform reporting for all beta-strep	November 21, 2005	Dr. T. Mazzulli
All beta strep from non-sterile sites – report sensi with "Susceptibility completed as requested"	November 21, 2005	Dr. T. Mazzulli
Resp – *S. Milleri* – add report p, e, cc	November 21, 2005	Dr. T. Mazzulli
Resp and Misc – SPICE group – add comment if ceftriaxone is S	November 21, 2005	Dr. T. Mazzulli
Resp – add linezolid, synercid to enterococcus if van-R and am-R except *E. gal* and *E. cass*.	November 21, 2005	Dr. T. Mazzulli
Statement for reporting sensi on eye and ear sources	November 21, 2005	Dr. T. Mazzulli
Resp an Misc. *H. flu* – report b-lactamase and message	November 21, 2005	Dr. T. Mazzulli

Page Number / Item	Date of Revision	Signature of Approval
CSF – add linezolid, synercid to enterococcus if van-R and am-R except E. gal and E. cass.	November 21, 2005	Dr. T. Mazzulli
Bloods – add linezolid, synercid to enterococcus if van-R and am-R except E. gal and E. cass.	November 21, 2005	Dr. T. Mazzulli
CSF – cip suppress	November 21, 2005	Dr. T. Mazzulli
CSF – fep suppress	November 21, 2005	Dr. T. Mazzulli
Report of *Haemophilus* spp. And *Neisseria gonorrhoeae* added to Resp and misc, CSF and Bloods	November 21, 2005	Dr. T. Mazzulli
Enterobacterales, report all 3rd 2nd and 1st generation cephalosporins I or R if any 3rd generation cephalosporin is I or R.	November 21, 2005	Dr. T. Mazzulli
Enterobacterales, report all 2nd and 1st generation cephalosporins I or R if any 2nd generation cephalosporin is I or R.	November 21, 2005	Dr. T. Mazzulli
Double Disk for cc and e for beta-strep and *S. pneumo*	November 21, 2005	Dr. T. Mazzulli
KB ESBL interpretation page changed	November 21, 2005	Dr. T. Mazzulli
Table for reporting different ESBL classes added	November 21, 2005	Dr. T. Mazzulli
Drug – ESBL Inhibitor name changed to ESBL Potentiation – result as Y or N	November 21, 2005	Dr. T. Mazzulli
e-test link to reading result pictures	November 21, 2005	Dr. T. Mazzulli
If MRSA isolated from MRSA screen and tet/doxy AND sxt=R, set up fusidic acid e-test	December 30, 2005	Dr. T. Mazzulli
Page 5 – S. bovis from blood and sterile site – set up e-test	January 11, 2006	Dr. T. Mazzulli
Page 5 – S. milleri from blood and sterile site – "pure culture" removed	January 11, 2006	Dr. T. Mazzulli
MRSA screen susceptibility reporting message change	February 15, 2006	Dr. T. Mazzulli
Add inducible Class C ESBL message base on D zone	February 15, 2006	Dr. T. Mazzulli
Page 40 interpretation table – include D zone	February 15, 2006	Dr. T. Mazzulli
Message for fusidic acid and mupirocin interpretation	February 15, 2006	Dr. T. Mazzulli
Page 4 – NEVER refer clinical isolates to isolates from infection control screens. – added	March 06, 2006	Dr. T. Mazzulli
Remove Cefepime testing and reporting	April 05, 2006	Dr. T. Mazzulli
MRSA – mupirocin by KB; if KB-MUP=R, repeat with etest	April 05, 2006	Dr. T. Mazzulli
Add KB piptazo for all *P. aeruginosa*	April 05, 2006	Dr. T. Mazzulli
Report Pen with MIC for *S. bovis* if isolated from blood and sterile sites	April 15, 2006	Dr. T. Mazzulli
All KB-esbl panels – include cefepime	April 15, 2006	Dr. T. Mazzulli
Annual Review	April 15, 2006	Dr. T. Mazzulli
Unusual susceptibility pattern to be rechecked – pip=S and tzp=R	May 26, 2006	Dr. T. Mazzulli
Change list of SPICE bugs for reporting pages 13, 17, 20 and 24 as per QMP-LS broadsheet	May 26, 2006	Dr. T Mazzulli
Report IC screen with revised message – Susceptibility results are provided for infection control purposes only.	May 26, 2006	Dr. T. Mazzulli
Add column for Shigella species reporting for blood, sterile sites and CSF isolates	July 22, 2006	Dr. T. Mazzulli
Revised resulting message for S. saprophyticus and CNST from urine.	September 8, 2006	Dr. T. Mazzulli
Add e-test linezolid and synercid for *E. gallinarum* or *E. casseliflavus* from blood or sterile sites	October 23, 2006	Dr. T. Mazzulli
Modify Vitek link	October 23, 2006	Dr. T. Mazzulli
S. milleri set up pen as e-test for non-sterile and urine sites	November 20, 2006	Dr. T. Mazzulli

Page Number / Item	Date of Revision	Signature of Approval
S. bovis and viridans *Streptococcus* changed from mixed culture to mixed morphotypes susceptibility not done.	November 20, 2006	Dr. T. Mazzulli
Annual Review	April 27, 2007	Dr. T. Mazzulli
Page 3, 4 – resistant gnb – set up CO etest as well as PO	April 27, 2007	Dr. T. Mazzulli
Appendix IX – MIC broth set up change	April 27, 2007	Dr. T. Mazzulli
Report Polymyxin B, with message and MIC, on interpretation	April 27, 2007	Dr. T. Mazzulli
Change from testing and reporting imipenem to meropenem	April 27, 2007	Dr. T. Mazzulli
Always report meropemen for *Acinetobacter* species	April 27, 2007	Dr. T. Mazzulli
Change CNST sensi testing rules for blood cultures	February 13, 2008	Dr. T. Mazzulli
Added canned comments for CNST, S. lugdunensis, Proprionibacterium, Bacillus and Corynebacterium sp when isolated from blood cultures	February 13, 2008	Dr. T. Mazzulli
Added report doxycycline, mupirocin and fusidic acid on MRSA if isolated from any source of Bridgepoint patients	February 13, 2008	Dr. T. Mazzulli
Annual Review	April 10, 2008	Dr. T. Mazzulli
S. millerr, viridans strep, small colonies beta strep from non-sterile sites, set up susceptibility testing on request only	April 10, 2008	Dr. T. Mazzulli
Added –set up KB for amikacin on all *Acinetobacter baumanii*	April 10, 2008	Dr. T. Mazzulli
Urine – Staphylococcus reporting table – combined MSH, UHN and RVHS columns	April 10, 2008	Dr. T. Mazzulli
Urine – Enterococcus reporting table – combined MSH, UHN and RVHS columns	April 10, 2008	Dr. T. Mazzulli
S. aureus vancomycin mic=2.0 mcg/L reporting message	April 10, 2008	Dr. T. Mazzulli
Do not report Ampicillin, Cefazolin and Nitrofurantoin for *Acinetobacter* species	April 10, 2008	Dr. T. Mazzulli
Disk Diffusion – expanded interpretation procedure	April 10, 2008	Dr. T. Mazzulli
S. pneumo reporting – changed reporting moxifloxacin for UHN patients to levofloxacin	April 10, 2008	Dr. T. Mazzulli
Report meropenem on all CSF with Enterobactericeae and Acinetobacter	April 10, 2008	Dr. T. Mazzulli
Appendix XIII – ertapenem added to drug list	August 20, 2008	Dr. T. Mazzulli
"For E. coli, Klebsiella *species and* Proteus *species that are confirmed to have an ESBL of any class, report all penicillins and first, second and third generation cephalosporins and* ***piperacillin/tazobactam*** *as R."* **piperacillin/tazobactam** – rule added to Enterobacterales reporting	August 20, 2008	Dr. T. Mazzulli
Ceftriaxone – Report on all Acinetobacter *species (change in Urine reporting)*	August 20, 2008	Dr. T. Mazzulli
?BORSA added confirmation of mic by PHL	August 20, 2008	Dr. T. Mazzulli
Polymyxin B and Colistin reporting change for P.aeruginosa *and* Acinetobacter *species*	August 20, 2008	Dr. T. Mazzulli
Report moxifloxacin for RVHS and Ajax for S. pneumo	August 20, 2008	Dr. T. Mazzulli
Report 161ipro instead of orflox for Bridgepoint, RVHS and Ajax	August 20, 2008	Dr. T. Mazzulli
Clarify interpretation of vancomycin macro e-test for Staphylococcus	November 03, 2008	Dr. T. Mazzulli
Changed S. anginosus *group, viridans strep group suspectibility testing on non-sterile sites to KB (if requested)*	January 05, 2009	Dr. T. Mazzulli
"or I" added to SPICE message:	January 05, 2009	Dt. T. Mazzulli

Page Number / Item	Date of Revision	Signature of Approval
Citrobacter spp., *Enterobacter* spp., *Hafnia* spp., *Morganella morganii, Proteus vulgaris, Providencia* species, *Serratia* species, if **S or I**, report with comment "Resistance to extended-spectrum penicillins, beta-lactam/beta-lactamase inhibitor combinations, and cephalosporins may develop during therapy with these agents. For serious infections, these agents should be avoided and consultation with a medical microbiologist or infectious disease physician is strongly recommended."		
Modify appearance of Table of Contents	January 05, 2009	Dr. T. Mazzulli
Annual Review	May 10, 2009	Dr. T. Mazzulli
Added double disk CC/E for Staphyloccoccus testing	May 10, 2009	Dr. T. Mazzulli
Remove reflex rules for CC/E on Staphylococcus	May 10, 2009	Dr. T. Mazzulli
VISA screen added	May 10, 2009	Dr. T. Mazzulli
Remove CNST from vancomycin screen plate	May 10, 2009	Dr. T. Mazzulli
Modified etest procedure	May 10, 2009	Dr. T. Mazzulli
Modified medium for S. aureus on screen plate and etest	May 10, 2009	Dr. T. Mazzulli
Added Modified Hodge Test	May 10, 2009	Dr. T. Mazzulli
Modified MHT resulting phrases	March 15, 2010	Dr. T. Mazzulli
Modified S. anginosus reporting phrases if susceptibility is not tested	March 15, 2010	Dr. T. Mazzulli
Report moxifloxacin on S. pneumo for UHN patients instead of levo	March 15, 2010	Dr. T. Mazzulli
Modified ESBL template	May 26, 2010	Dr. T. Mazzulli
Added reporting phrase for ESBL D zone=Y, Potentiation=Y	May 26, 2010	Dr. T. Mazzulli
Annual Review	June 04, 2010	Dr. T. Mazzulli
Modified GBS from urine panel and reporting	June 04, 2010	Dr. T. Mazzulli
Revised Modified Hodge Test "Interpretation" section	September 15, 2010	Dr. T. Mazzulli
E coli and Kleb species, if VT is S to Ceftriaxone and I/R to pip/tazo, set up KB pip/tazo to confirm	September 15, 2010	Dr. T. Mazzulli
Stool Transplant Study comment added to C. diff isolates	October 08, 2010	Dr. T. Mazzulli
Report meropenem for Gram negatives if I added to statement of report if R.	October 16, 2010	Dr. T. Mazzulli
Cipro in urines reporting for RVHS corrected (it was omitted in error in the last revision)	October 16, 2010	Dr. T. Mazzulli
Added picture of NDM-1 in Modified Hodge Test section	October 22, 2010	Dr. T. Mazzulli
Removed Modified Hodge Test section.	November 11, 2010	Dr. T. Mazzulli
Send all erta I or R Enterobacterales to PHL for KPC PCR	November 11, 2010	Dr. T. Mazzulli
Carbapenemase reporting with ESBL screen added	November 11, 2010	Dr. T. Mazzulli
Added meropenem disk to IC ESBL screen plate	November 11, 2010	Dr. T. Mazzulli
Removed pip/tazo from routine reporting in Enterobacterales. If requested, set up KB and report using KB results.	November 11, 2010	Dr. T. Mazzulli
Added Carbapenemase Screening Section	November 17, 2010	Dr.T. Mazzzulli
Modified Steno resulting phrase	November 17, 2010	Dr.T. Mazzzulli
Added Tigecycline reporting phrases	November 17, 2010	Dr.T. Mazzzulli
Updated Carbapenemase send out procedure	November 23, 2010	Dr. T. Mazzulli
Updated Carbapenemase testing to Rosco KPC MBL Confirm Kit disks	January 20, 2011	Dr. T. Mazzulli
Rosco disk added to Antimicrobial table	January 20, 2011	Dr. T. Mazzulli
Updated criteria for susceptibility table	January 20, 2011	Dr. T. Mazzulli

Page Number / Item	Date of Revision	Signature of Approval
Page 4, added KB for cefazolin if requested or from sterile sites	February 23, 2011	Dr. T. Mazzulli
Page 18 changed cefazolin reporting to – report from KB only if requested	February 23, 2011	Dr. T. Mazzulli
Page 25 changed cefazolin reporting to – report from KB only	February 23, 2011	Dr. T. Mazzulli
Added TREK sensititre for S. pneumoniae – procedure and modified reporting sections	March 14, 2011	Dr. T. Mazzulli
For E. gallinarum or E. casseliflavus and VA=R, E. faecalis or E. faecium from Blood & Sterile sites updated to include set up linezolid and synercid	June 15, 2011	Dr. T. Mazzulli
"Susceptibility tested on pure cultures ONLY" – added to Criteria for testing for clarity.	June 15, 2011	Dr. T. Mazzulli
Cephalexin added to Urine gram negative reporting	July 18, 2011	Dr. T. Mazzulli
Annual Review	July 18, 2011	Dr. T. Mazzulli
Added Colistin etest to resistant Enterobacterales + colistin reporting canned messages	July 18, 2011	Dr. T. Mazzulli
Added not to report Erythromycin on GBS for GBS screen	November 18, 2011	Dr. T. Mazzulli
Added Tigecycline to reporting tables	November 18, 2011	Dr. T. Mazzulli
Added ertapenem reporting if ertapenem=I/R or MDR	November 18, 2011	Dr. T. Mazzulli
Zone size interpretation change for ROSCO meropenem 10	December 12, 2011	Dr. T. Mazzulli
Blood Culture Yeast sensi refer back up to 7 days	August 2, 2012	Dr. T. Mazzulli
Annual Review	August 2, 2012	Dr. T. Mazzulli
Revised OXA, VANC workflow instructions	August 2, 2012	Dr. T. Mazzulli
Remove Teico etest with 0.5 McFarland for S. aureus	August 2, 2012	Dr. T. Mazzulli
Removed routine KB tzp testing for Ps aeruginosa and Enterobacterales	December 12, 2012	Dr. T. Mazzulli
Added line for IC isolates refer back 3 months on page 4 – criteria for testing	December 12, 2012	Dr. T. Mazzulli
Acinetobacter, test for KB Amikacin if Gent and Tob are R	December 12, 2012	Dr. T. Mazzulli
Interpretations for reporting changed as per CSLI M100-S22 (zone size and mic changes)	December 12, 2012	Dr. T. Mazzulli
Revised resulting messages for Colistin and Tigercycline	January 25, 2013	Dr. T. Mazzulli
Reflexed tzp to be I/R if any 3rd gen cephalosporin is I or R	January 25, 2013	Dr. T. Mazzulli
Revised table format of "set up criteria	July 17, 2013	Dr. T. Mazzulli
Removed all polymyxin testing and reporting	July 17, 2013	Dr. T. Mazzulli
Added sent to PHL for MIC for S.maltophilia and B. cepacia	July 17, 2013	Dr. T. Mazzulli
Updated canned message section	July 17, 2013	Dr. T. Mazzulli
Remove RVHS from all reporting tables .	July 17, 2013	Dr. T. Mazzulli
Interpretations for reporting changed as per CSLI M100-S23	July 17, 2013	Dr. T. Mazzulli
Annual Review	July 17, 2013	Dr. T. Mazzulli
Merged MSH and UHN columns on all reporting tables	October 10, 2013	Dr. T. Mazzulli
Added set up and reporting for Aeromonas species	October 10, 2013	Dr. T. Mazzulli
Added meropenem screen to replace ertapenem screen for infection control CRE screens	October 10, 2013	Dr. T. Mazzulli
Report Amox/clavu for Enterobacteriacae; set up kb for amox clav	October 10, 2013	Dr. T. Mazzulli
Remove reporting am, cf, nitro, sxt for all sites in Pseudomonas aeruginosa	October 10, 2013	Dr. T. Mazzulli

Page Number / Item	Date of Revision	Signature of Approval
Vitek interpretation change for cepodoxime – from ≤2=R to ≤0.5=R; 4=I removed, from ≥8=R to ≥4=R	October 10, 2013	Dr. T. Mazzulli
cc=R e=S for Staph and Strep – repeat and confirm id; freeze, added to page 12	October 10, 2013	Dr. T. Mazzulli
Remove cefurozime from Resp Enterobacterales	October 10, 2013	Dr. T. Mazzulli
Report HLGR results on enterococcus from blood and sterile sites only when vancomycin is susceptible	October 10, 2013	Dr. T. Mazzulli
Remove cloxacillin from reporting on Staph in spinal fluid	October 10, 2013	Dr. T. Mazzulli
Staphylococcus from Blood Culture – remove e, cc and sxt from reporting	October 10, 2013	Dr. T. Mazzulli
Remove ciprofloxacin from reporting for enterococci to TRI urine	October 10, 2013	Dr. T. Mazzulli
Salmonella – remove nalidic acid testing, set up kb and etest for 164ipro and suppress cip from reporting from Vitek (Vitek has older breakpoints)	October 10, 2013	Dr. T. Mazzulli
Report Doxycycline on Staphs from all tissues. Wounds (not from respiratory sites) and urine	October 10, 2013	Dr. T. Mazzulli
Report moxifloxacin on staphs from bone/joint	October 10, 2013	Dr. T. Mazzulli
Modified positive Carbapenemase reporting phrase	October 10, 2013	Dr. T. Mazzulli
No sensi set up for CNST other than S. lugdenensis isolated from all blood cultures, report with new message	November 13, 2013	Dr. T. Mazzulli
Report erta and mero when it is I/R for all gram negatives	November 13, 2013	Dr. T. Mazzulli
For Enterobacterales, report erta and mero when it is I/R OR I/R to 2 of the 3 antimicrobial agents: amikacin, ciprofloxacin, 3rd Generation Cephalosporins	November 19, 2013	Dr. T. Mazzulli
For *P. aeruginosa*, report mero when it is I/R OR I/R to 2 of the 3 antimicrobial agents: amikacin, ciprofloxacin, 3rd Generation Cephalosporins AND pipercillin/tazobactam	November 19, 2013	Dr. T. Mazzulli
Report Doxycycline on *S. aureus* from all tissues. Wounds (not from respiratory sites) and urine (change frm all *Staphylococcus*)	November 19, 2013	Dr. T. Mazzulli
Modified KB panel set up for Enterobacterales to	November 19, 2013	Dr. T. Mazzulli
Modified *Aeromonas* set up panel	November 19, 2013	Dr. T. Mazzulli
Added secondary reporting drugs to all *Aeromonas*	November 19, 2013	Dr. T. Mazzulli
Page 73 macro etest table second category 2, modified teicoplanin from "< or > 12 mg/L" to "≥8 mg/L"	November 19, 2013	Dr. T. Mazzulli
Neisseria gonorrhoeae all sites – send to PHL for susceptibility	November 19, 2013	Dr. T. Mazzulli
Enterobacterales – Report meropenem and ertapenem if I/R **OR** if I/R to 3 of the 4 antimicrobial agents: amikacin, ciprofloxacin, 3rd Generation Cephalosporins, Septra **OR** if requested	December 24, 2013	Dr. T. Mazzulli
P. aeruginosa – Report meropenem if I/R **OR** if I/R to 3 of the 4 antimicrobial agents: amikacin, ciprofloxacin, 3rd Generation Cephalosporins, Piperacillin/tazobactam **OR** if requested	December 24, 2013	Dr. T. Mazzulli
Doxycycli ne R comment added	December 24, 2013	Dr. T. Mazzulli
B. cepacia to PHL – change request to levo aand tcc	December 24, 2013	Dr. T. Mazzulli
Report all SPICE bugs R to all beta-lactem and beta-lactem/inhibitors drugs	December 24, 2013	Dr. T. Mazzulli
Added ceftriaxone etest to S. anginosis isolated from non-sterile sites. To be reported when Pen is I/R	December 24, 2013	Dr. T. Mazzulli

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Report if I/R to **All** other Antimicrobial Agents **OR** if only aminoglycoside is S **OR** if requested *Report if I/R to **All** oral Antimicrobial Agents (i.e. amoxicillin, amoxicillin-clavulanic acid, cephalexin, TMP-SMX, ciprofloxacin, doxycycline, tetracycline, nitrofurantoin) **OR** if requested.*	January 25, 2014	Dr. T. Mazzulli
Added insructions for set up if Vitek card is terminated under "Criteria for Susceptibility Testing"	February 10, 2014	Dr. T. Mazzulli
Updated S. maltophilia and B, cepacia etest	February 10, 2014	Dr. T. Mazzulli
Stenotrophomonas maltophilia Send to PHL for MIC for TCC and Minocycline	March 31, 2014	Dr. T. Mazzulli
Acinetobacter set up meropemen by KB; send to NML for PCR id mero=I/R	May 25, 2014	Dr. T. Mazzulli
Updated zone size and etest breakpoint for *Acinetobacter* to 2014 CLSI guidelines	May 25, 2014	Dr. T. Mazzulli
Added KB panel for resistant gnb (kbxdr panel)	May 25, 2014	Dr. T. Mazzulli
Annual Review	May 25, 2014	Dr. T. Mazzulli
Added KB for sxt for MSSA and MRSA	May 25, 2014	Dr. T. Mazzulli
*B*LACTA Test added	June 27, 2014	Dr. T. Mazzulli
Base on Cefoxitin to rule out MRSA	June 27, 2014	Dr. T. Mazzulli
CRE reporting changes	June 27, 2014	Dr. T. Mazzulli
Confirmed by KB if Vitek SXT = R	June 27, 2014	Dr. T. Mazzulli
Revised Salmonella set up and reporting, removed Vitek	June 27, 2014	Dr. T. Mazzulli
Vitek AES breakpoint changes	July 12, 2014	Dr. T. Mazzulli
Changed rule out MRSA "Cefoxitin" to "Cefoxitin Screen"	July 12, 2014	Dr. T. Mazzulli
Modified Hemophilus beta-lactamase reporting phrase for non-sterile sites	July 12, 2014	Dr. T. Mazzulli
Updated BLACTA reporting phrases	July 12, 2014	Dr. T. Mazzulli
Update UHN/MSH logo Correct file path in footer	August 27, 2014	Dr. T. Mazzulli
Teico and Vancomycin Etest for Staph aureus	September 27, 2014	Dr. T. Mazzulli
VISA-Hvisa-vrsa	September 27, 2014	Dr. T. Mazzulli
Changed B cepacia to etest for SXT	September 27, 2014	Dr. T. Mazzulli
Change Enterococcus from KB to etest for linezolid	September 27, 2014	Dr. T. Mazzulli
Revised addition of Linezolid, daptomycin, tigercillin for BORSA/MRSA	September 27, 2014	Dr. T. Mazzulli
Pseudomonas aeruginosa – Changed from "If resistant to **all** routinely tested_antimicrobials (**including** aminoglycosides). KB (kbxdrpa). ATM, FEP, TIM" to: "If resistant to **all** routinely tested_antimicrobials and colistin (**excluding** aminoglycosides). KB (kbxdrpa). ATM, FEP, TIM"	October 25, 2014	Dr. T. Mazzulli
Enterococcus change panel to astgp67	October 25, 2014	Dr. T. Mazzulli
Perform susceptibilities on CNST in BC if isolated from patients with endocarditis	October 25, 2014	Dr. T. Mazzulli
Added link to bactericidal vs static drug table for Etest reading	December 30, 2014	Dr. T. Mazzulli
Remove Piperacillin/Tazobactam on Enterococcus	December 30, 2014	Dr. T. Mazzulli
Report levo and add linezolid to enterococci from urine if no other oral options	February 9, 2015	Dr. T. Mazzulli

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BORSA detection update	February 9, 2015	Dr. T. Mazzulli
Annual Review	February 9, 2015	Dr. T. Mazzulli
p.28 viridans note#4: added "only if in BC or heart tissue specimen"	April 30, 2015	Dr. T. Mazzulli
Urine p. 13, clarified clinda/levo comments, added #17	April 30, 2015	Dr. T. Mazzulli
Urine GPC – chart change: s. 166ipro166 to s. species	April 30, 2015	Dr. T. Mazzulli
Aeromonas: Added tetracycline to all sites for *Aeromonas* spp. With comment >13yrs Aeromonas panel added for Enterics with reporting results.	May 27, 2015	Dr. T. Mazzulli
Fosfomycin KB from FOS to FOT, change concentration to 200ul Fixed typo: all sites enterococcus Screen added 'va"	July 21, 2015	Dr. T. Mazzulli
Removed setting up double disk KB for all Staphylococcus when vitek is ICR-/cc=S/e=R. For MRSA suppress clindamycin and release with comment.	July 29, 2015	Dr. T. Mazzulli
Removed b-lacatamase testing for blood/sterile sites enterococci	July 29, 2015	Dr. T. Mazzulli
Added Previous positive CRE and ESBL LIS comments to canned message section. Added previous positive refer back criteria and reporting for clinical and IC screen on ARO detection.	August 20, 2015	Dr. T. Mazzulli
Under "When to test" at end of page added reference to link to folder with CLSI guidelines.	August 27, 2015	Dr. T. Mazzulli
Added hyperlinks to CRE How To Detect Section for reporting.	October 6, 2015	Dr. T. Mazzulli
Under "What to Test" for Enterobacterales for : B. coli, K. pneumonia, K. oxytoca or P. mirabilis: If CPD=I or R or BLACTA+ Added: **ONLY on specimens : MSH newborn:D1-M13 & MSH Female 12-50yrs**	October 28, 2015	Dr. T. Mazzulli
p.10 Removed in section for Enterococcus spp: if Nitro I/R from Vitek: set up KB FD p.94 Enterococcus QUAD screen log: added BHI 24h column and added "bench" with No. column. Staph Ox screen, added "bench" with No. column	November 25, 2015	Dr. T. Mazzulli
Update "How to detect" section for CRE: Routine and Screening" Added CRE Clinical/IC screen flowcharts Added CRE reporting tables Updated What to test table for Enterobacterales Updated ROSCO with procedure and reporting Added BCARBA procedure	December 21, 2015	Dr. T. Mazzulli
Ceftazidime for Enterobacterales suppressed from reporting for all sites	December 30, 2015	Dr. T. Mazzulli
For fastidious and nonfermenting GNBs in What to test section, Added: Blood and Sterile sites send to PHOL and HACEK group to name. For Blood cultures and Sterile sites "What to report" added footer note to report as per PHOL susceptibilities.	January 7, 2016	Dr. T. Mazzulli
"What to set up" section: Moraxella added for BC and Sterile sites send to PHL for sensi	January 19, 2016	Dr. T. Mazzulli

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"What to report" for Aeromonas in each site, Tetracycline: Report if I/R to **All** ciprofloxacin, amoxicillin/clavalacnic acid and trimethoprim/sulfamethoxazole		
Remove link to TREK Manual Resp/non-sterile: Vancomycin Ceftriaxone: Report if Pen I or R or send to PHL Report Ceftriaxone, Vancomycin for S.pneumo from sterile sites, no conditions	February 24, 2016	Dr. T. Mazzulli
For *Staphylococcus species* Cloxacillin and Cefazolin reporting added note: "for *Staphylococcus pseudointermedius* base on Oxacillin result. BCARBA test: added Brilliance UTI agar to acceptable testing agars. Added Etest Drug Ceftolozane-Tazobactam Remove Amoxicillin Clavulanic acid from Aeromonas set up panel & reporting tables Remove Amoxicillin Clavulanic acid, Piperacillin/Tazobactam, Ertapenem, Tigecycline from reporting on gram negatives for Spinal Fluid specimens Aminoglycosides and Septra suppressed unless Ceftriaxone is non-susceptible for CSF specimens.	April 4, 2016	Dr. T. Mazzulli
Annual Review *Aerococcus* species added to what to test table. Susceptibilty comment added to reporting tables.	May 9, 2016	Dr. T. Mazzulli
-Updated MSH logo in header -Updated CRE comments \NCRB, \CNML, }NCRB, \pCRB, \PCRB -Removed "CNSIP" send out in CRE flowcharts for IC / Clinical -For *S. pneumo* on all sites, add Oxacillin KB; report for sterile sites as: If Oxacillin=S and Penicillin etest=S, report as S. If Oxacillin=R and Penicillin etest=R, report as R. If Oxacillin=S and Penicillin etest=R, unusual result, confirm. If Oxacillin=R and Penicillin etest=S, report base on PHOL Penicillin MIC Report on non-sterile sites: Base on Oxacillin result if S. **OR** if Oxacillin is R, base on Penicillin etest if I or R **OR** if Oxacilin is R, and Penicillin etest is S, base on PHOL MIC Added to list of unusual results for *S.pneumo*Oxacillin=S & Penicillin etest R -Do NOT set CZ on proteus mirabilis -Enterobacterales from urines: Report Fosfomycin if I/R to all of the following: amoxicillin/ampicillin, amox/clav, cephalexin, ciprofloxacin, nitrofurantoin and TMP/SMX, or if Requested. Report *E. coli* with interpretation. Report other *Enterobacterales* with zone diameter and Isolate Message. For *E. coli* where fosfomycin is not reported, add Isolate Message "*E. coli* is generally susceptible to fosfomycin for treatment of acute uncomplicated cystitis." -Enterococcus from urines: Report Fosfomycin for I/R to ampicillin and nitrofurantoin. For *E. faecalis* report interpretation. For *E. faecium* report with with zone diameter and Isolate message. For *E.*	July 26, 2016	Dr. T. Mazzulli

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faecalis where fosfomycin is not reported, add Isolate Message "*E. faecalis* is generally susceptible to fosfomycin for treatment of acute uncomplicated cystitis." -Annual Review		
Changed N.meningitidis from No sensi to send to PHL as per IQMH 2016.07.06 practice recommendations for AST. Added Vibrio to "What to set up" table for enterics (not sensi) and sterile sites (send to PHL) as per IQMH 2016.12.14 Stool reporting Practice recommendations. Addition of Appendix "AGENTS NEVER TO BE REPORTED BY SITE" Added etest panel to Acinetobacter, Steno, Burkholderia for resistant etest (etresa)	July 26, 2016	Dr. T. Mazzulli
Addition of routine septra etest set up for *S.maltophilia* and send out to PHL if KB and etest disagree.	November, 18, 2016	Dr. T. Mazzulli
Amoxicillin Etest added to Abbreviation list Aeromonas susceptibility removed from Enteric sites (on request only). Added 168ipro168tibility comment for Aeromonas spp. To report with "Resistance to non-carbapenem beta-lactam antimicrobials may develop in Aeromonas species during therapy. Choosing a non-beta-lactam antimicrobial should be considered for serious infections. Consultation with infectious diseases or medical microbiology is advised." Added link to TREK manual in the TOC.	February 1, 2017	Dr. T. Mazzulli
Addition of susceptibility options for *Shigella* spp when requested on enteric sites. Reporting ciprofloxacin phrase for *Shigella* Etest added to reporting table for enteric sites.	February 6, 2017	Dr. T. Mazzulli
Under What to Test, temporary procedure change instructions for Vitek card recall added. Added to appendix XIX for temporary procedure change instructions for Vitek astn213 and Vitek astgp67 susceptibliity results.	April 26, 2017	Dr. T. Mazzulli
Annual Review Urine cephalexin reflexed from cefazolin vitek2 result. Reported only for *E.coli, Klebciella pneumonia* and *Proteus mirabilis.* Cephalexin =I/R will reflex Ampicillin =R for *E.coli, Klebciella pneumonia.* Added *Staphylococcus pseudointermedius* & *Staphylococcus intermedius* in "What to set up" table with *Staphylococcus lugdenensis*	May 3, 2017	Dr. T. Mazzulli
Removed Amp/Sulbactam from set up and reporting for enterobacteriaciae and Acinetobacter spp.	May 17, 2017	Dr. T. Mazzulli
Added set up etest Daptomycin for Enterococcus: All sites, if VA=R or *vanA* positive, *E. faecalis* or *E.faecium*	May 18, 2017	Dr. T. Mazzulli
Updated What to set up for Urine GBS and Urine Group A, C, F. Resp and non-sterile Beta strep reporting, removed duplicate comment #20 (duplicate to comment #2) For Urine Beta strep comment with Clinda for insignificant amounts, updated comment to include "for intrapartum chemoprophylaxis"	June 2, 2017	Dr. T. Mazzulli

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Urine Beta strep comment #12 removed "and erythromycin".		
Removed: Under What to Test, temporary procedure change instructions for Vitek card recall added. Added to appendix XIX for temporary procedure change instructions for Vitek astn213 and Vitek astgp67 susceptibliity results.	July 22, 2017	Dr. T. Mazzulli
For *P. aeruginosa* what to set up, • removed KB disks ATM/FEP/TIM to set up "If resistant to **all** routinely tested antimicrobials and colistin (**excluding** aminoglycosides)" • Added ATM /FEP KB to "If resistant to **all** routinely tested antimicrobials (**excluding** aminoglycosides)"	July 28, 2017	Dr. T. Mazzulli
Implementation of Acinetobacter for CPO screening: Added Acinetobacter to CPO flowcharts for both clinical and IC The resulting comment codes/notifications for Acinetobacter have been added.	August 2, 2017	Dr. T. Mazzulli
Addition of results phrases/canned messages for Acinetobacter negative and positive comments when returning from NML (\ACCN & \ACCP)	August 8, 2017	Dr. T. Mazzulli
Rifampicin and Amoxicillin etest abbreviations added. Modified Shigella "What to set up" from All sites Vitek to only non-enteric sites Vitek.	August 18, 2017	Dr. T. Mazzulli
Added result of Haze as acceptable for OX screen plate with *S.aureus* ATCC43387	September 25, 2017	Dr. T. Mazzulli
Added reporting of Ceftolozane/Tazobactam for Enterobacterales comment and *P.aeruginosa* when: I/R to **All** other Antimicrobial Agents **OR** if only aminoglycoside is S **OR** when requested	October 27, 2017	Dr. T. Mazzulli
Urine what to report for entero: linked comment 20 "if "S" for *E.faecalis* add Isolate Message "*E. faecalis* is generally susceptible to fosfomycin for treatment of acute uncomplicated cystitis." To reported drugs Amp, Tet, Nitro Added set up KB fos to Etnerobacterieacea "If resistant to **all** routinely tested antimicrobials(**excluding** aminoglycosides)" for Urines Added for Aeromonas isolated from enterics, no sensi to be set up unless requested. Removed Chloramphenicol from Urine Enterobacterales reporting. All other sites Non-Urine release Chloramphenicol if I/R to **All** other Antimicrobial Agents	December 7, 2017	Dr. T. Mazzulli
Annual Review	May 20, 2018	Dr. T. Mazzulli
Increased templates for screen plates. Replaced Etests and PHOL testing for *S.pneumoniae* and replaced with TREK set up. Modifed reporting rules accordingly.	August 29, 2018	Dr. T. Mazzulli
Added CRE IC canned message	Sept 14, 2018	Dr. T. Mazzulli
New etest Ceftobiprole added for MRSA on request only to all applicable sections.	November 18, 2018	Dr. T. Mazzulli
Added reporting phrase for tigecycline: "Results for tigecycline is based on Etest gradient strips (bioMérieux)	November 18, 2018	Dr. T. Mazzulli

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which have been validated with well-characterized laboratory (ATCC) strains. Verification on clinical isolates against a gold standard method has been limited. Please take this into consideration when interpreting these results."		
Added reporting phrase for Ceftolozane/tazobactam: "Results for ceftolozane/tazobactam is based on Liofilmchem gradient strips (Alere) which have been validated with well-characterized laboratory (ATCC) strains. Verification with clinical isolates against a gold standard method has been limited. Please take this into consideration when interpreting these results."	November 18, 2018	Dr. T. Mazzulli
Updated Table of Contents links	November 18, 2018	Dr. T. Mazzulli
Add Rifampin (RI) to list of e-test abreviations Added *H.pylori* susceptibility requirements	November 30, 2018	Dr. T. Mazzulli
Corrected *H.pylori* set up from CM to CH (Clari) Corrected spelling of tetracycline in comments for reporting *H.pylori*	December 10, 2018	Dr. T. Mazzulli
Modified step three in Denka procedure from water bath to 100C heating block with note.	January 11, 2019	Dr. T. Mazzulli
Annual Review pg 7-9 "Section: What to Test Enterobacterales (not SPICE) sterile and on request – KB cefazolin, ertapenem, tobramycin, amikacin Enterobacterales (not SPICE) on request – KB ampicillin SPICE – KB 170ipro, gentamicin, tobramycin, amikacin Proteus non-vulgaris on request – KB amox/clav Pseudomonas aeruginosa – KB pip-tazo, ceftaz, 170ipro, tobra, amikacin Salmonella, Shigella – KB/gradient strip all antimicrobials deleted Shigella enteric isolates AST testing reference"	July 16, 2019	Dr. T. Mazzulli
pg 13 Enterococcus – add "excluding respiratory" to all sites for DPC pg 26 Resp/Misc Gram Pos Reporting Table: update Footnote 14 for Daptomycin to exclude respiratory pg 35 Blood/Sterile Sites Table: change Footnote 11 to Footnote 7 for Daptomycin/delete Footnote 11	July 31, 2019	Dr. T. Mazzulli
Update MRSA Screen test from DENKA to PBP2 Screen	November 29, 2019	Dr. T. Mazzulli
Removed need for CPO workup comment } \ NCRB and notification to ICPs.	December 31st, 2019	Dr. T. Mazzulli
Safety Precautions added to PBP2 MRSA Screen	February 21, 2019	Dr. T. Mazzulli
Added a section to Enterobacterales chart: All sites – E.coli and P. mirabilis from NICU to add AMP KB	March 30, 2020	Dr. T. Mazzulli
Changed Burkholderia *cepacia from Sensitivity chart to Burkholderia cepacia Complex including [1]:* Burkholderia cepacia Burkholderia gladioli Burkholderia vietnamiensis Burkholderia multivorans Burkholderia pyrrocinia Burkholderia ubonensis Burkholderia stabilis Burkholderia ambifaria	April 16, 2020	Dr. T. Mazzulli

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Annual Review		

Full document review included in all updates. Bi-annual review conducted when no revision had been made within 2 years.

Page Number / Item	Date of Revision	Edited by:
For Spinal fluids – Enterobacterales: Removed Genta, tobra and amikacin as reportable drugs Mero released based on Ceftriatxone I or R only.	November 18, 2020	Dorna Zareianjahromi
Acinetobacter sp – Resistant to all routine drugs, removed Tetracycline for non-urine samples	Dec 21, 2020	Dorna Zareianjahromi
Added Amp KB to sterile non-spice, updated Acinetobacter amikacin KB, updated S aureus mupiricin fusidic acid, formatting change to WHAT TO TEST Enterobacterales section	Mar 12, 2021	Wayne Chiu
Updated Ps aeruginosa What to setup – KB	Mar 15, 2021	Wayne Chiu
Updated wording for ERT/MEM suppression rules for GNB in urine, resp, other non-sterile sites	Mar 19, 2021	Wayne Chiu
Changed what to setup for GNB other afermenters – refer PHOL all sites. Updated Enterococcus what to setup for cass/gall	Mar 23, 2021	Wayne Chiu
Minor formatting change	April 11, 2021	Jessica Bourke
Nomenclature update – C diff, cuti acnes, enterobacterales	April 19, 2021	Wayne Chiu
Added doxy for MDR enterobacterales , and add doxy fosfo for MDR enterococcus Updated SH IPAC comments for MRA, VRE, CRE and ESBL to standard “contact precautions” comment \ICPR. Specified colistin reporting on Acinetobacter sp is for baumanii complex only. Added dori,ert,imi,lefamulin to “never report” list for CSF	June 9, 2021	Wayne Chiu
Updated clinical CRE flowchart Removed colistin etest	June 11, 2021	Wayne Chiu
Included examples of hacek group and other afermenter group Included Eye specimen in erythromycin reporting from strep pneumo	June 18, 2021	Wayne Chiu
Specified enterobacterales MDR testing dx and co Inserted section: Comment Template for infrequently tested antibiotics	June 29, 2021	Wayne Chiu
Updated ESBL inducible class C	July 8, 2021	Wayne Chiu
Updated Enterobacterales MDR and blacta section	July 14, 2021	Wayne Chiu
Added comment 5 for enterobacterales nonspice nonsterile Added KB option for stalug	July 22, 2021	Wayne Chiu
Adjusted fosfo agar dilution comment	July 27, 2021	Wayne Chiu
Removed “non-carbapenem” from aeromonas comment	Aug 27, 2021	Wayne Chiu
Replaced DENKA with PBP2a test	Aug 31, 2021	Wayne Chiu
Updated infrequent AST comments	Sep 20, 2021	Wayne Chiu
Updated Imi-rele comments, CLSI interp now available for Enterob and Pseudo	Sep 21, 2021	Wayne Chiu
Added cefiderocol KB comments	Sep 27, 2021	Wayne Chiu
Added AMC to KB panel when vitek fails Enterobacterales	Oct 5, 2021	Wayne Chiu

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Updated cefiderocol KB for Acineto and Steno	Nov 11, 2021	Wayne Chiu
Added "considering combination therapy" to aeromonas comment	Nov 15, 2021	Wayne Chiu
Added levo and doxy to enterococcus and viridans strep testing and reporting for bone/joint, non-sterile sites	Dec 10, 2021	Wayne Chiu
Added section for filamentous fungus "What to Test"	Jan 7, 2022	Wayne Chiu
Added note SXT, do not report <2 months old	March 24, 2022	Wayne Chiu
When referring AST to another sample, please specify collection date AND time	April 12, 2022	Wayne Chiu
Updated mupirocin and fusidic acid comments	May 25, 2022	Wayne Chiu
Update testing for H. pylori – sendout to Mayo only and report per Mayo report. Clarified send out to PHOL for AST for Pseudo other than aerug and other afermenters – all sites Included Capnocytophaga under "other fastidious gram neg"	July 11, 2022	Wayne Chiu
Removed fusidic acid from testing, no longer available	July 31, 2022	Wayne Chiu
Add Ampicillin –S as uncommon results to Enterococcus faecium	August 10, 2022	Qin Liu
Removed notification instructions for rosco, refer to Isolate notification table instead Added pantoea sp to SPICE group, added hyperlinks to replace lists Specified not to test or report cefazolin on P mirabilis In Appendix A unusual result section, specifiy viridian strep includes anginosus group	November 11, 2022	Wayne Chiu
Minor wording updates to AST Appendix C New/Old work up . Clarification on CLSI and Eucast comment additiona for Fosfo as Infrequently tested antibiotic comments.	May 19, 2023	Jessica Bourke
Temporary procedure for Shigella starting May 20th, 2023 added to manual with reporting phrase. Minor formatting on page 12 p.9 Steno if fully resistnant moved Minocycline KB to in house testing.	August 14, 2023	Jessica Bourke
Romoved High level gm500 and st2000 from non-blood sterile site (page 12) Changed from *See Blood and Sterile Fluids HLGR Results Reporting* to *See Blood HLGR Results Reporting* (page 30). Changed from *See Blood and Sterile Fluids HLGR Results Reporting* to *See Blood HLGR Results Reporting* (page 34). Updated in the section of Reporting Results (page 124): *Blood cultures when ampicilline and / or vancomycin is susceptible*	October 27, 2023	Qin Liu
Corrected comment #7 for urine GBS sensitivity reporting to match wording of other non-sterile sites comment and LIS canned message. "This organism is intrinsically susceptible to penicillin. If treatment is required AND this patient cannot be treated with penicillin, please contact the Microbiology Department within 48 hours to request sensitivity testing.	January 22, 2024	Jessica Bourke
Added Beta-Lactamase Testing (Cefinase disk) for persistent Enterococcus faecalis positive from the same steril site (eg. Blood or CSF or the like) over the span of >=five days under WHAT TO	July 24, 2024	Oliver Li

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TEST-> *Enterococcus* species (page 12)		
Updated sensitivity setup for What to do section of *Streptococcus anginosus* group and small colony-ß-haemolytic *Streptococcus* for blood and sterile sites	September 09,2024	Oliver Li
Added the sensi setup needed if bone/joint in Blood & sterile sites for Streptococcus anginosus group and Viridans Streptococcus (page 14) Updated what to report section accordingly (page 36)	October 01, 2024	Oliver Li
Added "Write the date 3 months from the first opening as the new expiry date on R1 and reconstituted R2 reagents. " in section of "**XVII – *B*CARBA Test**" on page 144.	Feburary 11, 2025	Oliver Li
Updated: Using reflected light to read linezolid KB for Staph spp.- page 78	Feburary 12, 2025	Qin Liu
Burkohlderia cepacia complex: Removed all disk disffusions from B. cepacia; Send B.cepacia to PHOL for MIC		
Stenotrophomonas maltophilia: Removed ceftazidime as antibiotic to be tested and reported from S. maltophilia Added the following new comment as isolate comment "*If treatment is deemed clinically warranted for S. maltophilia, combination therapy with two effective antimicrobials is recommended until clinical improvement is observed.*" Updated the breakpoints of Minocycline in LIS		
Pseudomonas aeruginosa: Removed gentamicin as antibiotic to be tested and reported from P.aeruginosa		
Appendix B. Agents never to be reported by site: Added moxifloxacin should never be reported for urine specimens. Added agents should never be reported for pediatric and pregnancy patients.		
Removed ticarcillin/clavulanic acid from entire antimicrobial susceptibility manual		
Removed tigecycline as antibiotic to be tested and reported rom Urine Specimens.		
Updated the age group for reporting tetracycline use from > 13 ys to > 7 ys		
Updated **What to Test Table**: Additional Testing Notes for Enterobacterales (SPICE/non-SPICE): Added new kb panel of kbxeru including both FOS and DO Remove TGC and/or MH from unrine site Remove LVX and MH from CSF/VP Shunt &brain Change from temporary to definitive testing of stool isolates for ampicillin (KB), 173ipro (etest) , septra (KB), ceftriaxone (KB))	Feburary 14, 2025	Qin Liu

Page Number / Item	Date of Revision	Edited by:
Updated in **WHAT TO REPORT**: Changed the tile from Spinal Fluids to Spinal Fluids, including VP shunts and Brain Tissue Changed the title from Blood and other Sterile sites to Blood and other Sterile Sites, excluding Spinal Fluids/VP Shunts or Brain Tissue		
Stenotrophomonas maltophilia from urine site-page 21: Added the following new comment as isolate comment "*If treatment is deemed clinically warranted for S. maltophilia, combination therapy with two effective antimicrobials is recommended until clinical improvement is observed.*"	Feburary 17, 2025	Qin Liu
Updated Salmonella and shigella: **Enteric sites:** For Salmonella (Typhi – all; non-Typhi – upon microbiologist approval) and Shigella (all), set up and report): Ampicillin Ciprofloxacin (Etest) TMP-SMX Azithromycin (exception: exclude for Salmonella non-Typhi) If resistant to all of the above, set up and report: Ceftriaxone Ertapenem Meropenem Doxycycline **Non-enteric sites:** Salmonella and Shigella set up and report: Ampicillin Ceftriaxone do not report Ciprofloxacin (Etest) TMP-SMX Azithromycin (exception: exclude for Salmonella non-Typhi) (except CSF/Urine) If resistant to ceftriaxone, then set up and report- Ertapenem (except for CSF) Meropenem Doxycycline (except for CSF)	March 12, 2025	Qin Liu
Levofloxacin Testing Adjustments for Bone/Joint Isolates: • Enterococcus: Changed Levofloxacin Etest → Kirby-Bauer (KB) • Anginosus group: Levo KB already set up; removed Levo Etest • Viridans group: Changed Levofloxacin Etest → Kirby-Bauer (KB)		

Page Number / Item	Date of Revision	Edited by:
• Beta-hemolytic Streptococcus: Added Levofloxacin and Tetracycline KB Updated Levofloxacin comments to align with KB for Enterococcus isolated from bone/joints. Updated doxycycline comment for Viridans group, S. anginosus group, and beta-hemolytic streptococci from bone/joint		
Appendex B-Agents Never To Be reported By Site: Changed Respiratory Specimens to BAL/ETT/sputum/ lung biopsy	March 12, 2025	Qin Liu
Removed macro e-test from enterococcus spp.-page13 Minor format in revision section Added to S.maltophilia: If I/R to all other drugs, set up and report : • Cefiderocol • Ceftazidime-aztreonam combination result based on the diffusion between these two disks Add Ceftazidime-Avibactam and Aztreonam Combination Test Procedure – page 95 Revised appendix numbering (Roman numerals) to include newly added Ceftazidime-Avibactam and Aztreonam Combination Test-page 95	March 21, 2025	Qin Liu
Report levofloxacin for *BHS* isolated from bone and joint specimens–page 40 Report vancomycin for *S. pneumoniae* isolated from blood cultures and other sterile sites **only if penicillin is I/R –** page 38		
• Updated the CPO flowchart for both IC and Non-IC workups: set up CARBR for BCARB-/ROSCO+ samples; report as positive if CARBR is positive; send BCARB-/ROSCO+/CARBR- samples to PHOL for genotypic confirmation.. • Removed CPO isolate comment (\CNML): ~This organism is negative by the βCARBA test (Bio-Rad) ~but phenotypic testing based on the KPC+MBL+OXA48 ~Confirm Kit inhibitor tablets (ROSCO) cannot rule out ~carbapenemase production. ~Genotypic confirmation from the National Microbiology ~Laboratory to follow. • Added CPO isolate comment(\CPHL): ~Phenotypic testing cannot rule out carbapenemase ~production. Genotypic confirmation from Public ~Health Ontario Laboratory to follow.		
Updated the additional testing notes for MDR Enterobacerales-page 7		

Page Number / Item	Date of Revision	Edited by:
• Updated to set up Clindamycin and Erythromycin double disks routinely for viridans • Delete footnote" Report with isolate comment \icr- "If clindamycin susceptibility testing is required, please contact the microbiology laboratory within 48 hours." For Blood and other Sterile Sites reporting. • Re-added "Tetracycline" to the list of reportable antibiotics for Enterococcus isolated from urine. It was mistakenly removed in the previous version. • APPENDIX E. Annual CLSI Updates Implementation		
• Re-added VRE extra anitbiotics (LZD,DPC-excluding respiratory)section that was mistakenly remoed in the previous version. • Removed DPC for Enterococcus from all sites if the isolate is resistant to all routinely tested antimicrobials or if requested as a duplicate test • Removed TGC for Enterococcus from all sites if the isolate is resistant to all routinely tested antimicrobials or if requested to match lab practice.	April 23, 2025	Qin Liu
Updated What to set up section: • Added plain MH agar for setting up the following antimicrobials: Minocycline Chloramphenicol Cefiderocol • Updated the panel name from kbxeru to kbfodo for additional oral drug for urines • Added the panel name of kbxdru for extreme drug resistant urine isolates • Added additional antimicrobials for extreme drug resistant pseudomonas aeruginosa	May 27, 2025	Qin Liu
• For Enterococci and streptococcus species – added testing and reporting for doxy and levo in sterile sites • Strep. Pneumo – removed set up for clinda and erythro • Updated cefiderocol reporting comments	June 27, 2025	Karin Schoer