Department of Microbiology Aerobic Staphylococci Work-up



Gross Appearance	Body Site			Preliminary Report	Set Up	Results	•	Final Report	Billing
	All sites when indicated by Q score for work-up or MRSA screen.	+	_	S. aureus	Phoenix AST		_	S. aureus & AST *see Note 1	URB1 or ORGB1 ASB1
	Multiple sets of blood cultures positive with staph or	-	_	Staph species	Phoenix AST KB FOX & SXT PYR: if + then set up tube coag (TC),	PYR +	TC – ORN + BASE –	Rounds consultation S. lugdunensis & AST Report Phoenix OX/FOX result Enter SXT in Epicenter	ORGB1 ASB1 KBB1
	Normally sterile body sites with staph being the sole isolate.				ornithine, and base decarboxylase	PYR –		Staph coag-neg & AST (enter FOX & SXT in Epicenter)	ORGB1 ASB1 KBB1
	Sterile body site, staph not seen in direct smear, and culture is mixed	_	_	Staph coag-neg				Staph coag-neg No AST	ORGB1
	Nonsterile body site and staph is seen in direct smear and no <i>S. aureus</i>	_	_	Staph coag-neg	Phoenix AST KB FOX & SXT PYR: if + then set	PYR +	TC – ORN + BASE –	S. lugdunensis & AST Report Phoenix OX/FOX result Enter SXT in Epicenter	ORGB1 ASB1 KBB1
	grows in culture				up tube coag (TC), ornithine, and base decarboxylase	PYR –		Staph coag-neg & AST (enter FOX & SXT in Epicenter)	ORGB1 ASB1 KBB1
	Cath tip > 15 col	_	_	Staph coag-neg				Staph coag-neg & [C15] No AST	ORGB1
	Cath tip < 16 col	-	_	Staph coag-neg				Staph coag-neg & [C14] No AST	ORGB1
CHROM = light pink to rose	Urine significant count	_	_	S. sapro	Phoenix AST KB FOX & SXT			S. sapro & AST (enter FOX & SXT in Epicenter)	URB1 ASB1 KBB1
CHROM = cream	Urine significant count	_	_	Staph coag-neg	Phoenix AST KB FOX & SXT			Staph coag-neg & AST (enter FOX & SXT in Epicenter)	URB1 KBB1 ASB1
Bright yellow	All sites	-	_	Staph species	TC (tube coag) smear OF Glucose	TC – tetrads oxidizer	/nonoxidizer	Micrococcus	ORGB1
*NIA A NADOA		-::6			MDOA - I	fermente	er	Staph coag-neg	ORGB1

*Note 1 MRSA is automatically flagged as a significant isolate in the computer. MRSA should be called to inpatient unit and faxed to Epidemiology.

MRSA isolates with a VA MIC \geq 4 should be brought up on Rounds. For *S. aureus* isolates with a VA MIC = 2 or 4, perform E-test confirmation. If E-test MIC = 2 or 4, consult Rounds. MIC values of 2 or 4 should be reported.

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Department of Microbiology Aerobic Streptococci Work-up (α-hemolytic)

Body Site	Test	Results	Preliminary Results	Set Up	Results	Final Report	Billing
Respiratory	Bile Solubility	+	Strep pneumoniae	Phoenix AST		S. pneumoniae & AST (enter Pen MIC value in LIS under oral and parenteral) Mixed flora	ORGB1 ASB1
		?		Optochin (P disk) & Phoenix AST	P disk zone ≥14 mm	S. pneumoniae & AST (enter Pen MIC value in LIS under oral and parenteral)	ORGB1 ASB1
Bile Solubility – En	Bile Solubility	+	Strep pneumoniae	Phoenix AST	P disk zone < 14 mm	Mixed Flora S. pneumoniae & AST (enter Pen MIC value in LIS under oral and parenteral)	ORGB1 ASB1
	Enterococcus	If site is a normally sterile body fluid, set up 6.5% NaCl, Control Broth, BE, Phoenix AST	Broth control + 6.5% NaCl + BE + Neg control broth, NaCl, or BE = not Enterococcus	Enterococcus & AST *see Note 2 Rounds consultation	Urine: URB1 Other: ORGB1 AST: ASB1		
				If site is a normally sterile body fluid, set up 6.5% NaCl, Control Broth, BE. Perform Phoenix AST if normally sterile body site or if seen in direct smear and within Q score of	Broth control + 6.5% NaCl - BE - Broth control + 6.5% NaCl + BE - Broth control + 6.5% NaCl - Broth control + 6.5% NaCl - BE +	Viridans Strep & AST if applicable Rounds consultation Viridans Strep & AST if applicable Rounds consultation	URB1/ORGB1 & ASB1 if applicable URB1/ORGB1 & ASB1 if applicable
				non-sterile site.	6.5% NaCl + BE +	Rounds consultation	

^{*}Note 2: For isolates identified as VRE, perform additional testing to rule out *E. gallinarum* or *E. casseliflavus/flavescens*. These species are intrinsically resistant to vancomycin and not associated with epidemiologic outbreaks. These organisms typically produce vancomycin MIC values between 2 and 16 µg/mL. They are generally motile and most isolates of *E. casseliflavus/flavescens* produce a distinct yellow pigment, which can be observed by collecting growth on a swab. Consult on Rounds. VRE is automatically flagged as a significant isolate in the computer. If isolate ID is *E. faecalis/faecium*, call VRE to inpatient unit.

Department of Microbiology Aerobic Streptococci Work-up (β-hemolytic)



Body Site	Test	Results	Preliminary Results	Set Up	Results	Final Report	Billing
Respiratory	Group A latex	+				Group A Strep AST comment [BSAS]	ORGB1
		_				Mixed flora	
Throat	Group A latex	+				β-Strep Group A isolated & AST comment [BSAS]	ORGB1
		_				No Group A Strep isolated	
Genital G	Group B latex	+		KB if Pen allergic		Group B Strep isolated & AST comment [BTSBS] AST if pen allergic	ORGB1 KBB1 if done for Pen allergic
		_				No Group B Strep isolated	
All other sites	A & B latex PYR if latex –	A +				Group A Strep & AST comment [BSAS]	Urine: URB1 Other: ORGB1
		B +				Group B Strep & AST comment [BTSBS]	Urine: URB1 Other: ORGB1
		A – B – PYR +	Enterococcus	If site is a normally sterile body fluid: Control Broth	Broth control + 6.5% NaCl + BE +	*see Note 2	Urine: URB1 Other: ORGB1 AST: ASB1
		PIRT		6.5% NaCl BE AST	Neg control broth, NaCl, or BE = not <i>Enterococcus</i>	Rounds consultation	
		A – B –	β-hemolytic strep not group A or B	If site is a normally sterile body fluid: 6.5% NaCl	Broth control + 6.5% NaCl – BE –	Beta-hemolytic strep not group A or B	Urine: URB1 Other: ORGB1
		PYR –		Control Broth BE	Broth control + NaCl + / BE – or NaCl – / BE +	Rounds consultation	

*Note 2: For isolates identified as VRE, perform additional testing to rule out *E. gallinarum* or *E. casseliflavus/flavescens*. These species are intrinsically resistant to vancomycin and not associated with epidemiologic outbreaks. These organisms typically produce vancomycin MIC values between 2 and 16 µg/mL. They are generally motile and most isolates of *E. casseliflavus/flavescens* produce a distinct yellow pigment, which can be observed by collecting growth on a swab. Consult on Rounds. VRE is automatically flagged as a significant isolate in the computer. If isolate ID is *E. faecalis/faecium*, call VRE to inpatient unit.

Department of Microbiology Aerobic Streptococci Work-up (non-hemolytic)



Body Site	Test	Results	Preliminary Results	Set Up	Results	Final Report	Billing
Genital	Group B latex	+		KB if Pen allergic		Group B Strep isolated & AST comment [BTSBS] AST if pen allergic	ORGB1 KBB1 if done for pen allergic
		_				No Group B Strep isolated	
Other body sites except respiratory	PYR	+	Enterococcus	If site is a normally sterile body fluid: Control Broth	Broth control + 6.5% NaCl + BE +	Enterococcus & AST *see Note 2	Urine: URB1 Other: ORGB1 AST: ASB1
, ,				6.5% NaCl BE AST	Neg control broth, NaCl, or BE = not Enterococcus	Rounds consultation	
		_	Non-hemolytic strep	If site is a normally sterile body fluid, set up Control Broth, 6.5%	Broth control + 6.5% NaCl – BE –	Viridans Strep & AST if applicable	URB1/ORGB1 & ASB1 if applicable
				NaCl, BE. Perform AST if normally	Broth control + 6.5% NaCl + BE –	Rounds consultation	
				sterile body site <u>or</u> if seen in direct smear and within Q score of sterile site.	Broth control + 6.5% NaCl – BE +	Viridans Strep & AST if applicable	URB1/ORGB1 & ASB1 if applicable
		_	Non-hemolytic strep	Looks like Group B strep colony	B+	Group B Strep & AST comment [BTSBS]	Urine: URB1 Other: ORGB1
				If site is a urine, set up Group B latex	B –	Viridans Strep	Urine: URB1 Other: ORGB1
		_		If site is any other non sterile body site		Viridans Strep	Urine: URB1 Other: ORGB1

^{*}Note 2: For isolates identified as VRE, perform additional testing to rule out *E. gallinarum* or *E. casseliflavus/flavescens*. These species are intrinsically resistant to vancomycin and not associated with epidemiologic outbreaks. These organisms typically produce vancomycin MIC values between 2 and 16 µg/mL. They are generally motile and most isolates of *E. casseliflavus/flavescens* produce a distinct yellow pigment, which can be observed by collecting growth on a swab. Consult on Rounds. VRE is automatically flagged as a significant isolate in the computer. If isolate ID is *E. faecalis/faecium*, call VRE to inpatient unit.

Department of Microbiology Neisseria species Work-up



Gross Appearance	Body Site	Oxidase	Gram Stain	CarboFerm	Growth on MTM	Final Report	Billing
Small, grey translucent	Genital Vaginal	_				No N. gonorrhoeae isolated	
	Cervical Urethral	+	GNR			No N. gonorrhoeae isolated	
		+	GNDC	More than just GLUC +	+	Rounds consultation	
		+	GNDC	Glucose + Maltose - Lactose - Sucrose -	+	Rounds consultation N. gonorrhoeae Report to DOH	ORGB1
	+	GNDC	Glucose + Maltose - Lactose - Sucrose -	_	Rounds Consultation		
	Non-genital	_	GNDC		+/-	Rounds Consultation	
		+	GNR			Perform NFID	
		+	GNDC	Glucose + Maltose - Lactose - Sucrose -	+/-	Rounds consultation N. gonorrhoeae Report to DOH	ORGB1
		+	GNDC	Glucose + Maltose + Lactose – Sucrose –	+	N. meningitidis Rounds consultation Call patient care area, physician, and epidemiology. Send isolates from sterile sites to DOH.	ORGB1
		+	GNDC	Any other combination other than above	+/-	Rounds Consultation	

Department of Microbiology Moraxella species Work-up



Gross Appearance	Body Site	Oxidas e	Gram Stain	Other Tests	Results	Report	Billing	
Dry, wrinkled yellow or white	Respiratory	+	GNDC	Catarrhalis Disk	+	M. catarrhalis & AST comment [MCATAS]	ORGB 1	
		+	GNDC	Catarrhalis Disk	-	Mixed flora		
	Other sites	+	GNDC?	Penicillin disk elongation test	Cell elongation at P disk	Rounds Consultation		
		+	GNDC	CarboFerm	Glucose – Maltose – Lactose – Sucrose – Butyrate +	Rounds Consultation M. catarrhalis & AST comment [MCATAS]	ORGB 1	
Smooth transparent	All sites	+	GNCB	If catalase + and non-motile set up: OFGL		Rounds Consultation for other <i>Moraxella</i> spp.		

Department of Microbiology Haemophilus species Work-up



Gross Appearance	Site	Gram Stain from CHOC	Set Up	Growth on BAP	XV	AST	Final Report	Billing
Small transparent colonies satelliting around staph streak	Non-sterile sites	tiny pleomorphic GNR	XV disks cefinase	_	Both required	Cefinase –	H. influenzae Beta lactamase negative & [HFLUS]	ORGB1
or mucoid grey colonies on CHOC				_	Both required	Cefinase +	H. influenzae Beta-lactamase positive & [HFLUS]	ORGB1
				_	Both not required		Haemophilus species not influenzae	ORGB1
				+			Rounds Consultation	
	Sterile sites	tiny pleomorphic GNR	XV disks cefinase	_	Both required	Cefinase – Set up KB on HTM	H. influenzae Beta-lactamase negative & AST	ORGB1 KBB1
				_	Both required	Cefinase + Set up KB on HTM	H. influenzae Beta-lactamase positive & AST	ORGB1 KBB1
				_	Both not required		Rounds Consultation Haemophilus sp. not influenzae	ORGB1
				+			Rounds Consultation	

Cultures for Haemophilus ducreyi

- A. Gram stain
 - 1. Gram negative pleomorphic coccobacilli with many single chains representing "schools of fish".
- B. Culture
 - 1. Examine plates incubated in CO₂ at day 2 and day 7.
 - 2. Gram stain any growth on CHOC.
 - 3. Bring up culture on Rounds.

Department of Microbiology Aerobic Gram-Negative Rods Work-up (Other Than *Haemophilus* species)



Gross Appearance	Oxidas	Indole	Preliminary	Set Up	Final Report	Billing
BAP, and/or MAC	е	muole	Report	-	•	
MAC = lactose positive flat, donut-shaped	_	+	E. coli & report direct AST results	Phoenix AST	E. coli & Phoenix AST	Urine: URB1 Other: ORGB1
MAC = lactose positive	_	_	for Blood isolates Coliform & report direct AST results for Blood isolates	Phoenix ID/AST	Report Phoenix ID and AST	AST: ASB1 *See Note 3
MAC = lactose negative	+	+	GNR	Phoenix ID/AST	Report ID and AST may require Rounds consultation	
BAP = swarming MAC = lactose negative	-	-	Proteus species	Phoenix AST	Proteus sp. With AST	
BAP = swarming MAC = lactose negative	_	+	Proteus vulgaris	Phoenix AST	Proteus vulgaris & AST	
BAP = greenish pigment MAC = lactose negative	+		Pseudomonas aeruginosa	Phoenix AST KB for CF isolates	Pseudomonas aeruginosa with AST	
BAP = metallic sheen or grape-like odor MAC = lactose negative	+		Pseudomonas aeruginosa	Phoenix AST KB for CF isolates	Pseudomonas aeruginosa with AST	
MAC = lactose negative	+		Gram negative rod	Phoenix ID/AST or NF ID and AST	Report ID and AST May require Rounds consultation CF isolates identified as <i>B. cepacia</i> must be confirmed by tube ARG and LYS <i>B. cepacia</i> = ARG –, LYS +	
MAC = lactose negative Purplish colonies	-		Gram negative rod	Phoenix ID/AST If GNCB & non- motile and PHX fails to yield an ID set up OFGL & nitrate	Report ID & AST from Phoenix If offline ID, consult Rounds for reporting Acinetobacter species	
MAC = lactose negative	_		Gram negative rod	Phoenix ID/AST	Report ID and AST	
BAP = brick red pigment MAC = lact neg but red	_		Serratia marcescens	Phoenix AST	Serratia marcescens & AST	
BAP = growth MAC = no growth	+ or –		Gram negative rod	Phoenix ID/AST or NF ID and AST	Rounds consultation Report ID and AST	

^{*}Note 3 If confirming ESBL or KPC for *Enterobacteriaceae* by disk diffusion, bill KBB1.

Department of Microbiology Campylobacter Work-up



Bring up on Rounds Notify patient care area, Physician and Epidemiology

Oxidase & Catalase Positive Growth on Campylobacter media at 42°C Gram stain Gram Neg Rods Gram Neg Rods-Faint Staining Not curved or Campy-like (short, curved, S-shaped, coccoid) Transfer to MAC at 35°C Wet prep Growth No Growth (Motility) Consult supervisor Report: Not typical No Campylobacter sp. Darting isolated (not darting) Consult Supervisor Report: Campylobacter sp. **Bill ORGB1**

Helicobacter pylori

Gram stain: slender spirally curved gram-negative rods

- 1. Examine plates incubated in a Campy environment at 35°C at day 3 and day 7.
- 2. Gram stain any growth on the plates.
- 3. Bring up culture on Rounds.

Oxidase: positive

Urea: positive (very rapid)

Department of Microbiology Aerobic Gram-Positive Rods Work-up



Gross Appearance	Hemolysis	Gram stain of plate	Motility	Catalase	Prelim. Report	Set-Up	Results	Final Report	Billing
BAP									
Large, flat, dull, rough, gray, "medusa head"	_	Uniform large gram positive rods with squared off ends	-	+	Rounds consultation				
Large, flat, dull, rough gray green, ground glass	β	Uniform large gram positive rods with squared off ends	+/-	+	Bacillus species	If ? Gram +/- set AST To help induce spore production, inoculate urea, CHROMagar, and reincubate older plates. Spore stain	Formation of spores	Bacillus species not anthracis	ORGB1
Large, flat, dull, smooth, gray	_	Uniform large gram positive rods (may be gram negative)	+	+	Bacillus species	If ? Gram +/– set AST To help induce spore production, inoculate urea, CHROMagar,	Formation of spores	Bacillus species not anthracis	ORGB1
						and reincubate older plates. Spore stain	No spores	Rounds consultation	
Small sometimes dry dull, white opaque	_	Short gram positive rods (diphtheroids) in palisading arrangements	-	+	Corynebacteriu m species	If sterile site or multiple sets of Blood Cx, set up Etests. Set up API CORYNE for ID only if requested.		Corynebacterium species Rounds if API set	ORGB1 MICB for E-test
Urine isolate: Pinpoint, convex, smooth, and whitish gray	_	Short gram positive rods (diphtheroids) in palisading arrangements	-	+		For Urine isolates: Urea agar	Rapid positive (< 2 h)	Corynebacterium urealyticum	URB1
Very small colonies which	– or α	Gram positive coccobacilli (may be	_	_	Gram positive rod	TSI	H ₂ S +	Erysipelothrix rhusiopathiae	ORGB1
may require 48 h for growth		diphtheroids)					H ₂ S –	Rounds consultation	
Small flat white or gray	β	Gram positive rods (short rods to cocci)	Tumblin g at 22°C Slow or	+	Gram positive rod	BE	+	Rounds Listeria monocytogenes Report to DOH	ORGB1
			– at 35°C				_	Rounds consult	
Very small opaque	α	Long thin gram positive rods	-	_		Vancomycin disk	R	Lactobacillus spp. & [LACBSS]	URB1

Department of Microbiology Aerobic Gram-Positive Rods Work-up



(consult Rounds				S	Lactobacillus-like	No ID
for non-urine					species	charge
isolates)						

References

- A. Finegold, S.M., Baron, E.J. 1986. Bailey and Scott's Diagnostic Microbiology, 7th ed., C.V. Mosby Co., St. Louis, pp. 404, 525.
- B. Murray, P.R., Baron, E.J., Pfaller, M.A., Tenover, F.C., Yolken, R.H. 1999. Manual of Clinical Microbiology, 7th ed., ASM Press, Washington, DC, pp. 272, 288, 298, 319-341, 349, 362, 443-446, 459-474, 502, 512, 518-519, 544, 595, 608, 717.

Document Control History

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History of Changes: 06/25/2012 Removed acetate from *Moraxella* spp. workup. 10/10/2013 Added comment for S. aureus, "For *S. aureus* isolates with a VA MIC = 2 or 4, perform E-test confirmation. If E-test MIC = 2 or 4, consult Rounds. MIC values of 2 or 4 should be reported." Updated billing code MICB1 to ASB1 for applicable organisms tested on the Phoenix instrument. Added, "or Latex" to the slide coag column on Staphylococci work-up. Updated workup for *Corynebacterium* to perform AST if from sterile site or multiple blood culture sets. 05/08/2014 Added workup for *C. urealyticum* urine isolates. 06/24/2014 Added Vancomycin Disk Test for identification of Lactobacillus isolates from urine cultures and added susceptibility comment. 04/30/2015 Updated for Viridans strep... "Perform Phoenix AST if normally sterile body site or if seen in direct smear and within Q score of non-sterile site."