

## Enterococci

<b>SPECIMEN</b>	Blood, wounds and sterile sites (tissues, peritoneal fluid), urines (urosepsis)		
<b>PROCEDURE</b> Enterococci (general)	<b>Agar medium</b>	Mueller Hinton agar	
	<b>Inoculum</b>	Suspension in saline to 0.5 McFarland turbidity	
	<b>Incubation</b>	35°C/ ambient/ 16-20h; always read vancomycin at 24h	
	<b>Reading</b>	For bactericidal drugs, read at complete inhibition of all growth including microcolonies, hazes and isolated colonies. For bacteriostatic drugs, read at 80% inhibition when trailing is seen.	
<b>HLAR</b> High Level Aminoglycoside Resistance	<b>Agar medium</b>	Mueller Hinton agar	
	<b>Etest range</b>	Gentamicin and streptomycin 0.064-1024 µg/ mL	
	<b>Inoculum</b>	Suspension in saline to 0.5-1 McFarland (heavier inoculum preferable)	
	<b>Incubation</b>	35°C/ ambient/ 24h (always confirm streptomycin at 48 hours)	
	<b>Reading</b>	Read at complete inhibition including microcolonies, hazes and isolated colonies.	
<b>VRE</b> Vancomycin Resistant Enterococci	<b>Agar medium</b>	<b>In the USA, this modified procedure is for Investigational Use Only.</b> Brain Heart Infusion agar (BHI)	
	<b>Inoculum</b>	Suspension in broth to 2 McFarland	
	<b>Inoculation</b>	Dispense 0.1 mL per 90 mm agar plate and streak evenly.	
	<b>Incubation</b>	35°C/ ambient/ 24-48h; always confirm at 48h.	
	<b>Reading</b>	Look for hazes, microcolonies and isolated colonies. Use a magnifying glass, oblique light and/or tilt the plate to look for all growth.	

**LITERATURE** Package insert, product labels, Customer Information Sheet 002 and 007, Etest Technical Guide, Media/ Inoculum/ Incubation, Reading & Trouble Shooting charts and Etest reference list.

QUALITY CONTROL (MIC µg/ml)	<i>E. faecalis</i> ATCC® 29212			
	AM	LE	LZ	QDA
Ampicillin	0.5-2	Teicoplanin <sup>1)</sup>	TP	0.125-0.5
Levofloxacin	0.25-2	Tetracycline	TC	8-32
Linezolid	1-4	Vancomycin	VA	1-4
Quinupristin/ Dalfopristin	2-8			

HLAR	<i>E. faecalis</i> ATCC® 29212		VRE (BHI) <sup>1)2)</sup>	<i>E. faecalis</i> ATCC® 29212
	GM	SM		
Gentamicin	4-16	Teicoplanin <sup>1)</sup>	TP	0.25-1
Streptomycin <sup>1)</sup>	64-256	Vancomycin	VA	2-6

INTERPRETATION CLSI MIC Criteria (µg/ml) <sup>3)</sup>	S I R			S I R		
	AM	LE	LZ	TP	TC	VA
Ampicillin	≤8	-	≥16	Teicoplanin <sup>1)</sup>	≤8	16 ≥32
Levofloxacin	≤2	4	≥8	Tetracycline	≤4	8 ≥16
Linezolid	≤2	4	≥8	Vancomycin	≤4	8-16 ≥32
Quinupristin/ Dalfopristin	≤1	2	≥4			

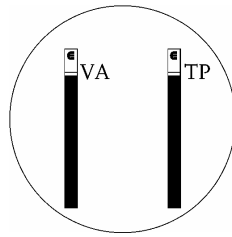
Phenotype Interpretation	HLAR	
	Negative	Positive
Gentamicin	≤512	>512
Streptomycin <sup>1)</sup>	≤1024	>1024

VRE (BHI) Phenotype <sup>1)2)</sup>	Vancomycin (µg/ml)	Teicoplanin (µg/ml)	Species
vanA	≥32 (R)	and ≥16 (I-R)	<i>E. faecalis</i> <i>E. faecium</i>
vanB	≥8-256 (I-R)	and ≤4 (S)	<i>E. faecalis</i> <i>E. faecium</i>
vanC1	4-16 (S-I)	and ≤4 (S)	<i>E. gallinarum</i>
vanC2	4-16 (S-I)	and ≤4 (S)	<i>E. casseliflavus</i> <i>E. flavescens</i>
vanD	64 (R)	and ≤4 (S)	<i>E. faecium</i>
vanE	16 (I)	and ≤4 (S)	<i>E. faecalis</i>

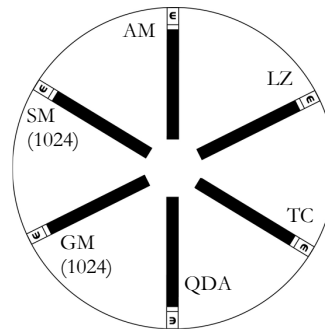
### Notes

1. In the USA, for Investigational Use Only.
2. Dogson, K. (2004). VRE detection: a new gold standard. *Clinical Microbiology Newsletter*. 26 (4):25-30. VRE phenotype interpretation is based on currently available literature.
3. Please refer to CLSI M100- S (latest version).

## ANTIBIOGRAMS



**Panel A**  
Detection of VRE (BHI)



**Panel B**  
Other antibiotics and  
HLAR (Mueller Hinton)  
Observe: QDA for *E. faecium* only.

Examples only, please use your own formulary.

## REFERENCES

1. Schulz *et al.* (1993). Reliability of Etest for detection of ampicillin, vancomycin, and high-level aminoglycoside resistance in *Enterococcus* spp. *JCM*. 31(12): 3336- 3339.
2. Jones *et al.* (1995). Emerging multiply resistant enterococci among clincial isolates: II. Validation of the Etest to recognize glycopeptide-resistant strains. *DMID*. 21: 95- 100.
3. Tenover *et al.* (1995). Ability of commercial and reference antimicrobial susceptibility testing methods to detect vancomycin resistance in enterococci. *JCM*. 33(6): 1524-1527.
4. Yamane *et al.* (1997). Laboratory evaluation of antimicrobial susceptibility testing to detect vancomycin resistant enterococci. *Japanese J. Clin. Path.* 45(4): 381- 391.
5. Kohner *et al.* (1997). Comparison of agar dilution, broth microdilution, Etest, disk diffusion and automated Vitek methods for testing susceptibilities of *Enterococcus* spp. to vancomycin. *JCM*. 35(12): 3258- 3263.
6. Endtz *et al.* (1998). Comparison of eight methods to detect vancomycin resistance in enterococci. *JCM*. 36(2): 592- 594.
7. Bolmström *et al.* (1998). Phenotyping of vancomycin-resistant enterococci (VRE) using Etest under different conditions. *ICAAC*, poster no. D-57.