

Multi-Site Evaluation of Dalbavancin and Vancomycin MIC Test Strip Compared To Broth Microdilution MICs

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Abstract:

Background: Dalbavancin and vancomycin are antibiotics used for treatment of acute bacterial skin and skin structure infections caused by Gram-positive organisms. This study was performed to evaluate the performance of the dalbavancin and vancomycin MIC test strips (MTS) from Liofilchem (Roseto degli Abruzzi, Italy) compared to a broth microdilution method (BMD) for a FDA 510(k) submission. **Methods:** Clinical and challenge isolates were tested by BMD with frozen panels and by MTS. For dalbavancin, 311 *S. aureus* and 319 *E. faecalis* from recent clinical sources were collected and tested at 3 sites, 76 *S. aureus*, 39 *E. faecalis* and 37 *E. faecium* challenge isolates were tested at 1 site, and for reproducibility 10 *S. aureus* and 10 *E. faecalis* were tested 10 times at 3 sites. For vancomycin, 312 *S. aureus*, 20 *S. epidermidis*, 378 *E. faecalis*, and 62 *E. faecium* from recent clinical sources were collected and tested at 3 sites, 76 challenge isolates (41 *S. aureus*, 5 *S. epidermidis*, 11 *E. faecalis* and 24 *E. faecium*) were tested at 1 site, and for reproducibility 10 isolates were tested 10 times at 3 sites. QC strains (*S. aureus* ATCC 29213, *E. faecalis* ATCC 29212) were tested a minimum of 20 times by each site. **Results:** As shown in the table, dalbavancin and vancomycin MTS MIC results for consolidated clinical and challenge organisms were within +/- one doubling dilution (essential agreement) of BMD MIC results for all isolates with only one exception

Agent	Organism	N	% Essential Agreement	% Category Agreement
Dalbavancin	<i>S. aureus</i>	387	100%	99.7%
Dalbavancin	<i>E. faecalis</i>	319	99.7	100%
Vancomycin	<i>S. aureus</i>	353	99.7	98.6
Vancomycin	<i>S. epidermidis</i>	65	100	98.5
Vancomycin	<i>E. faecalis</i>	389	100	99.7
Vancomycin	<i>E. faecium</i>	81	100	97.5

Conclusions: The dalbavancin MTS against *S. aureus* and *E. faecalis* and the vancomycin MTS against *S. aureus*, *S. epidermidis*, *E. faecalis* and *E. faecium* performs similar to the reference broth microdilution method.

Introduction

- Liofilchem (Roseto degli Abruzzi, Italy) manufactures MIC test strips (MTS) for a variety of antimicrobial agents, including vancomycin and recently developed a strip for dalbavancin. The Liofilchem MIC test strip is a quantitative agar-based diffusion assay for determining the minimum inhibitory concentration (MIC).
- This study was performed as part of a 510(k) study (for "in vitro diagnostic use" label in the U.S.)
- This study compared the dalbavancin MTS MIC to broth microdilution MIC for the indicated Gram positive organisms: *S. aureus* and *E. faecalis*
- This study compared the vancomycin MTS MIC to broth microdilution MIC for the FDA indicated Gram positive organisms: *S. aureus*, *S. epidermidis*, *E. faecalis* and *E. faecium*

References:

- Clinical and Laboratory Standards Institute. 2015. Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria that Grow Aerobically. 10th ed. Approved standard, CLSI M7-10, Wayne, PA.
- Clinical and Laboratory Standards Institute. 2016 Performance Standards for Antimicrobial Susceptibility Testing. Approved Standard - 26th Edition. CLSI document M100-26 Wayne, PA
- http://www.liofilchem.net/en/mov_mic_test_strip.php

Methods

Study Strains (Clinical isolates collected at three sites and challenge isolates)

Organism Group	DALBAVANCIN		VANCOMYCIN	
	Clinical	Challenge	Clinical	Challenge
<i>Staphylococcus aureus</i>	311	76	312	41
MRSA	155	66 ⁽¹⁾	156	39 ⁽²⁾
MSSA	156	10 ⁽³⁾	156	2
<i>Staphylococcus epidermidis</i>	NA	NA	60	5
<i>Enterococcus faecalis</i>	319	39	378	11
<i>Enterococcus faecium</i>	NA	37	62	19
TOTAL	630	152	812	76

¹53 VISA, 3 VRSA ²32 VISA, 3 VRSA ³5 VRSA

All clinical isolates were collected within one year of testing and at least 50% were tested within 7 days of collection. QC strains: *S. aureus* ATCC 29213 and *E. cloacae* ATCC 29212

Testing sites:

- Laboratory Specialists, Inc., Westlake, OH
- University of Rochester Medical Center, Rochester, NY
- Cardiff University, Cardiff, UK

MIC methods:

- Each isolate was tested once by broth microdilution according to CLSI method (1) with frozen panels containing dalbavancin and vancomycin concentrations of 0.002-8 µg/mL and 0.016-256 µg/mL respectively, and by dalbavancin (DAL) and vancomycin (VAN) MTS (3) containing concentrations of 0.002-32 µg/mL and 0.016-256 µg/mL respectively (Liofilchem, Roseto degli Abruzzi, Italy) on 100 mm Mueller Hinton Agar II plates (2 sites used MHA from Becton Dickinson [Sparks, MD] and one site used MHA from Oxoid [Basingstoke, UK]). Challenge isolate testing was performed by one site (Laboratory Specialists)
- Quality control strains were tested each day of testing and a total 20 replicates/site were tested.
- MTS results were rounded up to next doubling dilution for analysis. MIC results were interpreted according to FDA breakpoints



S. aureus
Dalbavancin MIC = 0.06 µg/mL

Figure 1. Dalbavancin MTS MIC compared to BMD MIC for 387 clinical and challenge *S. aureus* isolates*

MTS Results	BMD Reference Results											Evaluation					
	0.008	0.015	0.03	0.06	0.12	0.25 S	0.5 R	1	2	4	8		>8				
0.002																	
0.004																	
0.008	1																
0.015																	
0.03				5	1												
0.06				23	106	64											
0.12					27	116	1										
0.25 S						1	17	7									
0.5 R							3	3	2								
1								1	1	1							
2										2							
4											2						
8											1						
16																	
32																	
>32																	2

*221 MRSA, 166 MSSA

Figure 3. Vancomycin MTS MIC compared to BMD MIC for 353 *Staphylococcus aureus*

Test Results	Reference Results											Evaluation					
	0.25	0.5	1	2	4	8	16	32	64	128	256		>256				
0.25	1																
0.5		3	5														
1			8	209	40	1											
2				12	43	4											
4					9	6											
8						1	5										
16								3									
32									1								
64																	
128																	
256																	
>256																	2

*195 MRSA, 158 MSSA

Figure 2. Dalbavancin MTS MIC compared to BMD MIC for 395 *Enterococcus* spp.*

MTS Results	BMD Reference Results											Evaluation					
	0.008	0.015	0.03	0.06	0.12	0.25 S	0.5 R	1	2	4	8		>8				
0.002																	
0.004																	
0.008																	
0.015			2														
0.03				9	1	1											
0.06					109	147	8										
0.12					1	53	24										
0.25 S							2										
0.5 R								1									
1									1								
2										2							
4											1	2					
8													1				
16														2			
32																	
>32																	30

*358 *E. faecalis*, 37 *E. faecium*

Figure 4. Vancomycin MTS MIC compared to BMD MIC for 470 *Enterococcus* spp.*

Test Results	Reference Results											Evaluation					
	0.25	0.5	1	2	4	8	16	32	64	128	256		>256				
0.25																	
0.5		1	4	6													
1			10	118	36												
2				39	99	12											
4					21	31	1										
8						2	5	2									
16							2	2									
32									1	1							
64									1	2							
128																	
256													1	2			
>256														2	69		

*389 *E. faecalis*, 81 *E. faecium*



S. aureus
Vancomycin MIC = 1.5 µg/mL

Table 1. Dalbavancin and Vancomycin MTS Quality Control Results by Testing Site

QC Organism	Dalbavancin MTS					Vancomycin MTS					
	MIC (µg/mL)	Number of results			MIC (µg/mL)	MTS Frequency					
		Site 1	Site 2	Site 3		Site 1	Site 2	Site 3			
<i>S. aureus</i> ATCC 29213	0.015				0.25						
	0.03				0.5						1
	0.06	20	17	27	1	19	27	25			
	0.12		8	5	2	1					3
	0.25					4					
<i>E. faecalis</i> ATCC 29212	0.015				0.5						
	0.03				1						
	0.06	9	10	15	2	4					7
	0.12	11	15	17	4	16	25	22			
	0.25					8					

CLSI QC range

Conclusions

- The dalbavancin and vancomycin MTS against *S. aureus* and *Enterococcus* spp. performed similar to BMD testing.
- There was a tendency for one dilution higher dalbavancin MIC results for strains with BMD MIC results of 0.03-0.06 µg/mL.
- 86% of *S. aureus* with dalbavancin BMD MIC results of >0.12 µg/mL were challenge VISA and VRSA isolates.
- Although essential agreement was 100% for dalbavancin against *S. aureus*, based on a high prevalence of very major errors (7/11) for *S. aureus* with dalbavancin MTS MIC of 0.25 µg/mL and broth microdilution MIC of 0.5 µg/mL, additional replicate testing with these and additional strains with borderline MIC results is warranted.
- The vancomycin MTS was recently cleared for in vitro diagnostic use and the dalbavancin MTS is currently under review by the FDA.