

# Tigecycline Evaluation Surveillance Trial (T.E.S.T.) - United States In Vitro Antibacterial Activity Against Selected Species of *Enterococcus* spp.

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## REVISED ABSTRACT

**Background:** Tigecycline, a member of a new class of antimicrobials (glycylcyclines), has been shown to have potent expanded broad spectrum activity against most commonly encountered species responsible for community and hospital acquired infections. The T.E.S.T. program determined the in vitro activity of tigecycline compared to vancomycin, linezolid, ampicillin, imipenem, ceftriaxone, levofloxacin, minocycline, penicillin and piperacillin/tazobactam against members of *Enterococcus* spp. collected from hospitals in the USA. **Methods:** A total of 2,777 clinical enterococci were identified to the species level at each participating site and confirmed by the central laboratory. Isolates were collected from 2004 through 2006. Minimum Inhibitory Concentration (MICs) were determined by the local laboratory using broth microdilution panels and interpreted according to CLSI guidelines. **Results:** Of 1,897 *E. faecalis* evaluated, vancomycin resistance was noted in 90 (4.7%) isolates. These isolates were all susceptible to linezolid, penicillin, ampicillin and tigecycline. Tigecycline presented the lowest MIC<sub>50/90</sub> (0.06/0.12 mcg/ml) against all enterococci among the antimicrobial agents evaluated. As a typical profile of *E. faecalis*, fluoroquinolone (levofloxacin) and tetracycline (minocycline) had limited activities against this species. Among 772 *E. faecium*, 515 (66.7%) were resistant to vancomycin, of which 17 isolates were non-susceptible to linezolid. Tigecycline also presented the lowest MIC<sub>50/90</sub> of 0.06/0.12 mcg/ml against all vancomycin-resistant enterococci. **Conclusion:** Tigecycline's in vitro activity was comparable to or greater than most commonly prescribed antimicrobials. The presented data suggest that tigecycline may be an effective and reliable therapeutic option against *Enterococcus* spp. including vancomycin-resistant strains.

## INTRODUCTION

Tigecycline is a broad-spectrum antimicrobial agent and first-in-class of the semisynthetic glycylcyclines to be approved for human use [1]. This synthetic analogue of the minocycline molecule exhibits significant antibacterial activity that is both bacteriostatic and, in certain instances, bactericidal with killing activity that is as much as fourfold better than vancomycin and daptomycin [2, 3]. The development of tigecycline is important in that tigecycline and other glycylcyclines are active against bacterial strains carrying either or both of the two major forms of tetracycline resistance: efflux and ribosomal protection. Certain substituents at the 9-position of the tetracycline molecule restored activity against bacteria harboring genes encoding either or both efflux and ribosomal protection. A single chemical modification of tigecycline overcomes the two molecularly distinct forms of resistance while maintaining activity against susceptible gram-positive, gram-negative, aerobic, and anaerobic bacteria [4]. Furthermore, resistance to tigecycline is difficult to produce even in the laboratory.

Previous studies have demonstrated excellent in vitro activity for tigecycline against clinical and laboratory strains of gram-positive and gram-negative bacteria with minimum inhibitory concentrations for the 90th percentile inhibited at or below 2 mcg/ml, including difficult to treat methicillin-resistant *S. aureus* (MRSA), vancomycin-resistant enterococci (VRE) and extended-spectrum beta-lactamase (ESBL) producing *Enterobacteriaceae* [5-9]. This study was undertaken to document the in vitro activity of tigecycline against significant numbers of *Enterococcus* spp. collected from laboratories in the United States. This study is part of the larger ongoing global Tigecycline Evaluation and Surveillance Trials (T.E.S.T.) program.

## MATERIALS & METHODS

- All isolates were derived from blood, respiratory tract, urine, skin, wound, body fluids and other defined sources. Only one isolate per patient was accepted into the study. Clinical isolates were collected and tested between January 2004 and December 2006 from 187 study centers in the United States. Isolates were identified to the species level and tested at each site by the participating laboratory.
- Organism collection, transport, confirmation of organism identification, as well as, development and management of a centralized database was coordinated by Laboratories International for Microbiology Studies (LIMS), a division of International Health Management Associates, Inc. located in Schaumburg, IL, USA.
- All organisms were deemed clinically significant by local participant criteria. Isolate inclusion was independent of medical history, antimicrobial use, age or gender. All sites identified each study isolate utilizing local laboratory site criteria.
- Minimum inhibitory concentrations (MICs) were determined by the CLSI recommended broth microdilution testing method [10]. Tigecycline was supplied by Wyeth Pharmaceuticals (Collegeville, PA, USA). All other agents were supplied by the panel manufacturer, MicroScan (Dade Behring Inc., Sacramento, CA, USA). The following antimicrobial agents were included on the panels with their dilution ranges (expressed in mcg/ml): amoxicillin/clavulanic acid (0.12/0.06-32/16); ampicillin (0.06-16); ceftriaxone (0.06-64); imipenem (0.06-16); linezolid (0.5-8); levofloxacin (0.008-8); minocycline (0.5-16); tigecycline (0.008-16); penicillin (0.06-8); piperacillin/tazobactam (0.06/4-128/4) and vancomycin (0.12-32). MIC interpretive criteria followed published guidelines established by the Clinical and Laboratory Standards Institute [11] and recent US Food and Drug Administration packaging insert for tigecycline [12], where applicable.
- Quality controls (QC) were performed by each testing site on each day of testing using ATCC control strains *S. aureus* ATCC 29213 and *Enterococcus faecalis* ATCC 29212. Results were included in the analysis only when corresponding QC isolates tested within the acceptable range according to CLSI (2006) guidelines [11].

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The results are listed in the following Tables.

Table 1. In vitro activity of tigecycline and comparative agents against 2,777 *Enterococcus* species in the United States.

Organisms	Drug	%Sus <sup>a</sup>	%Int	%Res	MIC (mcg/ml)		
					MIC <sub>50</sub>	MIC <sub>90</sub>	Range
<i>Enterococcus</i> spp (n=2,777)	Tigecycline	100	0	0	0.06	0.12	≤0.008 - 0.25
	Ampicillin	75.3	0	24.7	1	>16	≤0.06 - >16
	Levofloxacin	43.6	1	55.3	16	>32	≤0.06 - >32
	Linezolid	98.3	1.7	0	2	2	≤0.5 - 4
	Minocycline	55.1	37.6	7.4	4	8	≤0.25 - >8
	Penicillin	74.7	0	25.3	2	>8	≤0.06 - >8
<i>Enterococcus</i> <i>avium</i> (n=18)	Tigecycline	100	0	0	0.06	0.06	≤0.008 - 0.06
	Ampicillin	83.3	0	16.7	1	>16	≤0.06 - >16
	Levofloxacin	72.2	5.6	22.2	2	32	≤0.06 - >32
	Linezolid	100	0	0	1	2	1 - 2
	Minocycline	55.6	38.9	5.6	2	8	≤0.25 - >8
	Penicillin	77.8	0	22.2	1	>8	≤0.06 - >8
<i>Enterococcus</i> <i>casseliflavus</i> (n=23)	Tigecycline	100	0	0	0.06	0.12	0.03 - 0.25
	Ampicillin	100	0	0	0.5	1	0.25 - 1
	Levofloxacin	82.6	4.3	13	2	8	1 - 16
	Linezolid	82.6	17.4	0	2	4	1 - 4
	Minocycline	78.3	17.4	4.3	≤0.25	8	≤0.25 - >8
	Penicillin	100	0	0	1	2	0.5 - 8
<i>Enterococcus</i> <i>durans</i> (n=15)	Tigecycline	100	0	0	0.06	0.12	0.03 - 0.25
	Ampicillin	33.3	0	66.7	>16	>16	0.25 - >16
	Levofloxacin	33.3	6.7	60	32	>32	0.12 - >32
	Linezolid	93.3	6.7	0	2	2	1 - 4
	Minocycline	46.7	26.7	26.7	8	8	≤0.25 - >8
	Penicillin	33.3	0	66.7	>8	>8	0.5 - >8
<i>Enterococcus</i> <i>faecalis</i> (n=1,897)	Tigecycline	100	0	0	0.06	0.12	0.015 - 0.25
	Ampicillin	100	0	0	1	1	≤0.06 - 8
	Levofloxacin	56.8	0.9	42.3	1	>32	≤0.06 - >32
	Linezolid	99	1	0	2	2	≤0.5 - 4
	Minocycline	47.1	45.5	7.4	8	8	≤0.25 - >8
	Penicillin	100	0	0	2	4	≤0.06 - 8
<i>Enterococcus</i> <i>faecium</i> (n=772)	Tigecycline	100	0	0	0.03	0.12	0.015 - 0.25
	Ampicillin	13.9	0	86.1	>16	>16	0.12 - >16
	Levofloxacin	9.6	1	89.4	>32	>32	0.12 - >32
	Linezolid	97.2	2.8	0	2	2	≤0.5 - 4
	Minocycline	73.8	19.7	6.5	≤0.25	8	≤0.25 - >8
	Penicillin	12.2	0	87.8	>8	>8	0.12 - >8
<i>Enterococcus</i> Group D (n=29)	Tigecycline	100	0	0	0.12	0.25	0.03 - 0.25
	Ampicillin	96.6	0	3.4	1	2	0.12 - >16
	Levofloxacin	51.7	0	48.3	2	>32	0.5 - >32
	Linezolid	96.6	3.4	0	2	2	≤0.5 - 4
	Minocycline	48.3	24.1	27.6	8	>8	≤0.25 - >8
	Penicillin	96.6	0	3.4	2	4	0.5 - >8
<i>Enterococcus</i> <i>raffiniosus</i> (n=4)	Tigecycline	100	0	0	0.06	0.06	0.06 - 0.06
	Ampicillin	75	0	25	0.5	>16	0.5 - >16
	Levofloxacin	50	0	50	2	>32	1 - >32
	Linezolid	100	0	0	2	2	1 - 2
	Minocycline	100	0	0	4	4	≤0.25 - 4
	Penicillin	50	0	50	1	>8	0.5 - >8
<i>Enterococcus</i> non-specified (n=19)	Tigecycline	100	0	0	0.06	0.12	0.03 - 0.12
	Ampicillin	68.4	0	31.6	1	>16	0.5 - >16
	Levofloxacin	36.8	0	63.2	32	>32	≤0.06 - >32
	Linezolid	100	0	0	2	2	≤0.5 - 2
	Minocycline	63.2	31.6	5.3	2	8	≤0.25 - >8
	Penicillin	63.2	0	36.8	4	>8	2 - >8
<i>Enterococcus</i> Group D (n=29)	Tigecycline	100	0	0	0.12	0.25	0.03 - 0.25
	Ampicillin	96.6	0	3.4	1	2	0.12 - >16
	Levofloxacin	51.7	0	48.3	2	>32	0.5 - >32
	Linezolid	96.6	3.4	0	2	2	≤0.5 - 4
	Minocycline	48.3	24.1	27.6	8	>8	≤0.25 - >8
	Penicillin	96.6	0	3.4	2	4	0.5 - >8

<sup>a</sup> Interpretive criteria as defined by CLSI, M100-S16 (2006), where applicable [10]; Tigecycline FDA breakpoints for enterococci are approved for vancomycin-susceptible *E. faecalis*, only [12]; Breakpoints for tigecycline were applied to other enterococci for comparison purposes only.

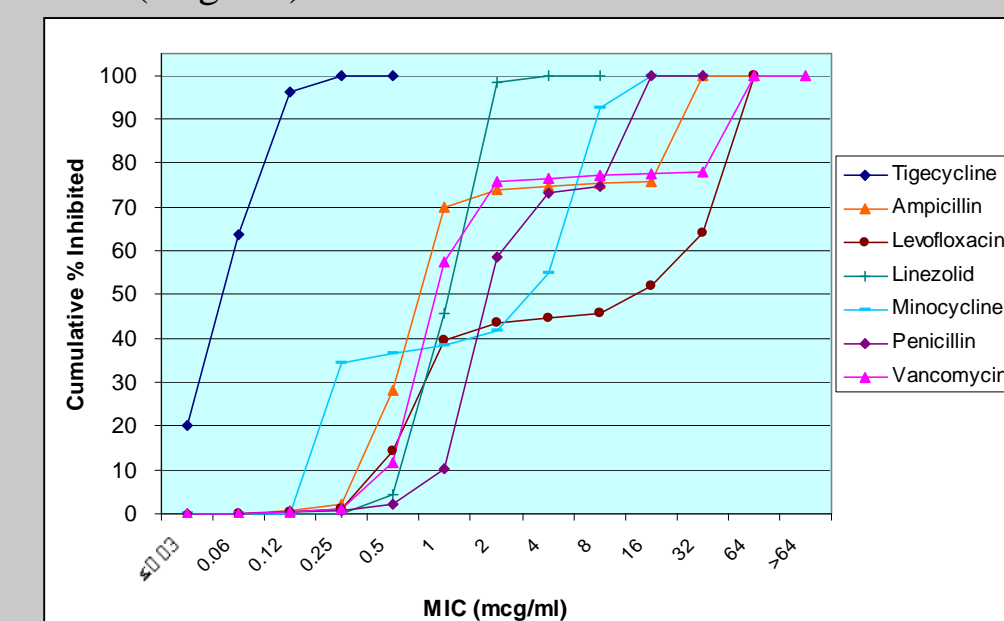
## RESULTS

Table 2. In vitro activity of tigecycline and comparative agents against 605 vancomycin-resistant enterococci in the United States.

Organisms	Drug	%Sus <sup>a</sup>	%Int	%Res	MIC (mcg/ml)		
					MIC <sub>50</sub>	MIC <sub>90</sub>	Range
Vancomycin-Resistant <i>E. faecium/faecalis</i> (n=605)	Tigecycline	100	0	0	0.06	0.12	0.015 - 0.25
	Ampicillin	16.9	0	83.1	>16	>16	0.25 - >16
	Levofloxacin	0.8	0.2	99	>32	>32	1 - >32
	Linezolid	97.2	2.8	0	2	2	≤0.5 - 4
	Minocycline	70.9	22.6	6.4	≤0.25	8	≤0.25 - >8
	Penicillin	15.9	0	84.1	>8	>8	0.5 - >8
Vancomycin-Resistant <i>Enterococcus</i> <i>faecium</i> (n=515)	Tigecycline	100	0	0	0.03	0.12	0.015 - 0.25
	Ampicillin	2.3	0	97.7	>16	>16	0.5 - >16
	Levofloxacin	0.4	0.2	99.4	>32	>32	2 - >32
	Linezolid	96.7	3.3	0	2	2	≤0.5 - 4
	Minocycline	72.4	20.6	7	≤0.25	8	≤0.25 - >8
	Penicillin	1.2	0	98.8	>8	>8	2 - >8
Vancomycin-Resistant <i>Enterococcus</i> <i>faecalis</i> (n=90)	Tigecycline	100	0	0	0.06	0.12	0.03 - 0.25
	Ampicillin	100	0	0	1	1	0.25 - 8
	Levofloxacin	3.3	0	96.7	32	>32	1 - >32
	Linezolid	100	0	0	1	2	≤0.5 - 2
	Minocycline	62.2	34.4	3.3	4	8	≤0.25 - >8
	Penicillin	100	0	0	2	4	0.5 - 8
Vancomycin-Resistant <i>Enterococcus</i> Group D (n=29)	Tigecycline	100	0	0	0.12	0.25	0.03 - 0.25
	Ampicillin	96.6	0	3.4	1	2	0.12 - >16
	Levofloxacin	51.7	0	48.3	2	>32	0.5 - >32
	Linezolid	96.6	3.4	0	2	2	≤0.5 - 4
	Minocycline	48.3	24.1	27.6	8	>8	≤0.25 - >8
	Penicillin	96.6	0	3.4	2	4	0.5 - >8

<sup>a</sup> Interpretive criteria as defined by CLSI, M100-S16 (2006), where applicable [10]; Tigecycline FDA breakpoints for enterococci are approved for vancomycin-susceptible *E. faecalis*, only [12]; Breakpoints for tigecycline were applied to other enterococci for comparison purposes only.

Figure 1. Cumulative percents inhibited (%) of tigecycline and comparative agents against 2,777 enterococci at each MIC (mcg/mL).



## CONCLUSIONS

- Tigecycline inhibited 100% of all *Enterococcus* spp. at the FDA susceptible breakpoints of 0.25 mcg/ml without regard to vancomycin-resistant phenotype.
- Tigecycline demonstrated equivalent in vitro potency to penicillin, ampicillin, vancomycin and linezolid against all individual *Enterococcus* species with MIC<sub>90</sub> values ranging from 0.12 to 0.25 mcg/ml.
- Tigecycline's MIC<sub>90</sub> of 0.12 mcg/ml against both vancomycin-resistant *E. faecalis* and vancomycin-resistant *E. faecium* was the lowest of all comparator agents in this study.
- The in vitro activity of tigecycline in this study suggests that tigecycline is highly active against vancomycin-resistant *Enterococcus* species and may be an effective treatment option for these frequently difficult to treat phenotypes.