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Tigecycline in vitro Activity against Staphylococcus aureus and Enterococcus strains Resistant To Other Drugs In The T.E.S.T. Program - United States, 2004 - 2005

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REVISED ABSTRACT

Background: Tigecycline (TIG), a member of a new class of antimicrobials (glycylcyclines), has been shown to have potent activity against many Gram-positive and -negative organisms. The T.E.S.T. program determined the in vitro activity of TIG against *S. aureus* and enterococci resistant to 10 commonly prescribed antimicrobials: amoxicillin-clavulanic acid (AUG), piperacillin-tazobactam (PT), levofloxacin (LVX), ceftriaxone (CAX), linezolid (LZD), minocycline (MIN), vancomycin (VAN), ampicillin (AMP), penicillin (P) and imipenem (IMP). Study strains were collected from clinical laboratories in the United States during 2004-2005. **Methods:** A total of 2,777 clinical isolates (1081 enterococci, 1,696 *S. aureus*) from 77 laboratories were identified to the species level at each participating site and confirmed by the central laboratory. Minimum Inhibitory Concentrations (MICs) were determined by the local laboratory using broth microdilution panels. Antimicrobial resistance was interpreted according to CLSI breakpoints with TIG susceptible breakpoints defined as ≤0.5 mcg/mL for *S. aureus* and ≤0.25 mcg/mL for enterococci. **Results:** 686/1081 (64%) enterococci and 1591/1696 (94%) *S. aureus* (including MR + MS strains) were resistant to at least one of the study drugs. Among the enterococci, resistance rates were LVX 56%, P27%, AMP 26%, VAN 23%, MIN 10%, and LZD 0.6%. Resistant rates for *S. aureus* were P 93%, AMP 92%, AUG 40%, LVX 37%, PT 36%, CAX 22%, IMP 7%, LZD 0%, MIN 0.1% and VAN 0%. TIG inhibited 99.2% of the enterococci and 100% of the *S. aureus* resistant to other drugs. Modal TIG MICs ranged between 0.06 and 0.12 mcg/mL for all resistant strains. **Conclusions:** TIG retained potent activity against drug-resistant *S. aureus* and enterococcal isolates, inhibiting >99% of all resistant strains tested at the defined susceptibility breakpoints. TIG should prove to be a useful drug for therapy of infections with these resistant gram-positive pathogens.

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 -negative bacteria with minimum inhibitory concentrations for the 90th percentile inhibited at or below 2 mcg/mL, including difficult to treat methicillin-resistant *Staphylococcus 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 drug resistant *Staphylococcus aureus* and enterococci within 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 - October 2005 from 77 study centers across 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.

Antimicrobial Susceptibility Testing

- 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. The FDA susceptible breakpoint of 0.25 mcg/mL against vancomycin-susceptible *E. faecalis* was adapted for all enterococci for comparative purposes only.
- Quality controls (QC) were performed by each testing site on each day of testing using the corresponding 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 (2005) guidelines [11].

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RESULTS

The results are listed in the following Tables.

Table 1. Study Organisms with Phenotypes.

Genus/Species/Phenotype *	Phenotype	Total N
Staphylococcus aureus		1,696
Staphylococcus aureus , MRSA	870 (51.3%)	
Staphylococcus aureus , MSSA	826 (48.7%)	
Enterococcus avium		8
Enterococcus casseliflavus		11
Enterococcus durans		10
Enterococcus faecalis		750
Enterococcus faecalis , VRE	40 (5.3%)	
Enterococcus faecium		283
Enterococcus faecium , VRE	192 (67.8%)	
Enterococcus Group D		1
Enterococcus raffinosus		2
Enterococcus , non-speciated		16
Total		**2,777**

*Phenotypes: MRSA = methicillin-resistant *S. aureus*; MSSA = methicillin-susceptible *S. aureus*; VRE = vancomycin-resistant enterococci.

Table 2. In Vitro Susceptibility of Tigecycline and comparators against *Staphylococcus aureus* and *Enterococcus* species.

Drug	MIC mcg/mL)			%Sus	%Res
	MIC_{50}	MIC_{90}	Range		
***Staphylococcus aureus* (n=1,696)**					
Tigecycline	0.12	0.25	0.015 - 0.5	100	0
AmoxClav	4	>8	≤0.03 - >8	60.6	39.4
Ampicillin	16	>16	≤0.06 - >16	7.6	92.4
Ceftriaxone	16	>64	0.12 - >64	46.6	21.5
Imipenem	0.25	4	≤0.12 - >16	90.6	7.4
Levofloxacin	0.5	>32	≤0.06 - >32	54	36.7
Linezolid	2	4	≤0.5 - 4	100	0
Minocycline	≤0.25	0.5	≤0.25 - >8	99.2	0.1
Penicillin	>8	>8	≤0.06 - >8	6.7	93.3
PipTazo	4	>16	≤0.25 - >16	64.3	35.7
Vancomycin	1	1	≤0.12 - 4	100	0
***Enterococcus* spp (n=1,081)**					
Tigecycline a	0.06	0.12	≤0.008 - 0.5	99.5	0.5
Ampicillin	1	>16	0.12 - >16	74	26
Levofloxacin	16	>32	≤0.06 - >32	42.6	56.2
Linezolid	2	2	≤0.5 - >8	96.9	0.6
Minocycline	4	8	≤0.25 - >8	50.8	9.5
Penicillin	2	>8	≤0.06 - >8	72.8	27.2
Vancomycin	1	>32	≤0.12 - >32	75.7	23.2

[a]Tigecycline susceptible breakpoint defined here as 0.25 mcg/mL for all enterococci for comparison purposes only (FDA approved breakpoint of 0.25 mcg/mL is for *E. faecalis*, vancomycin susceptible, only).

Table 3. Frequency Distribution of Drug Resistant Strains at each Tigecycline MIC (mcg/mL) for each comparator antimicrobial agent against 1591 *Staphylococcus aureus* strains resistant to one or more drugs.

S. aureus	Tigecycline MICs (mcg/mL)						
Resistant To:	0.015	0.03	0.06	0.12	0.25	0.5	Total
AmoxClav (R)	0	1	80	453*	107	27	668
Ampicillin (R)	1	7	256	1116*	154	32	1566
Ceftriaxone (R)	0	1	49	217*	71	26	364
Imipenem (R)	0	1	9	61*	39	16	126
Levofloxacin (R)	0	2	79	411*	103	26	621
Linezolid (R)	0	0	0	0	0	0	0
Minocycline (R)	0	0	0	0	0	1	1
Penicillin (R)	1	7	260	1125*	156	32	1581
PipTazo (R)	0	2	82	392*	99	29	604
Vancomycin (R)	0	0	0	0	0	0	0
Total No. Drug Resistant Values	2	21	815	3775*	729	189	5531

*Modal Value

Table 4. Frequency Distribution of Drug Resistant Strains at each Tigecycline MIC (mcg/mL) for each comparator antimicrobial agent against 686 *Enterococcus* spp strains resistant to one or more drugs.

Enterococci	Tigecycline MICs (mcg/mL)						
Resistant To:	0.015	0.03	0.06	0.12	0.25	0.5	Total
Ampicillin (R)	2	119	122*	30	5	3	281
Penicillin (R)	2	123	124*	36	6	3	294
Levofloxacin (R)	4	158	274*	155	12	4	607
Linezolid (R)	0	1	1	1	1	1	5
Minocycline (R)	0	1	27	57*	17	1	103
Vancomycin (R)	1	102	108*	35	3	2	251
Total No. Drug Resistant Values	9	504	656*	314	44	14	1541

*Modal Value

Table 5. In Vitro Susceptibility of Tigecycline against Drug Resistant *Staphylococcus aureus* and *Enterococcus* spp.

Drug	Total No. of Resistant Strains/Drug	Percent (%) Resistance for each Drug	Tigecycline %Susceptible against Resistant Strains	Tigecycline %Resistant against Resistant Strains
***Staphylococcus aureus* (n=1,696)**				
AmoxClav	668	39.4	100	0
Ampicillin	1566	92.3	100	0
Ceftriaxone	364	21.5	100	0
Imipenem	126	7.4	100	0
Levofloxacin	621	36.6	100	0
Linezolid	0	0	-	-
Minocycline	1	0.1	100	0
Penicillin	1581	93.2	100	0
PipTazo	604	35.6	100	0
Vancomycin	0	0	-	-
Total No. *S. aureus* resistant to at least 1 drug	1591	93.8	100	0
***Enterococcus* spp (n=1,081)[a]**				
Ampicillin	281	26	98.9	1.1
Penicillin	294	27.2	99	1
Levofloxacin	607	56.2	99.3	0.7
Linezolid	5	0.5	80	20
Minocycline	103	9.5	99	1
Vancomycin	251	23.2	99.2	0.8
Total No. *Enterococcus* spp resistant to at least 1 drug	686	63.5	99.2	0.8

[a]Tigecycline susceptible breakpoint defined here as 0.25 mcg/mL against all enterococci for comparison purposes only (FDA approved breakpoint of 0.25 mcg/mL is for *E. faecalis*, vancomycin susceptible, only).

CONCLUSIONS

- Tigecycline inhibited 100% of all *Staphylococcus aureus* and 99.5% *Enterococcus* species at the FDA susceptible breakpoints of 0.5 and 0.25 mcg/mL, respectively, without regard to either methicillin- or vancomycin-resistant phenotypes.
- 1591/1696 (94%) *S. aureus* and 686/1081 (64%) *Enterococcus* species were resistant to at least one of the study drugs.
- 100% of all drug resistant *Staphylococcus aureus* and 99.2% of all *Enterococcus* species with one or more drug resistant values were susceptible to tigecycline.
- Tigecycline demonstrated potent in vitro activity against all drug resistant *Staphylococcus aureus* and *Enterococcus* species with modal values (MIC50s) ranging from 0.06 to 0.12 mcg/mL.
- Tigecycline demonstrated equivalent in vitro potency to vancomycin and linezolid against all *Staphylococcus aureus* with MIC_{90} value of 0.25 mcg/mL and susceptibilities 100%.
- Tigecycline demonstrated equivalent in vitro potency to linezolid against all *Enterococcus* species with MIC_{90} value of 0.12 mcg/mL and susceptibilities >99%.
- The in vitro activity of tigecycline in this study suggests that tigecycline is a significant antimicrobial agent for the treatment of some of the most commonly encountered hospital and community acquired pathogens and is active against most drug resistant *Staphylococcus aureus* and *Enterococcus* species.