

Evaluation of the Activity of Tigecycline Against Worldwide Levofloxacin-Resistant Pathogens

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Revised Abstract

Objectives: Tigecycline (TIG) is the first member of the glycolcycline class of anti-infectives to be marketed, exhibiting a broad spectrum of antibacterial activity against many commonly-isolated pathogens. This study evaluates the in vitro activity of TIG versus levofloxacin (LEV)-resistant strains in a large set of isolates collected globally from 2004-2008. **Methods:** More than 25,000 LEV-resistant isolates were identified to the species level at participating sites and confirmed by the central laboratory. MICs were determined by each site using supplied broth microdilution panels and interpreted according to CLSI guidelines. **Results:** Percentages of LEV-resistant isolates susceptible to TIG are summarized in the following table:

Organism (n)	No. LEV-Resistant (%)	% Susceptible to TIG
<i>E. coli</i> (16174)	4451 (27.5)	99.9
<i>Klebsiella</i> spp. (15566)	2065 (13.3)	89.6
ESBL+ (3525)	2250 (63.8)	95.1
<i>Enterobacter</i> spp. (14330)	1472 (10.3)	81.0
<i>Serratia</i> spp. (5907)	222 (3.8)	85.6
<i>S. aureus</i> MR (6674)	4425 (66.3)	100
<i>S. aureus</i> MS (8781)	221 (2.5)	100
<i>Enterococcus</i> spp. (9528)	5305 (55.7)	99.8

Conclusions: LEV resistance varied in this study from 2.5% in methicillin-sensitive *S. aureus* to 66.3% in methicillin-resistant *S. aureus*. When resistance to LEV was seen, however, TIG retained activity in the vast majority of such strains. TIG's broad spectrum of activity, including strains resistant to other drugs, makes it a valuable tool for treating serious infections caused by bacteria that may be refractory to treatment with commonly-used antimicrobials such as LEV.

Introduction

Tigecycline is a novel antimicrobial with expanded broad-spectrum activity from a new class of compounds, the glycolcyclines. Tigecycline inhibits protein synthesis by binding to the 30S ribosomal subunit. Although it is perceived to be bacteriostatic, it has shown some bactericidal activity against key targeted pathogens [1,2]. Tigecycline was developed to provide activity against tetracycline and multi-drug-resistant gram-positive pathogens and has demonstrated significant broad-spectrum activity against aerobic and anaerobic gram-positive and gram-negative microorganisms [2-4].

Tigecycline resistance is very infrequent and is also difficult to induce in the laboratory [5, 6] with a selection frequency observed at less than 10⁻⁹ [3, 5, 7]. With the exception of *P. aeruginosa*, tetracycline-resistant bacteria with either tetracycline efflux pumps or ribosomal protective features are sensitive to tigecycline [2-4, 7-11]. Tigecycline has shown to be active against multi-resistant *Acinetobacter* spp., particularly *A. baumannii* that are commonly associated with serious nosocomial infections. Similar activity has been observed against *Enterobacteriaceae*, even extended-spectrum β -lactamase (ESBL) and AmpC producing strains [10]. Tigecycline has demonstrated MIC₉₀ values of ≤ 0.5 mcg/ml against methicillin-resistant *Staphylococcus aureus* (MRSA) and other gram-positive organisms [2, 4-6]. Tigecycline has shown potent activity in animal models infected with selected strains of multi-drug resistant *Enterococcus faecium* and *Enterococcus faecalis* [4,5] with diverse genotypes of vanA, vanB and vanC [6].

The T.E.S.T. program determined the in vitro activity of tigecycline compared to most commonly prescribed broad spectrum antimicrobials against gram-negative and gram-positive species collected from 1016 hospital sites in 53 countries from 2004 to 2008. This study was designed to evaluate the in vitro activity of tigecycline against levofloxacin resistant organisms.

Materials & Methods

- For the T.E.S.T. program all isolates were derived from blood, respiratory tract, urine (no more than 25% of all isolates), skin, wound, fluids, and other defined sources. Isolates were identified to genus and species by the local laboratory. Each site tested the isolates using common broth microdilution panels. Only one isolate per patient was accepted.
- For this study more than 25,000 clinical isolates were collected from 2004 to 2008 from 1016 hospitals in 53 countries.
- Minimum inhibitory concentrations (MICs) were determined by the CLSI recommended broth microdilution testing method [12]. 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): gram positive panel: amoxicillin/clavulanic acid (0.03/0.015-8/4, tested using a 2:1 ratio of amoxicillin/clavulanic acid; reported concentrations refer to amoxicillin); ampicillin (0.06-16); ceftriaxone (0.03-64); imipenem (0.06-16); linezolid (0.5-8); levofloxacin (0.06-32); minocycline (0.25-8); tigecycline (0.008-16); penicillin (0.06-8); piperacillin/tazobactam (0.25/4-16/4) and vancomycin (0.12-32); gram-negative panel: amikacin (0.5-64); amoxicillin/clavulanic acid (0.12/0.06-32/16, tested using a 2:1 ratio of amoxicillin/clavulanic acid; reported concentrations refer to amoxicillin); ampicillin (0.5-32); cefepime (0.5-32); ceftriaxone (0.06-64); ceftazidime (8-32); imipenem (0.06-16); levofloxacin (0.008-8); minocycline (0.5-16); tigecycline (0.008-16) and piperacillin/tazobactam (0.06/4-128/4).
- MIC interpretive criteria for all drugs except tigecycline followed published guidelines established by the CLSI where applicable [12]. MIC interpretive criteria for tigecycline followed criteria established by the Federal Drug Administration (FDA, United States, 2005) where applicable [13].
- Escherichia coli*, *Klebsiella pneumoniae*, and *Klebsiella oxytoca* were screened for ESBL activity when MIC results for ceftriaxone were >1 mcg/ml using broth microdilution panels. ESBL activity was confirmed using the CLSI (2006) phenotypic confirmatory disk test (Oxoid, Ogdensburg, NY, USA) on Mueller-Hinton agar (Remel Inc., Lenexa, KS, USA) according to CLSI (2006) guidelines. ESBL presence was confirmed by testing the following antibiotic disks: cefotaxime (30-mcg), cefotaxime/clavulanic acid (30/10-mcg), ceftazidime (30-mcg), and ceftazidime/clavulanic acid (30/10-mcg). Antimicrobial disks were manufactured by Oxoid, Inc. (Ogdensburg, NY, USA). Mueller-Hinton agar used in testing was manufactured by Remel, Inc. (Lenexa, KS, USA). An organism was interpreted as containing an ESBL if there was an increase of >5 mm in the inhibition zone of the combination disk when compared to that of the cephalosporin alone.
- Quality control of broth microdilution panels followed manufacturer's and CLSI guidelines using the following ATCC strains where applicable: *Enterococcus faecalis* ATCC 29212; *Escherichia coli* ATCC 25922; *Escherichia coli* ATCC 35218; *Klebsiella pneumoniae* ATCC 700603 (as positive ESBL control); *Haemophilus influenzae* ATCC 49247; *Haemophilus influenzae* ATCC 49766; *Staphylococcus aureus* ATCC 29213; *Streptococcus pneumoniae* ATCC 49619; and *Pseudomonas aeruginosa* ATCC 27853.
- The collection and transportation of organisms, confirmation of identification, and construction and management of a centralized database were conducted and coordinated by Laboratories International for Microbiology Studies (LIMS), a subsidiary of International Health Management Associates, Inc. (IHMA, Schaumburg, IL, USA).

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Results

Table 1. In vitro activity of tigecycline and comparative agents against selected levofloxacin resistant gram-negative organisms.

Organism (n)	%S ^a	MIC ₅₀ (mcg/mL)	MIC ₉₀ (mcg/mL)	
<i>Enterobacteriaceae</i> spp. (8210)	Tigecycline	93.5	0.5	
	Amikacin	88.7	4	
	AmoxClav	27.6	16	
	Ampicillin	4.2	>32	
	Cefepime	65.2	2	
	Ceftriaxone	45.9	16	
	Imipenem	98.6	0.25	
	Levofloxacin	0.0	>8	
	Minocycline	53.1	4	
	PipTazo	61.5	8	
<i>E. coli</i> (4451)	Tigecycline	99.9	0.25	
	Amikacin	95.9	2	
	AmoxClav	41.7	16	
	Ampicillin	7.7	>32	
	Cefepime	76.1	≤ 0.5	
	Ceftriaxone	65.2	0.12	
	Imipenem	99.8	0.25	
	Levofloxacin	0.0	>8	
	Minocycline	62.2	4	
	PipTazo	84.4	2	
<i>Klebsiella</i> spp. (2065)	Tigecycline	89.6	1	
	Amikacin	76.0	8	
	AmoxClav	18.4	32	
	Ampicillin	0.1	>32	
	Cefepime	42.1	16	
	Ceftriaxone	22.6	>64	
	Imipenem	95.6	0.5	
	Levofloxacin	0	>8	
	Minocycline	49.5	8	
	PipTazo	34.6	128	
ESBL-producers ^b (2250)	Tigecycline	95.1	0.5	
	Amikacin	83.0	8	
	AmoxClav	18.4	16	
	Ampicillin	0.3	>32	
	Cefepime	30.0	32	
	Ceftriaxone	8.6	>64	
	Imipenem	97.4	0.25	
	Levofloxacin	0	>8	
	Minocycline	55.4	4	
	PipTazo	53.8	16	
<i>Enterobacter</i> spp. (1472)	Tigecycline	81.0	1	
	Amikacin	85.6	4	
	AmoxClav	1.8	>32	
	Ampicillin	0	>32	
	Cefepime	63.1	8	
	Ceftriaxone	19.6	64	
	Imipenem	98.5	0.5	
	Levofloxacin	0	>8	
	Minocycline	32.1	8	
	PipTazo	30.8	64	
<i>Acinetobacter</i> spp. (3405)	Tigecycline	na	1	
	Amikacin	45.8	32	
	Cefepime	8.9	32	
	Ceftriaxone	2.4	>64	
	Imipenem	65.6	2	
	Levofloxacin	0	>8	
	Minocycline	77.4	1	
	PipTazo	13.5	128	
	<i>P. aeruginosa</i> (3799)	Tigecycline	na	16
		Amikacin	77.6	8
Cefepime		42.5	16	
Ceftriaxone		7.2	>64	
Imipenem		60.1	4	
Levofloxacin		0	>8	
Minocycline		na	>16	
PipTazo		67.9	32	

^a Susceptibilities are defined in CLSI document M100-S18 (2008) where applicable. Tigecycline breakpoints are defined in FDA package insert (Tygacil®). 2005). na = CLSI breakpoints not established for this species.

^b ESBL producing *E. coli*, *K. pneumoniae* and *K. oxytoca*.

Table 2. In vitro activity of tigecycline and comparative agents against selected levofloxacin resistant gram-positive organisms.

	%S ^a	MIC ₅₀ (mcg/mL)	MIC ₉₀ (mcg/mL)	
<i>S. aureus</i> (methicillin-susceptible) (221)	Tigecycline	100	0.12	
	AmoxClav	99.5	0.5	
	Ampicillin	88.5	0.25	
	Ceftriaxone	98.9	4	
	Imipenem	100	≤ 0.12	
	Levofloxacin	0	16	
	Linezolid	100	2	
	Minocycline	98.4	≤ 0.25	
	Penicillin	86.3	0.12	
	PipTazo	98.9	0.5	
<i>S. aureus</i> (methicillin-resistant) (4425)	Tigecycline	100	0.12	
	AmoxClav	0	8	
	Ampicillin	0	>16	
	Ceftriaxone	0	64	
	Imipenem	0	1	
	Levofloxacin	0	16	
	Linezolid	100	2	
	Minocycline	95.8	≤ 0.25	
	Penicillin	0	>8	
	PipTazo	0	>16	
<i>E. faecalis</i> (2774)	Tigecycline	99.8 ^b	0.12	
	Ampicillin	99.8	1	
	Levofloxacin	0	32	
	Linezolid	98.9	1	
	Minocycline	43.3	8	
	Penicillin	99.4	4	
	Vancomycin	91.9	1	
	<i>E. faecium</i> (2214)	Tigecycline	100 ^b	0.06
		Ampicillin	4.8	>16
		Levofloxacin	0	>32
Linezolid		95.4	2	
Minocycline		73.4	≤ 0.25	
Penicillin		4.3	>8	
Vancomycin		44.1	>32	
VRE ^c (1210)		Tigecycline	99.9 ^b	0.06
		Ampicillin	15.2	>16
		Levofloxacin	0	>32
	Linezolid	95.6	2	
	Minocycline	70	0.5	
	Penicillin	14.7	>8	
	Vancomycin	0	>32	

^a Susceptibilities are defined in CLSI document M100-S18 (2008) where applicable. Tigecycline breakpoints are defined in FDA package insert (Tygacil®). 2005).

^b Tigecycline breakpoints are defined in FDA package insert (Tygacil®). 2005) as susceptible less than or equal to 0.25 mcg/mL for vancomycin-susceptible *E. faecalis* only. This breakpoint was expanded to include all enterococci for comparative purposes only.

^c Vancomycin-resistant *E. faecium* and *E. faecalis*.

Conclusions

- While levofloxacin resistance in this study varied according to organism type, the overall global percentage of levofloxacin resistance against gram-positive and gram-negative aerobic bacteria in this study was 21.6% (25,425/117,854) (only major species are shown).
- Tigecycline has limited activity against *P. aeruginosa*, but excellent in vitro activity against *Acinetobacter* spp. with the lowest MIC_{50/90} (1/2 mcg/mL) values of all antimicrobials tested.
- For levofloxacin resistant *Enterobacteriaceae*, including ESBL producers, tigecycline MIC_{50/90} values were 0.5/2 mcg/mL, comparable to those of imipenem (0.25/1 mcg/mL).
- Against levofloxacin resistant gram-positive organisms, including MRSA and VRE, tigecycline showed excellent in vitro activity, with tigecycline MIC₉₀ values of ≤ 0.25 mcg/mL. 100% of MRSA and 99.9% VRE were susceptible to tigecycline at breakpoints of 0.5 and 0.25 mcg/mL, respectively.
- Tigecycline demonstrated broad spectrum in vitro activity against levofloxacin resistant pathogens, and may be a useful addition to hospital formularies against these potentially difficult-to-treat isolates.