

Revised Abstract

Background: Tigecycline (TIG), 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 TIG compared to amoxicillin-clavulanic acid, piperacillin-tazobactam, levofloxacin, ceftriaxone, linezolid (LZD), minocycline (MIN), vancomycin (VAN), ampicillin, penicillin, and imipenem (IMP) against methicillin-resistant *S. aureus* (MRSA) isolates collected from 320 sites in 24 European countries between 2004 and 2008. **Methods:** A total of 1016 clinical isolates of MRSA were identified to the species level at each participating site and confirmed by the central laboratory. Minimum Inhibitory Concentration (MICs) were determined by the local laboratory using supplied broth microdilution panels and interpreted according to EUCAST guidelines. **Results:** The %S for the study drugs with MRSA activity--TIG, VAN, LZD, and MIN--was 99.9, 100, 100, and 83.7, respectively. MIC_{50/90} (mg/L) for TIG, VAN, LZD, and MIN were 0.12/0.25, 1/1, 2/4, and ≤0.25/4, respectively. **Conclusions:** European susceptibility patterns of MRSA remain fairly consistent. TIG was as potent as VAN and LZD, inhibiting 1015/1016 (99.9%) of the MRSA isolates at their respective breakpoints. TIG's excellent expanded broad spectrum of activity against MRSA should make it a very useful drug in treatment of difficult staphylococcal infections.

Introduction

Tigecycline is a novel antimicrobial with expanded broad-spectrum activity from a new class of compounds, the glycylcyclines. 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 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 *Pseudomonas aeruginosa*, tetracycline-resistant bacteria with either tetracycline efflux pumps or ribosomal protective features are sensitive to tigecycline [2-4, 7-11]. Tigecycline has been shown to be active against multi-resistant *Staphylococcus aureus* (MRSA), that are commonly associated with serious community and hospital-acquired nosocomial infections. Tigecycline has demonstrated MIC₉₀ values of 0.25 mg/L against methicillin-resistant *Staphylococcus aureus* (MRSA) and other gram-positive organisms [2, 4-6].

This study was designed to better define the *in vitro* activity of tigecycline in methicillin-resistant *Staphylococcus aureus* clinical isolates collected from 320 study centers in 24 European countries.

Materials & Methods

- All isolates were derived from blood, respiratory tract, urine (no more than 25% of all isolates), skin, wound, fluids, and other defined sources. Only one isolate per patient was accepted. Isolates were identified to genus and species by the local laboratory. Each site tested the isolates using broth microdilution.
- Clinical isolates (n=1,016) were collected tested between January 2004 – December 2008 from 320 study centers in 24 European countries.
- Custom broth microdilution panels were supplied by MicroScan (Dade MicroScan, Sacramento, CA, USA) with the following antimicrobial agents and concentrations (expressed in mg/L): amoxicillin/clavulanic acid (0.12-32); ampicillin (0.5-32); piperacillin/tazobactam (0.06-128); levofloxacin (0.008-8); ceftriaxone (0.06-64); linezolid (0.5-8); penicillin (0.06-8); imipenem (0.06-16); minocycline (0.5-16); tigecycline (0.008-16); and vancomycin (0.12-32).
- MIC interpretive criteria followed published guidelines defined by EUCAST, where available (<http://www.srga.org/eucastwt/MICTAB/index.html>, 2009); CLSI breakpoints were used where EUCAST not available. [12].
- Methicillin phenotype is based upon the susceptibility of *S. aureus* to ceftioxin, using the disk test according to CLSI document M100-S19 (2009) [12].
- Quality control of broth microdilution panels followed manufacturer's and CLSI guidelines using the following ATCC strains: *Staphylococcus aureus* ATCC 29213.
- The collection and transportation of organisms, confirmation of identification, as well as 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

Results are contained in the following tables.

Table 1. *In vitro* activity of tigecycline and comparative agents against 1,016 European clinical isolates of methicillin-resistant *S. aureus*.

Region	Drug	%SUS ¹	%INT	%RES	MIC (mg/L)		MIC range (mg/L)	
					MIC ₅₀	MIC ₉₀	Low	High
Europe	Tigecycline	99.9	0.0	0.1	0.12	0.25	0.03	1
	AmoxClav	4.9	0.0	95.1	>8	>8	0.25	>8
	Ampicillin	0.0	0.0	100.0	>16	>16	0.5	>16
	Ceftriaxone	0.0	0.0	100.0	64	>64	0.12	>64
	Imipenem	0.0	0.0	100.0	2	>16	≤0.12	>16
	Levofloxacin	12.6	2.8	84.6	8	32	≤0.06	>32
	Linezolid	100.0	0.0	0.0	2	4	≤0.5	4
	Meropenem	0.0	0.0	100.0	4	>16	≤0.12	>16
	Minocycline	83.7	2.2	14.2	≤0.25	4	≤0.25	>8
	Penicillin	0.0	0.0	100.0	>8	>8	0.25	>8
	PipTazo	7.4	0.0	92.6	>16	>16	≤0.25	>16
	Vancomycin	100.0	0.0	0.0	1	1	≤0.12	2

¹ Breakpoints as defined by EUCAST, where available; 2009; CLSI breakpoints were used where EUCAST not available; na = not available; breakpoints not defined. Tigecycline breakpoints are defined in FDA package insert (Tygacil®, 2005) as susceptible ≤0.5 mg/L[13]. All beta-lactams are resistant to MRSA as defined by EUCAST

Table 2. *In vitro* activity of tigecycline and comparators against European isolates of methicillin-resistant *S. aureus* (n=1,016) showing frequency distribution (n) and cumulative percent inhibited (%) at each MIC (mg/L).

MIC	0.03	0.06	0.12	0.25	0.5	1	2	4	8	16	32	64	128
n													
%													
Tigecycline	5	130	571	266	43	1							
	0.5	13.3	69.5	95.7	99.9	100							
AmoxClav				3	19	45	99	153	174	523			
				0.3	2.2	6.6	16.3	31.4	48.5	100.0			
Ampicillin				9	17	43	55	107	136	649			
				0.9	2.6	6.8	12.2	22.7	36.1	100.0			
Ceftriaxone			1	4	3	17	37	103	178	102	68	503	
			0.1	0.5	0.8	2.5	6.1	16.2	33.8	43.8	50.5	100.0	
Imipenem		37	78	64	33	27	19	13	22	148			
		8.4	26.1	40.6	48.1	54.2	58.5	61.5	66.4	100.0			
Levofloxacin	13	64	29	16	6	28	194	262	254	97	53		
	1.3	7.6	10.4	12.0	12.6	15.4	34.4	60.2	85.2	94.8	100.0		
Linezolid				18	227	660	111						
				1.8	24.1	89.1	100.0						
Minocycline			775	75	22	33	73	29	9				
			76.3	83.7	85.8	89.1	96.3	99.1	100.0				
Penicillin			11	7	16	31	45	96	810				
			1.1	1.8	3.3	6.4	10.8	20.3	100.0				
PipTazo		1	23	38	85	127	105	99	538				
		0.1	2.4	6.1	14.5	27.0	37.3	47.0	100.0				
Vancomycin		1	11	282	645	77							
		0.1	1.2	28.9	92.4	100.0							

Conclusions

- Tigecycline was as potent as VAN and LZD, inhibiting 100% of MRSA isolates at or below 0.5 mg/L.
- European susceptibility patterns of MRSA remain fairly consistent.
- The *in vitro* activity of tigecycline in this study suggests that it may be an effective agent for the treatment of methicillin-resistant *S. aureus* world-wide.