The Activity of Antimicrobial Agents Against Australian Bacteremia Isolates: The T.E.S.T. Program (2004-2008)

OB007

J. Johnson[1], R. Badal[1], S. Hawser[1], M. Hackel[1], S. Bouchillon[1], B. Johnson[1], D. Hoban[1], M. Renteria[1], M. Dowzicky[2]

[1]International Health Management Associates, Inc., Schaumburg, IL, USA
[2]Wyeth Pharmaceuticals, Collegeville, PA, USA

IHMA, Inc.
2122 Palmer Dr.
Schaumburg, IL
60173
Tel: 847.303.5003
Fax: 847.303.5601

Revised Abstract

Objectives: Tigecycline (TIG), a new glycylcycline, has been shown to have potent 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 and 10 comparators against bacteremia pathogens. Isolates were collected from 20 hospital sites in Australia throughout 2004-2008. **Methods**: 761 bacteremia 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**: Susceptibility of selected pathogens to tigecycline is summarized below:

Organism (#)	Tigecycline MIC_{50}	Tigecycline MIC_{90}	Tigecycline %S
Acinetobacter spp. (35)	0.25	1	-*
Enterococcus spp. (78)	0.12	0.25	100
E. coli (178)	0.12	0.25	100
Klebsiella spp. (113)	0.5	1	94.7
Enterobacter spp.(75)	0.5	1	96.0
P. aeruginosa (59)	8	16	-*
S. aureus MRSA (19)	0.12	0.25	100
S. aureus MSSA (69)	0.12	0.25	100
S. pneumoniae (57)	0.015	0.06	-*
S. agalactiae (36)	0.06	0.12	97.2
Serratia spp. (33)	1	2	100

*TIG breakpoint not defined.

Conclusions: TIG demonstrated a broad spectrum of antimicrobial activity, including *Acinetobacter* spp., *Enterobacteriaceae*, *S. aureus* (incl. MRSA), *S. pneumoniae* (all phenotypes), and *Enterococcus* spp. The wide spectrum of activity of tigecycline provides enhanced antimicrobial coverage of pathogens causing bacteremia.

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 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]. Activity has been observed against *Enterobacteriaceae*, including extended-spectrum beta-lactamase (ESBL) and *Amp*C producing strains [10]. Tigecycline has demonstrated MIC_{90} values of ≤0.5 mcg/ml against methicillin-resistant *Staphylococcus aureus* (MRSA) and other gram-positive organisms [2, 4-6].

The Tigecycline Evaluation and Surveillance Trials (T.E.S.T.) program determined the in vitro activity of tigecycline compared to most commonly prescribed broad spectrum antibiotics against gram-positive and gram-negative species. This study was designed to evaluate the in vitro activity of tigecycline against bacteremic pathogens collected from Australian hospitals.

Materials & Methods

- Clinical isolates from hematologic sources (n=761) were collected and tested between January 2004 and April 2008 from 20 sites in Australia. Isolates were identified to the species level and tested using broth microdilution at each site by the participating laboratory. All isolates were derived from blood culture specimens. Only one isolate per patient was accepted.
- Custom broth microdilution panels were supplied by MicroScan (Dade Behring Inc., Sacramento, CA, USA) and TREK (TREK Diagnostic Systems, West Sussex, England). Antimicrobial agents and concentrations tested (expressed in mcg/mL) were as follows: 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, MicroScan panels only); meropenem (0.12-16, MicroScan and TREK panels); 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, MicroScan panels only); meropenem (0.06-16, MicroScan and TREK panels); levofloxacin (0.008-8); minocycline (0.5-16); tigecycline (0.008-16) and piperacillin-tazobactam (0.06/4-128/4).
- Quality control of broth microdilution panels followed manufacturer's and CLSI guidelines using the following ATCC strains: *Enterococcus faecalis* ATTC 29212; *Escherichia coli* ATCC 25922; *Escherichia coli* ATCC 35218; *Haemophilus influenzae* ATCC 49247; *Haemophilus influenzae* ATCC 49766; *Staphylococcus aureus* ATCC 29213; *Streptococcus pneumoniae* ATCC 49619; *Klebsiella pneumoniae* ATCC 700603 and *Pseudomonas aeruginosa* ATCC 27853.
- The collection and transportation of organisms and the 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).

References

1 Sum, P.E. and P. Petersen, *Synthesis and structure-activity relationship of novel glycylcycline derivatives leading to the discovery of GAR-936.* Bioorg Med Chem Lett, 1999. **9**(10): p. 1459-62.
2 Abbanat, D., M. Macielag, and K. Bush, *Novel antibacterial agents for the treatment of serious Gram-positive infections.* Expert Opin Investig Drugs, 2003. **12**(3): p. 379-99.
3 Betriu, C., et al., *In vitro activities of tigecycline (GAR-936) against recently isolated clinical bacteria in Spain.* Antimicrob Agents Chemother, 2002. **46**(3): p. 892-5.
4 Gales, A.C. and R.N. Jones, *Antimicrobial activity and spectrum of the new glycylcycline, GAR-936 tested against 1,203 recent clinical bacterial isolates.* Diagn Microbiol Infect Dis, 2000. **36**(1): p. 19-36.
5 Henwood, C.J., et al., *Antibiotic resistance among clinical isolates of Acinetobacter in the UK, and in vitro evaluation of tigecycline (GAR-936).* J Antimicrob Chemother, 2002. **49**(3): p. 479-87.
6 Chopra, I., *New developments in tetracycline antibiotics: glycylcyclines and tetracycline efflux pump inhibitors.* Drug Resist Updat, 2002. **5**(3-4): p. 119-25.
7 Projan, S.J., *Preclinical pharmacology of GAR-936, a novel glycylcycline antibacterial agent.* Pharmacotherapy, 2000. **20**(9 Pt 2): p. 219S-223S; discussion 224S-228S.
8 Biedenbach, D.J., M.L. Beach, and R.N. Jones, *In vitro antimicrobial activity of GAR-936 tested against antibiotic-resistant gram-positive blood stream infection isolates and strains producing extended-spectrum beta-lactamases.* Diagn Microbiol Infect Dis, 2001. **40**(4): p. 173-
9 Patel, R., et al., *In vitro activity of GAR-936 against vancomycin-resistant enterococci, methicillin-resistant Staphylococcus aureus and penicillin-resistant Streptococcus pneumoniae.* Diagn Microbiol Infect Dis, 2000. **38**(3): p. 177-9.
10 Petersen, P.J., et al., *In vitro and in vivo antibacterial activities of a novel glycylcycline, the 9-t-butylglycylamido derivative of minocycline (GAR-936).* Antimicrob Agents Chemother, 1999. **43**(4): p. 738-44.
11 Petersen, P.J., et al., *In vitro and in vivo activities of tigecycline (GAR-936), daptomycin, and comparative antimicrobial agents against glycopeptide-intermediate Staphylococcus aureus and other resistant gram-positive pathogens.* Antimicrob Agents Chemother, 2002. **46**(8): p. 2595-601.
12 Clinical and Laboratory Standards Institute (CLSI). 2008. Performance Standards for Antimicrobial Susceptibility Testing; Fourteenth Informational Supplement. CLSI, document M100-S18. Wayne, PA, USA.
13 Tygacil®, 2005. Tigecycline FDA package insert.
14 Clinical and Laboratory Standards Institute (CLSI), 2007. *Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria That GrowAerobically; Approved Standard—Sixth Edition*, in *Document M7-A7.* CLSI, 940 Wayne, PA, USA.

Acknowledgements

We gratefully acknowledge contributions of the investigators, laboratory personnel and all members of the T.E.S.T. program group. This study was supported by a grant from Wyeth Pharmaceuticals.

Results

Figure 1. The distribution of 761 Australian blood culture isolates by species.

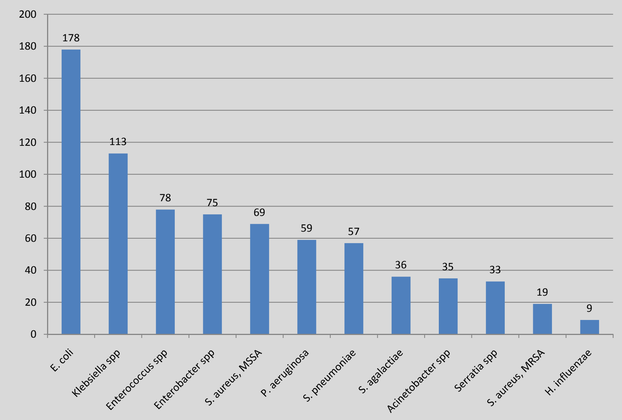

Table 1. The in vitro activity of tigecycline and comparative agents against *Enterobacteriaceae* isolated from blood specimens from Australia.

Organism	Drug	MIC (mcg/mL) MIC_{50}	MIC_{90}	Range	%Sus [a]
Enterobacter spp. (n=75)	Tigecycline	0.5	1	0.25 - 8	96.0
	Amikacin	2	4	≤0.5 - 8	100
	AmoxClav	>32	>32	1 - >32	5.3
	Ampicillin	>32	>32	2 - >32	1.3
	Cefepime	≤0.5	8	≤0.5 - >32	90.7
	Ceftazidime	≤8	>32	≤8 - >32	60.0
	Ceftriaxone	0.5	>64	≤0.06 - >64	65.3
	Imipenem	0.5	1	≤0.06 - 1	100
	Levofloxacin	0.06	0.5	≤0.008 - 8	94.7
	Meropenem	0.12	0.25	≤0.06 - 0.5	100
	Minocycline	4	16	1 - >16	78.7
	PipTazo	4	128	0.5 - >128	64.0
E. coli (n=178)	Tigecycline	0.12	0.25	0.06 - 2	100
	Amikacin	2	4	≤0.5 - 8	100
	AmoxClav	8	32	0.5 - >32	72.5
	Ampicillin	>32	>32	1 - >32	36.8
	Cefepime	≤0.5	≤0.5	≤0.5 - >32	98.9
	Ceftazidime	≤8	≤8	≤8 - 32	98.9
	Ceftriaxone	≤0.06	0.12	≤0.06 - >64	97.2
	Imipenem	0.25	0.25	≤0.06 - 1	100
	Levofloxacin	0.03	2	≤0.008 - >8	90.0
	Meropenem	≤0.06	≤0.06	≤0.06 - 0.5	100
	Minocycline	1	8	≤0.5 - >16	87.1
	PipTazo	1	4	0.25 - >128	93.8
Klebsiella spp. (n=113)	Tigecycline	0.5	1	0.25 - 8	94.7
	Amikacin	1	2	≤0.5 - 4	100
	AmoxClav	2	16	1 - >32	87.6
	Ampicillin	>32	>32	1 - >32	0.9
	Cefepime	≤0.5	≤0.5	≤0.5 - 8	100
	Ceftazidime	≤8	≤8	≤8 - 16	99.1
	Ceftriaxone	≤0.06	1	≤0.06 - >64	99.1
	Imipenem	0.25	0.5	≤0.06 - 0.5	100
	Levofloxacin	0.06	0.25	0.03 - 8	97.3
	Meropenem	≤0.06	0.12	≤0.06 - 0.5	100
	Minocycline	2	8	≤0.5 - >16	89.4
	PipTazo	2	8	0.5 - >128	97.3
Serratia spp. (n=33)	Tigecycline	1	2	0.5 - 2	100
	Amikacin	4	4	≤0.5 - 8	100
	AmoxClav	>32	>32	≤0.12 - >32	6.1
	Ampicillin	>32	>32	8 - >32	3.0
	Cefepime	≤0.5	≤0.5	≤0.5 - 2	100
	Ceftazidime	≤8	≤8	≤8 - ≤8	100
	Ceftriaxone	0.25	2	≤0.06 - 32	97.0
	Imipenem	1	2	0.12 - 2	100
	Levofloxacin	0.25	0.5	0.03 - 4	970.
	Meropenem	0.12	0.12	≤0.06 - 0.12	100
	Minocycline	4	8	2 - 8	84.8
	PipTazo	2	4	0.5 - 64	97.0

[a] Interpretive criteria as defined by CLSI document M100-S18, 2008, where available.
Tigecycline breakpoints defined by FDA (Tygacil®, 2005).

Table 2. The in vitro activity of tigecycline and comparative agents against *Acinetobacter* spp. and *Pseudomonas aeruginosa* isolated from blood cultures from Australia.

Organism	Drug	MIC (mcg/mL) MIC_{50}	MIC_{90}	Range	%Sus
Acinetobacter spp. (n=35)	Tigecycline	0.25	1	0.06 - 1	na
	Amikacin	2	8	≤0.5 - 64	91.4
	Cefepime	2	16	≤0.5 - >32	88.6
	Ceftazidime	≤8	>32	≤8 - >32	74.3
	Ceftriaxone	16	>64	0.25 - >64	48.6
	Imipenem	0.25	1	0.12 - >16	93.3
	Levofloxacin	0.12	1	0.015 - 8	94.3
	Meropenem	1	8	0.12 - >16	80
	Minocycline	≤0.5	2	≤0.5 - 8	94.3
	PipTazo	4	128	≤0.06 - >128	77.1
P. aeruginosa (n=59)	Tigecycline	8	16	2 - >16	na
	Amikacin	4	8	1 - 8	100
	Cefepime	2	8	1 - 32	91.5
	Ceftazidime	≤8	16	≤8 - >32	89.8
	Ceftriaxone	64	>64	8 - >64	5.1
	Imipenem	1	16	0.25 - 16	83.3
	Levofloxacin	0.5	4	0.25 - >8	86.4
	Meropenem	0.5	4	0.12 - >16	92.5
	Minocycline	16	>16	2 - >16	na
	PipTazo	8	32	2 - >128	96.6

[a] Interpretive criteria as defined by CLSI document M100-S18, 2008, where available. na=breakpoints not available.

Table 3. The in vitro activity of tigecycline and comparative agents against gram-positive pathogens isolated from blood specimens from Australia.

Organism	Drug	MIC (mcg/mL) MIC_{50}	MIC_{90}	Range	%Sus [a]
S. aureus, MRSA[b] (n=19)	Tigecycline	0.12	0.25	0.06 - 0.5	100
	AmoxClav	8	>8	2 - >8	5.3
	Ampicillin	>16	>16	16 - >16	0
	Ceftriaxone	32	>64	2 - >64	0
	Levofloxacin	0.25	16	≤0.06 - 16	78.9
	Linezolid	2	2	1 - 4	100
	Meropenem	2	>16	≤0.12 - >16	10.5
	Minocycline	≤0.25	4	≤0.25 - 4	100
	Penicillin	>8	>8	>8 - >8	0
	PipTazo	16	>16	1 - >16	10.5
	Vancomycin	1	2	0.5 - 2	100
S. aureus, MSSA[b] (n=69)	Tigecycline	0.12	0.25	0.06 - 0.5	100
	AmoxClav	1	2	0.12 - 2	100
	Ampicillin	4	>16	≤0.06 - >16	87.0
	Ceftriaxone	2	4	1 - 8	100
	Imipenem	≤0.12	0.25	≤0.12 - 0.25	100
	Levofloxacin	0.12	0.25	0.12 - 4	98.6
	Linezolid	2	4	1 - 4	100
	Meropenem	≤0.12	0.25	≤0.12 - 0.25	100
	Minocycline	≤0.25	≤0.25	≤0.25 - 2	100
	Penicillin	8	>8	≤0.06 - >8	87.0
	PipTazo	1	2	≤0.25 - 4	100
	Vancomycin	1	1	0.5 - 2	100
Enterococcus spp.[c] (n=78)	Tigecycline	0.12	0.25	0.03 - 0.25	100
	Ampicillin	1	>16	0.5 - >16	69.2
	Levofloxacin	32	>32	0.5 - >32	29.5
	Linezolid	2	2	1 - 4	100
	Minocycline	8	>8	≤0.25 - >8	44.9
	Penicillin	4	>8	0.5 - >8	70.5
	Vancomycin	1	4	0.5 - >32	94.9
S. agalactiae (n=36)	Tigecycline	0.06	0.12	0.03 - 0.5	97.2
	Ampicillin	0.12	0.25	≤0.06 - 0.25	100
	Ceftriaxone	0.12	0.12	≤0.03 - 0.12	100
	Levofloxacin	1	1	0.5 - 2	100
	Linezolid	1	2	1 - 2	100
	Meropenem	≤0.12	≤0.12	≤0.12 - ≤0.12	100
	Penicillin	0.12	0.12	≤0.06 - 0.25	97.2
	Vancomycin	0.5	1	0.25 - 1	100

[a] Interpretive criteria as defined by CLSI document M100-S18, 2008, where available.
[b] Methicillin phenotype based upon cefoxitin 30 mcg disk results; b-lactam susceptibilities based on methicillin phenotype.
[c] Tigecycline FDA breakpoints for enterococci are approved for vancomycin-susceptible *E. faecalis* only; susceptibilities for all other enterococci are entered for comparison purposes only.

Table 4. The in vitro activity of tigecycline and comparative agents against fastidious pathogens isolated from blood specimens from Australia.

Organism	Drug	MIC (mcg/mL) MIC_{50}	MIC_{90}	Range	%Sus [a]
H. influenzae (n=9)	Tigecycline	0.12	1	0.06 - 1	na
	AmoxClav	0.5	1	≤0.12 - 1	100
	Ampicillin	≤0.5	>32	≤0.5 - >32	77.8
	Cefepime	≤0.5	≤0.5	≤0.5 - ≤0.5	100
	Ceftriaxone	≤0.06	≤0.06	≤0.06 - ≤0.06	100
	Imipenem	0.12	0.12	0.12 - 0.12	100
	Levofloxacin	0.015	0.015	≤0.008 - 0.015	100
	Meropenem	≤0.06	0.12	≤0.06 - 0.12	100
	Minocycline	≤0.5	1	≤0.5 - 1	na
	PipTazo	≤0.06	0.12	≤0.06 - 0.12	100
S. pneumoniae (n=57)	Tigecycline	0.015	0.06	≤0.008 - 0.12	na
	AmoxClav	≤0.03	0.25	≤0.03 - 1	100
	Ceftriaxone	≤0.03	0.12	≤0.03 - 1	100
	Imipenem	≤0.12	≤0.12	≤0.12 - 0.5	94.1
	Levofloxacin	0.5	1	0.12 - 2	100
	Linezolid	≤0.5	1	≤0.5 - 1	100
	Meropenem	≤0.12	≤0.12	≤0.12 - 0.25	95
	Minocycline	≤0.25	1	≤0.25 - >8	na
	Penicillin	≤0.06	0.5	≤0.06 - 1	77.2
	Vancomycin	0.25	0.5	≤0.12 - 0.5	100

[a] Interpretive criteria as defined by CLSI document M100-S18, 2008, where available. na=breakpoints not available.

Conclusions

- The in vitro activity of tigecycline was comparable to amikacin, imipenem and meropenem against the *Enterobacteriaceae* associated with bacteremic sources with MIC_{90} values ranging from 0.25 to 2 mcg/mL among the selected species.
- Tigecycline's MIC_{50}/MIC_{90} values against *Acinetobacter* spp of 0.25 and 1 mcg/mL, respectively, were among the lowest of all agents in this study.
- Against gram-positive organisms, including methicillin-resistant *Staphylococcus aureus*, tigecycline showed excellent in vitro activity. All strains were inhibited at ≤0.5 mcg/ml and 100% of all *S. aureus* and *Enterococcus* spp. were susceptible to tigecycline.