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Activity of Tigecycline Tested Against Tetracycline-Resistant Enterobacteriaceae

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ABSTRACT

Background:
Tigecycline (TIG), a 9-butylglycylamido derivative of minocycline, is being developed as a parenteral broad-spectrum agent. This report summarizes the activity of TIG against a large collection of tetracycline (TET)-resistant (R) Enterobacteriaceae (ENT).

Methods:
6,309 ENT strains were acquired from recent (2000 to 2003) surveillance collections, of which 2,059 TET R (≥ 16 µg/ml) strains were evaluated. Susceptibility (S) was determined using NCCLS methods.

Results:
Activity of TIG is summarized in the table.

	$MIC_{50/90}$ (% S)[a]	
Organism (n; all/TET-R)	All ENT	TET-R ENT
E. coli (3,041/1,026)	0.12/0.25 (100)	0.25/0.5 (100)
Klebsiella spp. (1,122/172)	0.5/1 (99.9)	0.5/4 (99.4)
Enterobacter spp. (604/87)	0.5/2 (99.5)	2/4 (96.6)
P. mirabilis (309/303)	4/4 (90.0)	4/4 (90.1)
Proteae, indole + (157/85)	1/4 (97.5)	1/4 (96.5)
Serratia spp. (188/133)	1/2 (98.9)	1/2 (98.5)
Citrobacter spp. (136/16)	0.25/0.5 (100)	0.5/2 (100)
Salmonella spp. (530/97)	0.5/0.5 (100)	0.5/1 (100)
Shigella spp. (161/134)	0.12/0.25 (100)	0.12/0.25 (100)

a. Tentative breakpoint of ≤ 4 µg/ml, as applied to TET.

TIG remained highly active against all TET-R organisms with $MIC_{50/90}$s being 0.12 to 4 µg/ml, with 90.1 to 100.0% being S. TIG potency decreased 2- to 4-fold for TET-R *E. coli*, *Klebsiella* spp., *Enterobacter* spp., *Citrobacter* spp., and *Salmonella* spp., indicating overlapping binding sites, without % S changes.

Conclusions:
TIG offers advantages to existing agents including an enhanced spectrum of activity and stability to the commonly occurring TET R mechanisms (*tet*A or B and *tet*M or O). ENT R to other antimicrobial classes also remain S to TIG, making this glycylcycline an attractive candidate for continued clinical development.

INTRODUCTION

The 9-t-butylglycylamido derivative of minocycline, tigecycline (formerly GAR-936), has become the sentinel representative of a new class known as the glycylcyclines. This compound offers important advantages to existing antimicrobials including enhanced spectrum of activity and stability against tetracycline resistance mechanisms (Tet A or B efflux determinants and Tet M or O ribosomal protection factors). Its mode of action on bacterial ribosomes shows identical and overlapping binding sites when compared to tetracycline, but the position 9 substitution of tigecycline provides additional steric hindrance features and resulting greater spectra of activity.

Tigecycline is currently undergoing extensive clinical evaluation as a parenteral agent because of its potent activity against a broad range of commonly occurring species, including many resistant organisms such as penicillin-resistant *Streptococcus pneumoniae* (PRSP), oxacillin-resistant staphylococci (ORSA), vancomycin-resistant enterococci (VRE), and extended-spectrum ß-lactamase (ESBL) producing strains of Enterobacteriaceae. Tigecycline is also active against *Haemophilus influenzae*, *Moraxella catarrhalis,* pathogenic *Neisseria* spp. and many other Gram-negative species.

In this study we evaluated the in vitro activity of tigecycline against a total of 6,309 (2,059 tetracycline-resistant) isolates of Enterobacteriaceae recovered predominantly from patients with bloodstream, respiratory tract, skin and soft tissue and urinary tract infections, to determine the influence of tetracycline resistant determinants on tigecycline susceptibility in a large collection of wild-type strains.

MATERIALS AND METHODS

Specimen Collection. The collection initially consisted of 6,309 consecutively acquired, non-duplicate, patient isolates of Enterobacteriaceae submitted from nearly 100 participating medical centers representing over 25 countries on the five continents (Asia, Australia, Europe, North America, and South America) during 2000-2003. Isolates were identified by the submitting laboratory and subsequently shipped to the monitor (The JONES Group/JMI Laboratories, Iowa, USA) where identifications were confirmed using traditional methods and/or the Vitek system (bioMerieux Vitek, Hazelwood, MO, USA). Of this number, 2,059 strains were characterized as being resistant to tetracycline (MIC, ≥ 16 µg/ml).

Susceptibility Testing. MIC values for tigecycline and comparator agents were tested using validated dry-form broth microdilution panels (TREK Diagnostics, Cleveland, OH) with cation-adjusted Mueller-Hinton broth medium. Testing, incubation and MIC interpretation were performed using the manufacturer's recommendations and suggested technical details of the NCCLS (2003 and 2004). Susceptibility for tigecycline was defined as ≤ 4 µg/ml, that breakpoint used for all tetracyclines by the NCCLS. Quality control strains utilized included *Escherichia coli* ATCC 25922.

SELECTED REFERENCES

1. Bauer G, Berens C, Projan SJ, Hillen W. (2004). Comparison of tetracycline and tigecycline binding to ribosomes mapped by dimethylsulphate and drug-directed Fe^{2+} cleavage of 16S rRNA. *Journal of Antimicrobial Chemotherapy* 53:592-599.

2. Betriu C, Culebras E, Rodriguez-Avial I, Gomez M, Sanchez BA, Picazo JJ. (2004). In vitro activities of tigecycline against erythromycin-resistant *Streptococcus pyogenes* and *Streptococcus agalactiae*: Mechanisms of macrolide and tetracycline resistance. *Antimicrobial Agents and Chemotherapy* 48:323-325.

3. Cercenado E, Cercenado S, Gomez JA, Bouza E. (2003). In vitro activity of tigecycline (GAR-936), a novel glycylcycline, against vancomycin-resistant enterococci and staphylococci with diminished susceptibility to glycopeptides. *Journal of Antimicrobial Chemotherapy* 52:138-139.

4. Fritsche TR, Kirby JT, Jones RN. (2004). In vitro activity of tigecycline (GAR-936) tested against 11,859 recent clinical isolates associated with community-acquired respiratory tract and Gram-positive cutaneous infections. *Diagnostic Microbiology and Infectious Disease* 49:201-209.

5. Hoellman DB, Pankuch GA, Jacobs MR, Appelbaum PC. (2000). Antipneumococcal activities of GAR-936 (a new glycylcycline) compared to those of the nine other agents against penicillin-susceptible and –resistant pneumococci. *Antimicrobial Agents and Chemotherapy* 44:1085-1088.

6. National Committee for Clinical Laboratory Standards. (2003). *Methods for dilution antimicrobial susceptibility tests for bacteria that grow aerobically; approved standard-sixth edition*. Approved document M7-A6. Wayne, PA:NCCLS.

7. National Committee for Clinical Laboratory Standards. (2004). *Performance standards for antimicrobial susceptibility testing,* 14th informational supplement, M100-S14. Wayne, PA:NCCLS.

8. Petersen PJ, Jacobus NV, Weiss WJ, Sum PE, Testa RT. (1999). In vitro and in vivo antibacterial activities of a new glycylcycline, the 9-t-butylglycylamido derivative of minocycline (GAR-936). *Antimicrobial Agents and Chemotherapy* 43:738-744.

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RESULTS

- Among all Enterobacteriaceae, tigecycline was highly active ($MIC_{50/90}$, 0.25/1 µg/ml; 99.4% inhibited at ≤ 4 µg/ml); only amikacin (97.7%), cefepime (96.4%) and ertapenem (99.3%) among comparator agents tested had similar results (Table 1).

- Nearly one-third (2059/6309; 32.6%) of Enterobacteriaceae were tetracycline-resistant, but only two strains (both *Proteus mirabilis*) were tigecycline-resistant (MIC, 16 µg/ml; Tables 1 and 2).

Table 1. Activity of tigecycline and comparator antimicrobial agents tested against 6,309 strains of Enterobacteriaceae acquired from recent (2000 - 2003) global surveillance collections.

	MIC (µg/ml)				
Organism (no. tested)	50%	90%	Range	% susceptible	% resistant
All Enterobacteriaceae (6,309)					
Tigecycline	0.25	1	0.03-16	99.4[a]	-
Tetracycline	≤2	>8	≤2->8	65.0	32.6
Amikacin	2	4	≤0.25->32	97.7	1.3
Ampicillin	>16	>16	≤1->16	36.9	58.1
Amoxicillin/Clavulanate	8	>16	≤1->16	67.1	20.0
Cefepime	≤0.12	0.5	≤0.12->16	96.4	2.8
Ceftazidime	≤1	2	≤1->16	93.5	5.0
Ciprofloxacin	≤0.03	>4	≤0.03->4	87.8	11.4
Ertapenem	≤0.06	≤0.06	≤0.06->8	99.3	0.3
Piperacillin/Tazobactam	2	8	≤0.12->256	93.9	3.6
Trimethoprim/Sulfamethoxazole	≤0.5	>2	≤0.5->2	75.8	24.2

a. Susceptible breakpoint as applied to tetraclines (≤ 4 µg/ml) [NCCLS, 2004].

Table 2. Tigecycline activity against 6,309 strains of Enterobacteriaceae collected as part of global antimicrobial resistance surveillance programs (2000 - 2003).

	MIC (µg/ml)			
Organism (no. tested)	50%	90%	Range	% at ≤ 4 µg/ml[a]
Citrobacter spp. (136)	0.25	0.5	0.12-4	100.0
Enterobacter spp. (604)	0.5	2	0.06-8	99.5
E. coli (3,041)	0.12	0.25	0.03-4	100.0
E. coli 0157:H7 (3)	0.25	-	0.12-0.5	100.0
E. fergusonii (1)	0.25	-	0.25	100.0
E. hermannii (3)	0.25	-	0.25-0.5	100.0
E. vulneris (2)	0.25	-	0.25	100.0
Hafnia alvei (4)	0.5	-	0.25-1	100.0
Klebsiella spp. (1,122)	0.5	1	0.06-8	99.9
Kluyvera ascorbata (2)	0.25	-	0.25-0.5	100.0
Kluyvera spp. (1)	0.25	-	0.25	100.0
Leclercia adecarboxylata (2)	0.25	-	0.25-0.5	100.0
Pantoea agglomerans (22)	0.25	0.5	0.06-4	100.0
Proteae, indole-positive (157)	1	4	0.06-8	97.5
P. mirabilis (309)	4	4	0.25-16	90.0
P. penneri (2)	1	-	1-4	100.0
Salmonella spp. (530)	0.5	0.5	0.12-2	100.0
Serratia spp. (188)	1	2	0.12-8	98.9
Shigella spp. (161)	0.12	0.25	0.03-0.5	100.0
Tatumella spp. (1)	0.25	-	0.25	100.0
Yersinia enterocolitica (18)	0.25	0.25	0.12-0.5	100.0

a. Susceptible breakpoint applied to tetracyclines (≤ 4 µg/ml) [NCCLS, 2004].

- Tigecycline remained highly active against the vast majority of tetracycline-resistant organisms with all MIC_{50} and MIC_{90} values being between 0.12 and 4 µg/ml, and with 90.1 to 100.0% of strains being inhibited at a breakpoint of ≤ 4 µg/ml (Table 3).

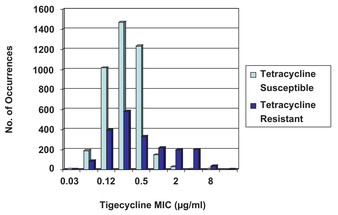
- While the central tendency of tigecycline MIC occurrences does not significantly vary with tetracycline-susceptible or -resistant populations, skewing towards higher MICs was apparent and occurs primarily in those species displaying innate resistance to the tetracyclines such as Proteae (Figure 1, Table 2).

Table 3. Activity of tigecycline tested against tetracycline-susceptible and -resistant subsets of species of Enterobacteriaceae (2000 - 2003).

	Tigecycline MIC (µg/ml)			% at MIC (µg/ml)	
Organism/ tetracycline-susceptibility (no. tested)	50%	90%	Range	≤2	≤4
E. coli					
Susceptible (2,001)	0.12	0.25	0.03-4	99.9	100.0
Resistant (1,026)	0.25	0.5	0.03-4	99.7	100.0
Klebsiella spp.					
Susceptible (911)	0.5	0.5	0.06-2	100.0	100.0
Resistant (172)	0.5	4	0.12-8	86.0	99.4
Enterobacter spp.					
Susceptible (486)	0.5	0.5	0.06-4	99.8	100.0
Resistant (87)	2	4	0.06-8	72.4	96.6
P. mirabilis					
Susceptible (4)	2	-	1-8	50.0	75.0
Resistant (303)	4	4	0.25-16	44.2	90.1
Proteae, indole-positive					
Susceptible (57)	1	2	0.06-8	94.7	98.2
Resistant (85)	1	4	0.06-8	84.7	96.5
Serratia spp.					
Susceptible (14)	0.5	1	0.12-1	100.0	100.0
Resistant (133)	1	2	0.5-8	95.5	98.5
Citrobacter spp.					
Susceptible (118)	0.25	0.5	0.12-2	100.0	100.0
Resistant (16)	0.5	2	0.25-4	93.8	100.0
Salmonella spp.					
Susceptible (427)	0.5	0.5	0.12-1	100.0	100.0
Resistant (97)	0.5	1	0.12-2	100.0	100.0
Shigella spp.					
Susceptible (25)	0.25	0.5	0.06-0.5	100.0	100.0
Resistant (134)	0.12	0.25	0.03-0.5	100.0	100.0

- Tigecycline potency decreased two- to four-fold among tetracycline-resistant *E. coli*, *Klebsiella* spp., *Enterobacter* spp., *Citrobacter* spp. and *Salmonella* spp., indicating overlapping resistance binding sites; susceptibility rates, however, remain unchanged (*E. coli*, *Citrobacter* spp. and *Salmonella* spp.) or decreased minimally (100.0 to 99.4%, *Klebsiella* spp.; 100.0 to 96.6%, *Enterobacter* spp.).

Figure 1. Activity of tigecycline against 6,156 isolates of Enterobacteriaceae (2,059 resistant to tetracycline).



CONCLUSIONS

- These findings confirm that tigecycline displays remarkable potency and breadth of spectrum against Enterobacteriaceae (99.4% of isolates were inhibited at ≤ 4 µg/ml versus 65.0% for tetracycline), including ESBL-producing isolates (data not shown).

- Tigecycline offers advantages to existing agents including potency, an enhanced spectrum of activity, and stability to the commonly occurring tetracycline resistance mechanisms (*tet*A or *tet*B and *tet*M or *tet*O).

- Enterobacteriaceae that are resistant to other antimicrobial classes also remain susceptible to tigecycline, attributes making this glycylcycline an attractive candidate for continued clinical development.