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In Vitro Antibacterial Activity of Tigecycline, a Novel Glycylcycline, Against Clinical Isolates of Enterobacteriaceae

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REVISED ABSTRACT

Background: Tigecycline (GAR-936, Wyeth) is a novel glycylcycline in development, which has been shown to have potent activity against most members of enterobacteriaceae. The activity of tigecycline was compared with those of other agents against *Escherichia coli*, *Enterobacter cloacae*, *Enterobacter aerogenes* and *Klebsiella pneumoniae* from hospitals throughout Europe, the Middle East and South Africa. **Methods:** Clinical isolates were collected between January 2001 – September 2002. Isolates were identified to genus and species at each site and confirmed by the central laboratory. MIC's were determined by the central laboratory using broth microdilution panels from Dade Microscan according to NCCLS guidelines and manufacturer's instructions. **Results:** The susceptibility results of the 1,499 isolates of enterobacteriaceae are listed in Table 1.

Table 1. In Vitro activity of Tigecycline Against 1,499 Enterobacteriaceae

Antibiotic	MIC (µg/ml)			
	Range Low	Range High	MIC ₅₀	MIC ₉₀
Tigecycline	0.12	8	0.5	1
Amox/Clav	0.5	>64	16	64
Amp/Sulb	0.5	>32	32	>32
Cefepime	0.5	>64	0.5	8
Ceftazidime	0.5	>64	0.5	>64
Ceftriaxone	0.5	>64	0.5	64
Imipenem	0.5	64	0.5	1
Levofloxacin	0.25	>64	0.25	8

Conclusion: Tigecycline exhibited excellent activity against all isolates tested. These results strongly suggest that tigecycline is a promising new antimicrobial agent with excellent activity against enterobacteriaceae.

INTRODUCTION

Glycylcyclines are tetracyclines with a 9-glycylamido substituent and were developed to avoid the two most prevalent tetracycline resistance mechanisms, ribosomal protection and efflux. Tigecycline, formerly GAR 936, is a glycylcycline derivative of minocycline, a semi-synthetic tetracycline, with a 9-t-butylglycylamido substitution [1]. Tigecycline inhibits protein synthesis by binding to the 30S ribosomal subunit. Although its antibacterial activity is significant, it is perceived to be bacteriostatic [2].

While developed to provide activity against tetracycline- and multi-drug-resistant gram-positive pathogens, it has also demonstrated significant broad-spectrum activity against aerobic and anaerobic gram-positive and gram-negative microorganisms [2-5].

Tigecycline resistance is very infrequent and is also difficult to induce in the laboratory [6, 7] with a selection frequency observed at less than 10⁻⁸[3, 6, 8]. Most tetracycline-

resistant bacteria with either tetracycline efflux pumps or ribosomal protective features are sensitive to tigecycline [2-4, 8-12]. Since tigecycline possesses no structural beta-lactam ring, it is particularly unaffected by beta-lactamase and extended-spectrum beta-lactamase producing enterobacteriaceae (ESBL) [9].

This study compared the activity of tigecycline with other agents against enterobacteriaceae including *Escherichia coli*, *Enterobacter cloacae*, *Enterobacter aerogenes* and *Klebsiella pneumoniae* from hospitals throughout Europe, the Middle East and South Africa.

MATERIALS & METHODS

All isolates were derived from blood, respiratory tract, urine (no more than 30% of all isolates), skin, wound, fluids and other defined sources. Only one isolate per patient was accepted.

Clinical isolates were collected between January 2001 – September 2002 from 38 study centers in 17 countries.

Antimicrobial agents tested with concentrations (expressed in µg/mL) were: amoxicillin/clavulanic acid (0.5 - 64); ampicillin/sulbactam (0.5 - 32); cefepime (0.5 - 64); ceftazidime (0.5 - 64); ceftriaxone (0.5 - 64); imipenem (0.5 - 64); levofloxacin (0.25 - 64); tigecycline (0.008 - 16). MIC interpretive criteria followed published guidelines established by the NCCLS [13].

Isolates were identified to genus and species at each site and confirmed by the central laboratory (Laboratories International for Microbiology Studies, Schaumburg, IL, USA).

Organism collection, transport, confirmation of organism identification, antimicrobial susceptibility testing, as well as, development and management of a centralized database was coordinated by Laboratories International for Microbiology Studies (LIMS).

Antimicrobial Susceptibility Testing

MIC's were determined by the central laboratory using microdilution panels by Dade Microscan (Dade Behring Inc., Sacramento, CA, USA) according to NCCLS guidelines and manufacturer's instructions.

Quality Control was performed using the following ATCC strains: *E. coli* ATCC 35218, *E. coli* ATCC 25922, *P. aeruginosa* ATCC 27853, *E. faecalis* ATCC 29212, and *S. aureus* ATCC 29213.

Extended-spectrum beta-lactamases were confirmed using the NCCLS recommended procedures for Screening and Confirmatory Tests for ESBLs in *Klebsiella pneumoniae*, *Klebsiella oxytoca*, and *Escherichia coli*. [13].

RESULTS

The results of this study are presented in the following tables.

Table 2. In Vitro MIC Values and Susceptibility Results of Tigecycline and Comparators Against 1,499 Enterobacteriaceae

Organism	Drug	MIC (mg/mL)			% Sus*
		Range	MIC ₅₀	MIC ₉₀	
Enterobacteriaceae (1499)	Tigecycline	0.12 - 8	0.5	1	97.3
	Amox/Clav	0.5 - >64	16	64	40.4
	Amp/Sulb	0.5 - >32	32	>32	28.0
	Cefepime	0.5 - >64	0.5	8	90.2
	Ceftazidime	0.5 - >64	0.5	>64	60.6
	Ceftriaxone	0.5 - 64	0.5	64	76.2
	Imipenem	0.5 - 64	0.5	1	99.2
	Levofloxacin	0.25 - >64	0.25	8	80.8
<i>E. aerogenes</i> (283)	Tigecycline	0.25 - 4	1	2	96.1
	Amox/Clav	1 - >64	64	64	3.9
	Amp/Sulb	1 - >32	32	>32	14.8
	Cefepime	0.5 - >64	0.5	4	94.3
	Ceftazidime	0.5 - >64	16	>64	49.5
	Ceftriaxone	0.5 - >64	1	16	86.6
	Imipenem	0.5 - 64	1	1	96.1
	Levofloxacin	0.25 - >64	0.25	16	73.1
<i>E. cloacae</i> (280)	Tigecycline	0.5 - 8	1	1	97.1
	Amox/Clav	2 - >64	64	>64	3.2
	Amp/Sulb	2 - >32	>32	>32	12.5
	Cefepime	0.5 - 64	0.5	4	98.2
	Ceftazidime	0.5 - >64	0.5	>64	66.8
	Ceftriaxone	0.5 - >64	0.5	32	76.8
	Imipenem	0.5 - 8	0.5	1	99.6
	Levofloxacin	0.25 - 64	0.25	0.5	94.6
<i>E. coli</i> (400)	Tigecycline	0.12 - 2	0.25	0.5	100
	Amox/Clav	0.5 - >64	8	32	64.0
	Amp/Sulb	0.5 - >32	16	>32	34.3
	Cefepime	0.5 - >64	0.5	32	84.5
	Ceftazidime	0.5 - >64	0.5	>64	73.0
	Ceftriaxone	0.5 - >64	0.5	>64	76.3
	Imipenem	0.5 - 1	0.5	0.5	100.0
	Levofloxacin	0.25 - >64	0.25	16	70.3
<i>K. pneumoniae</i> (536)	Tigecycline	0.12 - 8	0.5	2	95.9
	Amox/Clav	1 - >64	8	32	61.4
	Amp/Sulb	2 - >32	16	>32	38.4
	Cefepime	0.5 - >64	1	16	88.1
	Ceftazidime	0.5 - >64	4	>64	54.1
	Ceftriaxone	0.5 - >64	2	64	70.3
	Imipenem	0.5 - 4	0.5	0.5	100.0
	Levofloxacin	0.25 - >64	0.25	4	85.4

* %Sus = Percent Susceptible, based upon approved NCCLS breakpoints where available, from NCCLS document M100-S12; Tigecycline tentative susceptible breakpoint defined as ≤ 2 mg/mL

Table 3. In Vitro MIC Values and Susceptibility Results of Tigecycline and Comparators Against 420 ESBL Producers and 516 Non-ESBL Producing *E. coli* and *K. pneumoniae*

Organism / Phenotype	Drug	MIC (mg/mL)			% Sus*
		Range	MIC ₅₀	MIC ₉₀	
<i>E. coli</i> (142) ESBL producers	Tigecycline	0.25 - 2	0.25	1	100
	Amox/Clav	2 - >64	16	32	49.3
	Amp/Sulb	4 - >32	32	>32	15.5
	Cefepime	0.5 - >64	8	64	57.0
	Ceftazidime	0.5 - >64	32	>64	26.1
	Ceftriaxone	0.5 - >64	32	>64	33.8
	Imipenem	0.5 - 1	0.5	0.5	100.0
	Levofloxacin	0.25 - >64	4	32	50.0
<i>E. coli</i> (258) ESBL non-producers	Tigecycline	0.12 - 1	0.25	0.5	100
	Amox/Clav	0.5 - >64	8	32	72.1
	Amp/Sulb	0.5 - >32	16	>32	44.6
	Cefepime	0.5 - 16	0.5	0.5	99.6
	Ceftazidime	0.5 - >64	0.5	0.5	98.8
	Ceftriaxone	0.5 - >64	0.5	0.5	99.6
	Imipenem	0.5 - 1	0.5	0.5	100.0
	Levofloxacin	0.25 - >64	0.25	8	81.4
<i>K. pneumoniae</i> (278) ESBL producers	Tigecycline	0.25 - 8	1	2	97.1
	Amox/Clav	2 - >64	16	32	38.8
	Amp/Sulb	4 - >32	>32	>32	9.7
	Cefepime	0.5 - >64	2	32	78.1
	Ceftazidime	0.5 - >64	>64	>64	15.8
	Ceftriaxone	0.5 - >64	16	>64	45.7
	Imipenem	0.5 - 4	0.5	0.5	100.0
	Levofloxacin	0.25 - >64	0.5	8	78.1
<i>K. pneumoniae</i> (258) ESBL non-producers	Tigecycline	0.12 - 8	0.5	1	94.6
	Amox/Clav	1 - >64	2	16	85.7
	Amp/Sulb	2 - >32	8	>32	69.4
	Cefepime	0.5 - >64	0.5	0.5	98.8
	Ceftazidime	0.5 - >64	0.5	1	95.3
	Ceftriaxone	0.5 - >64	0.5	0.5	96.9
	Imipenem	0.5 - 1	0.5	0.5	100.0
	Levofloxacin	0.25 - 64	0.25	0.5	93.4

* %Sus = Percent Susceptible, based upon approved NCCLS breakpoints where available, from NCCLS document M100-S12; Tigecycline tentative susceptible breakpoint defined as ≤ 2 mg/mL

CONCLUSIONS

- Tigecycline inhibited 97.1% of all tested enterobacteriaceae in vitro at an MIC of 2 µg/mL.
- Tigecycline's MIC₉₀ of 1 µg/mL was equivalent to imipenem and 8 to 64 fold better than the beta-lactams, beta-lactam/beta-lactamase inhibitor combinations and levofloxacin against all enterobacteriaceae tested.
- Tigecycline demonstrated potent in vitro activity against both ESBL and non-ESBL producing *E. coli* and *K. pneumoniae*.
- The in vitro activity of tigecycline in this study suggests that tigecycline is a promising compound in the treatment of Gram-negative infections caused by selected enterobacteriaceae.

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