Poster #A1-1376

Pharmacodynamic Activity of Ceftobiprole Compared to Vancomycin vs. *Enterococcus faecalis* Using an In Vitro Model

G. G. Zhanel, N. Laing, L. Cox, E. Rossnagel, D. Voth, J. A. Karlowsky, and D. J. Hoban

Department of Medical Microbiology, Faculty of Medicine, University of Manitoba;
Departments of Clinical Microbiology and Medicine, Health Sciences Centre, Winnipeg, Canada

Dr. George Zhanel
Health Sciences Centre
MS673-820 Sherbrook St.
Winnipeg, MB, Canada R3A 1R9
Phone: (204) 787-4902
Email: ggzhanel@pcs.mb.ca

ABSTRACT

Background: Ceftobiprole is a novel cephalosporin with broad spectrum activity including MRSA. In addition, unlike other cephalosporins, Ceftobiprole is active versus *E. faecalis*. This study compared the pharmacodynamic (PD) activity of Ceftobiprole and Vancomycin versus *E. faecalis* using an in vitro model.

Methods: Ten *E. faecalis* clinical strains obtained from hospitalized patients in Canada were studied. The in vitro PD model was inoculated with $1x10^6$ cfu/mL. Ceftobiprole was dosed every 8 hours at 0, 8 and 16 hours to simulate fC_{max} and $t_{1/2}$ obtained after a standard 500mg intravenous every 8 hour dose in healthy volunteers (fC_{max} 30mg/L, t½ 3.5hrs). Vancomycin was dosed every 12 hours at 0 and 12 hours to simulate fC_{max} and $t_{1/2}$ obtained after a standard 1g intravenous every 12 hour dose in healthy volunteers (fC_{max} 20mg/L, t½ 8hrs). Sampling was performed over 24h to assess viable growth.

Results: Ceftobiprole $T_{>MIC}$ ≥ 100% (ceftobiprole MICs were all ≤1mg/L) resulted in bactericidal (≥3 log_{10} killing) activity against *E. faecalis* at 16 and 24 hours with no regrowth over the 24 hour study period. Ceftobiprole log_{10} killing (±SD) [relative to the starting inoculum] at 8, 16 and 24 hours is displayed below:

E. faecalis Strain (Ceftobiprole MIC, mg/L)	T>MIC (hr) [%]	log_{10} killing 8 hrs	log_{10} killing 16 hrs	log_{10} killing 24 hrs
84107 (0.5)	24 [100]	3.0±0.4	3.5±0.5	3.1±0.5
80889 (1.0)	24 [100]	1.8±0.4	3.6±0.5	3.2±0.5
81357 (0.5)	24 [100]	1.7±0.4	3.2±0.4	3.0±0.7
81417 (0.5)	24 [100]	1.5±0.4	3.0±0.5	3.0±0.5
81449 (0.5)	24 [100]	2.0±0.5	3.1±0.5	3.0±0.4
82065 (0.12)	24 [100]	3.1±0.5	3.5±0.4	3.1±0.5
82597 (0.25)	24 [100]	2.1±0.3	3.0±0.3	3.3±0.5
83132 (0.5)	24 [100]	2.1±0.5	3.0±1.0	3.3±0.9
84014 (0.12)	24 [100]	2.0±0.4	3.0±0.5	3.2±0.4
77780 (0.5)	24 [100]	2.1±0.5	3.0±0.5	3.2±0.5

Vancomycin $fAUC_{24}$/MIC of 340 (vancomycin MIC = 1mg/L against 8 strains) resulted in 1.6-1.8 log_{10} reduction at 24 hours, while vancomycin $fAUC_{24}$/MIC 170 (vancomycin MIC = 2mg/L against 2 strains) resulted in 0.4-0.8 log_{10} reduction at 24 hours.

Conclusion: Ceftobiprole $T_{>MIC}$ ≥ 100% (ceftobiprole MICs ≤1mg/L) resulted in bactericidal (≥3 log_{10} killing) activity against *E. faecalis* at 16 and 24 hours with no regrowth over the 24 hour study period. Vancomycin was bacteriostatic against *E. faecalis*.

BACKGROUND

Ceftobiprole, a cephalosporin, currently in Phase III clinical development in the US and UK, but has recently been approved for use in patients in both Canada and Switzerland. Ceftobiprole is a broad-spectrum cephalosporin with demonstrated *in vitro* activity against Gram-positive cocci, including MRSA and methicillin-resistant *Staphylococcus epidermidis* (MRSE), penicillin-resistant *Streptococcus pneumoniae*, *Enterococcus faecalis*, Gram-negative bacilli including AmpC producing *Escherichia coli* and *Pseudomonas aeruginosa* but excluding extended-spectrum β-lactamase (ESBL)-producing strains (1). Ceftobiprole is now being developed by Basilea Pharmaceutica AG. Currently, ceftobiprole has completed Phase III trials for complicated skin and skin structure infections due to MRSA, nosocomial pneumonia due to suspected or proven MRSA, while Phase III trials are ongoing for community-acquired pneumonia.

Unlike other cephalosporins, ceftobiprole is active against *E. faecalis*, an important cause of healthcare-acquired urinary tract infections and wound infections (1-3).

PURPOSE

The purpose of this study was to assess the pharmacodynamic activity of ceftobiprole and vancomycin against *E. faecalis* using an *in vitro* model.

RESULTS

Table 1. Susceptibility profiles (MIC, mg/L) of 10 *Enterococcus faecalis* strains to ceftobiprole, vancomycin, and comparator agents

Strain	Ceftobiprole	Vancomycin	Linezolid	Daptomycin
E. faecalis 82065	0.12	1	2	0.5
E. faecalis 84014	0.12	2	2	0.5
E. faecalis 82597	0.25	1	1	1
E. faecalis 84107	0.5	1	2	0.25
E. faecalis 77780	0.5	1	2	0.5
E. faecalis 81357	0.5	1	1	0.5
E. faecalis 81417	0.5	1	2	0.5
E. faecalis 81449	0.5	1	2	0.5
E. faecalis 83132	0.5	1	2	0.25
E. faecalis 80889	1	2	2	2

Table 2. Ceftobiprole pharmacodynamic parameters achieved in pharmacodynamic model

Strain	Ceftobiprole MIC (mg/L)	$T_{>MIC}$ h [%]	fC_{max} /MIC	$fAUC_{24}$ /MIC
E. faecalis 82065	0.12	24 [100]	224	688
E. faecalis 84014	0.12	24 [100]	224	688
E. faecalis 82597	0.25	24 [100]	112	344
E. faecalis 84107	0.5	24 [100]	56	172
E. faecalis 77780	0.5	24 [100]	56	172
E. faecalis 81357	0.5	24 [100]	56	172
E. faecalis 81417	0.5	24 [100]	56	172
E. faecalis 81449	0.5	24 [100]	56	172
E. faecalis 83132	0.5	24 [100]	56	172
E. faecalis 80889	1	24 [100]	28	86

Table 3. Vancomycin pharmacodynamic parameters achieved in pharmacodynamic model

Strain	Vancomycin MIC (mg/L)	$T_{>MIC}$ h [%]	fC_{max} /MIC	$fAUC_{24}$ /MIC
E. faecalis 82065	1	24 [100]	19.2	340
E. faecalis 84014	2	21 [87.5]	9.6	170
E. faecalis 82597	1	24 [100]	19.2	340
E. faecalis 84107	1	24 [100]	19.2	340
E. faecalis 77780	1	24 [100]	19.2	340
E. faecalis 81357	1	24 [100]	19.2	340
E. faecalis 81417	1	24 [100]	19.2	340
E. faecalis 81449	1	24 [100]	19.2	340
E. faecalis 83132	1	24 [100]	19.2	340
E. faecalis 80889	2	21 [87.5]	9.6	170

Table 4. Ceftobiprole killing of 10 strains of *E. faecalis* in a pharmacodynamic model simulating free serum concentrations

Strain	Log_{10} killing at 8, 16 and 24 h, respectively[a]		
	8 h	16 h	24 h
E. faecalis 82065	3.1±0.5	3.5±0.4	3.1±0.5
E. faecalis 84014	2.0±0.4	3.0±0.5	3.2±0.4
E. faecalis 82597	2.1±0.3	3.0±0.3	3.3±0.5
E. faecalis 84107	3.0±0.4	3.5±0.5	3.1±0.5
E. faecalis 77780	2.1±0.5	3.0±0.5	3.2±0.5
E. faecalis 81357	1.7±0.4	3.2±0.4	3.0±0.7
E. faecalis 81417	1.5±0.4	3.0±0.5	3.0±0.5
E. faecalis 81449	2.0±0.5	3.1±0.5	3.0±0.4
E. faecalis 83132	2.1±0.5	3.0±1.0	3.3±0.9
E. faecalis 80889	1.8±0.4	3.5±0.5	3.1±0.5

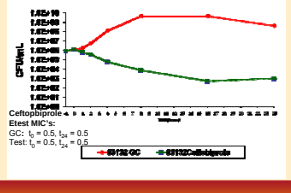
[a] = growth reduction relative to initial inoculum

Table 5. Vancomycin killing of 10 strains of *E. faecalis* in a pharmacodynamic model simulating free serum concentrations

Strain	Log_{10} killing at 8, 16 and 24 h, respectively[a]		
	8 h	16 h	24 h
E. faecalis 82065	1.2±0.3	1.4±0.5	1.5±0.5
E. faecalis 84014	0.3±0.3	0.6±0.4	0.8±0.4
E. faecalis 82597	1.1±0.6	1.0±0.4	2.0±0.6
E. faecalis 84107	0.3±0.5	1.1±0.4	1.6±0.6
E. faecalis 77780	0.8±0.5	1.0±0.5	1.8±0.6
E. faecalis 81357	0.5±0.3	0.8±0.4	1.5±0.5
E. faecalis 81417	0.9±0.5	1.2±0.4	1.7±0.5
E. faecalis 81449	0.6±0.4	0.8±0.4	1.4±0.4
E. faecalis 83132	0.6±0.4	1.0±0.4	1.5±0.5
E. faecalis 80889	0.2±0.5	0.6±0.5	0.4±0.4

[a] = growth reduction relative to initial inoculum

Figure 1. *In Vitro* Pharmacodynamic Modeling of Ceftobiprole (fC_{max} 30 mg/L, t½ 3.5 hrs) against *E. faecalis* 83132 (MIC: Ceftobiprole 0.5 mg/L)



CONCLUSIONS

Ceftobiprole $T_{>MIC}$ ≥ 100% (ceftobiprole MICs ≤1mg/L) resulted in bactericidal (≥3 log_{10} killing) activity against *E. faecalis* at 16 and 24 hours with no regrowth over the 24 hour study period. Vancomycin was bacteriostatic against *E. faecalis*.

ACKNOWLEDGMENTS

This study was supported in part by the University of Manitoba and Janssen Ortho Inc.

MATERIALS & METHODS

Bacterial strains. Ten *E. faecalis* strains were studied (Table 1). All isolates were clinical strains obtained from a national surveillance study assessing antimicrobial resistance in Canadian hospitals (CANWARD) [3].

Antibiotic preparations and susceptibility testing. Ceftobiprole was obtained as laboratory-grade powder from the manufacturer (Ortho-McNeil, New Jersey, USA). Vancomycin was purchased commercially. All MICs were determined by the CLSI broth microdilution method.

Pharmacokinetics of antibiotics in the in vitro pharmacodynamic model. Experiments were performed simulating peak free drug serum concentrations (fC_{max}) and $fAUC_{24}$ of ceftobiprole and vancomycin achieved in human serum after standard intravenous doses in health volunteers (ceftobiprole dose 500mg every 8 h [fC_{max} 30mg/L]; vancomycin 1 Gram dose every 12 h [fC_{max} 20mg/L]) [4]. Protein free (f), also known as unbound, serum concentrations were simulated using known protein binding fractions (ceftobiprole, ~16%; vancomycin ~50%). Ceftobiprole clearance was simulated using a reported serum half-life of 3.5 h, while for vancomycin a half-life of 8 h was simulated. The pharmacokinetics of ceftobiprole and vancomycin were evaluated by dosing using standard doses in the central compartment and sampling from this compartment at 0, 1, 2, 4, 6, 8, 12, 16 and 24 h. Ceftobiprole concentrations were determined in quadruplicate using *Escherichia coli* ATCC 25922 as the test organism with a lower limit of quantification of 0.25mg/L and was linear from 0.25mg/L to 128mg/L. Vancomycin concentrations were assessed using a fluorescence polarization immunoassay (Abbott Diagnostics TDx). The $fAUC_{24}$ (mg.h/L) for ceftobiprole and vancomycin were calculated using the trapezoidal rule. The $fAUC_{24}$/MIC was calculated for ceftobiprole and vancomycin against each of the *E. faecalis* strains studied.

In vitro pharmacodynamic model/pharmacodynamic experiments. The *in vitro* pharmacodynamic model used in this study has been previously described [4]. This initial inoculum was introduced into the central compartment (volume; 610 mL) of the *in vitro* pharmacodynamic model and exposed to ceftobiprole, simulating free (protein unbound) serum concentrations obtained after standard 500mg every 8 h or vancomycin 1 Gram every 12 h, dosing. Pharmacodynamic experiments were performed in ambient air at 37°C. At 0, 1, 2, 4, 6, 8, 12, 16 and 24 h, samples were removed from the central compartment and viable bacterial counts performed by plating serial 10-fold dilutions onto cation-supplemented Mueller-Hinton agar.

REFERENCES

1. Zhanel GG, Lam A, Schweizer F *et al.* Ceftobiprole: A review of a broad spectrum. *Am J Clin Derm* 2008;**9**(4):419-29.
2. Zhanel GG, DeCorby M, Laing N *et al.* Antimicrobial resistant pathogens in intensive care units across Canada: Results of the Canadian National Intensive Care Unit (CAN-ICU) Study, 2005/2006. *Antimicrob Agents Chemother* 2008;**52**:1430-7.
3. Zhanel GG, Karlowsky JA, DeCorby M, Nichol KA, Wierzbowski A, Baudry PJ, Lagace-Wiens P, Walkty A, Schweizer F, McCracken M, Mulvey MR, The Canadian Antimicrobial Resistance Alliance (CARA) and Hoban DJ. Prevalence of antimicrobial resistant pathogens in Canadian hospitals: Results of the Canadian ward surveillance study (CANWARD 2007). *Canadian Journal of Infectious Diseases and Medical Microbiology* 2009;**20** (Suppl A):9-19.
4. Zhanel GG, Voth D, Nichol K, Karlowsky JA, Noreddin AM and Hoban DJ. Pharmacodynamic activity of ceftobiprole to vancomycin versus methicillin-resistant *Staphylococcus aureus* (MRSA), vancomycin-intermediate *Staphylococcus aureus* (VISA) and vancomycin-resistant *Staphylococcus aureus* (VRSA) using an in vitro model. *Journal of Antimicrobial Chemotherapy* 2009;**64**:364-369.