E-137

In Vitro Activity of Ceftazidime Avibactam (CAZ104) Against Pathogens Collected During a Phase II Complicated Urinary Tract Infection (cUTI) Clinical Trial

DJ Biedenbach, ML Konrardy, HS Sader, RK Flamm, RN Jones

JMI Laboratories, North Liberty, Iowa, USA

Contact Information:
Douglas J Biedenbach
JMI Laboratories
345 Beaver Kreek Centre, Suite A
North Liberty, IA 52317
Tel: 001-319-665-3370 (ext. 220)
Fax: 001-319-665-3371
E-mail: douglas-biedenbach@jmilabs.com

Amended abstract

Background: Avibactam (NXL104) is a novel non-β-lactam β-lactamase (BL) inhibitor that inhibits Ambler class A, C, and D enzymes, and ceftazidime avibactam (CAZ104) is active against ESBL and AmpC producing bacteria. This study aimed to determine the *in vitro* activity of CAZ104 and comparators against pathogens collected during a Phase II trial of CAZ104 vs. imipenem (IMP) in adults with cUTI.

Methods: The trial was conducted in 5 countries/23 sites. Isolates from urine (220) or blood (18) were collected from 124 patients. Species included *E. coli* (186), other *Enterobacteriaceae* (ENT; 23), *P. aeruginosa* (PSA; 23) and other non-fermentative Gram-negative bacilli (NFB, 6). Susceptibility (S) testing was performed by a reference laboratory using CLSI broth microdilution for ceftazidime, cefotaxime, IMP, 2 BL inhibitor combinations (BLIC; amoxicillin-clavulanate, piperacillin-tazobactam), ciprofloxacin (CIP) and tigecycline according to CLSI and EUCAST breakpoints.

Results: The major pathogen was *E. coli* (39.8% ESBL phenotype). The highest CAZ104 MIC for *E. coli* was 0.5 µg/mL. $MIC_{50/90}$ values for ESBL producing strains (0.12/0.25 µg/mL) were 2-fold higher than non-ESBL strains (0.06/0.12 µg/mL), comparable to IMP ($MIC_{50/90}$, 0.12/0.12 µg/mL). MIC_{90} values for the other BLICs for all *E. coli* was 16 µg/mL, and CIP-S was 40.3% for *E. coli.* CAZ104 $MIC_{50/90}$ values for ENT, PSA and NFB were 0.06/0.25, 2/8 and 4/- µg/mL, respectively. CAZ104 (MIC_{90}, 8 µg/mL) was 4-fold more active than CAZ (MIC_{90}, 32 µg/mL) against PSA and CIP-S was only 26.1%.

Conclusion: CAZ104 is a potent β-lactam-β-lactamase inhibitor combination with potential for treating cUTI caused by common multidrug-resistant uropathogens. These results demonstrate that CAZ104 has significant activity against *E. coli*, the most common cause of cUTI, (including those harboring ESBL enzymes) and other ENT.

Introduction

The continued emergence of multidrug-resistant (MDR) Gram-negative bacteria limits potential therapeutic options for patients with complicated urinary tract infection (cUTI). In particular, the Gram-negative bacteria *Escherichia coli*, *Klebsiella pneumoniae*, *Enterobacter* spp. and *Pseudomonas aeruginosa* are a significant problem due to the increasing prevalence of MDR strains.

An approach to developing new agents to address the issue of resistance is to combine an established agent such as ceftazidime with a novel β-lactamase inhibitor which protects the co-drug from hydrolysis by β-lactamase enzymes, and enhances and expands the spectrum of the antibacterial. Ceftazidime plus avibactam (previously known as NXL104) (ceftazidime avibactam [CAZ104]) is such a combination which consists of the novel non-β-lactam β-lactamase inhibitor avibactam that inhibits Ambler class A, C, and some D enzymes with ceftazidime. This combination (ceftazidime avibactam), is active against extended-spectrum β-lactamases (ESBL)- and AmpC-producing bacteria.

The aim of this study was to determine the *in vitro* activity of ceftazidime avibactam and comparators against pathogens collected during a Phase II trial of ceftazidime avibactam versus imipenem in adults with cUTI.

Materials and methods

Isolate collection: Isolates were collected at study visits via urine culture, either midstream clean catch or catheterized urine from patients during a Phase II cUTI trial conducted in five countries; 23 sites in total. Blood cultures were performed when clinically indicated or in patients with indwelling catheters and stents. There were 220 isolates from urine, and 18 isolates from blood which were collected from 124 patients. Isolates included 186 *E. coli*, 23 other *Enterobacteriaceae*, 23 *P. aeruginosa* and six other non-fermentative Gram-negative bacilli.

Susceptibility testing: Isolates were susceptibility tested by a reference broth microdilution procedure (Clinical and Laboratory Standards Institute [CLSI], 2009) using validated microdilution panels manufactured by TREK Diagnostics (Cleveland, Ohio, USA). Antimicrobials tested included ceftazidime, cefotaxime, imipenem, two β-lactam-β-lactamase inhibitor combinations (amoxicillin/clavulanate, piperacillin/tazobactam), ciprofloxacin and tigecycline. Susceptibility testing results were interpreted according to CLSI or EUCAST criteria (CLSI, 2011; EUCAST, 2011); for ceftazidime avibactam, results were interpreted according to CLSI or EUCAST criteria for ceftazidime tested alone. *E. coli* ATCC 25922 and *P. aeruginosa* ATCC 27853 were concurrently tested for quality assurance; all results were within the published ranges.

E. coli and *Klebsiella* spp. isolates for which the MIC results of ceftriaxone or ceftazidime, or aztreonam were ≥2 µg/mL were considered to be phenotype-positive for ESBL production (CLSI, 2011).

Table 1. *Distribution of isolates by region and country*

Region and country	No. medical centers	No. of isolates
Middle East		
Jordan	3	46
Lebanon	5	79
Total	8	125
Latin America		
Guatemala	5	73
Asia-Western Pacific		
India	6	21
North America		
United States	4	19

Table 2. *Cumulative frequency distribution of ceftazidime avibactam MIC values for Gram-negative pathogens isolated from subjects in a Phase II cUTI trial*

Species (no. tested)	No. (cumulative %) of isolates inhibited at ceftazidime avibactam MIC (µg/mL)[a]								
	≤0.03	0.06	0.12	0.25	0.5	1	2	4	8
E. coli (186)	30 (16.1)	45 (40.3)	87 (87.1)	22 (98.9)	2 (100.0)	-	-	-	-
Wildtype (112)	25 (22.3)	38 (56.3)	43 (94.6)	6 (100.0)	-	-	-	-	-
ESBL (74)	5 (6.8)	7 (16.2)	44 (75.7)	16 (97.3)	2 (100.0)	-	-	-	-
Enterobacteriaceae (23)	6 (26.1)	10 (69.6)	3 (82.6)	3 (95.7)	0 (95.7)	0 (95.7)	1 (100.0)	-	-
P. aeruginosa (23)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (4.3)	3 (17.4)	8 (52.2)	7 (82.6)	4 (100.0)
Non-fermentative Gram-negative bacilli (6)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (16.7)	0 (16.7)	1 (33.3)	2 (66.7)	1 (83.3[b])

[a]Concentrations reported in the table for ceftazidime avibactam refer to the concentration of ceftazidime tested with fixed 4 µg/mL avibactam concentration; [b]One *Acinetobacter baumannii* isolate had a MIC value of 16 µg/mL

Table 3. *Activity of ceftazidime avibactam and comparator antimicrobial agents tested against 3233 Gram-negative pathogens isolated from subjects in a Phase II cUTI trial*

Antimicrobial agent (no. tested)	MIC_{50}	MIC_{90}	Range	CLSI[a] %S / %R	EUCAST[a] %S / %R
E. coli (186)					
Ceftazidime avibactam	0.12	0.25	≤0.03 – 0.5	- / -	- / -
Ceftazidime	0.25	32	≤0.03 – >32	64.5 / 33.3	60.2 / 35.5
Ciprofloxacin	>16	>16	≤0.015 – >16	40.3 / 59.7	35.5 / 59.7
Imipenem	0.12	0.12	≤0.03 – 0.5	100.0 / 0.0	100.0 / 0.0
Amoxicillin/clavulanate	16	16	1 – >32	44.1 / 9.7	- / 55.9
Piperacillin/tazobactam	4	16	0.5 – >64	90.9 / 5.4	83.3 / 9.1
Tigecycline[b]	0.12	0.25	≤0.03 – 1	100.0 / 0.0	100.0 / 0.0
Wildtype *E. coli* (112)					
Ceftazidime avibactam	0.06	0.12	≤0.03 – 0.25	- / -	- / -
Ceftazidime	0.12	0.5	≤0.03 – 1	100.0 / 0.0	100.0 / 0.0
Ciprofloxacin	0.25	>16	≤0.015 – >16	57.1 / 42.9	51.8 / 42.9
Imipenem	0.12	0.12	≤0.03 – 0.25	100.0 / 0.0	100.0 / 0.0
Amoxicillin/clavulanate	8	16	1 – >32	64.3 / 5.4	- / 35.7
Piperacillin/tazobactam	2	8	0.5 – >64	92.9 / 3.6	91.1 / 7.1
Tigecycline[b]	0.12	0.25	≤0.03 – 1	100.0 / 0.0	100.0 / 0.0
ESBL *E. coli* (74)					
Ceftazidime avibactam	0.12	0.25	≤0.03 – 0.5	- / -	- / -
Ceftazidime	32	>32	2 – >32	10.8 / 83.8	0.0 / 89.2
Ciprofloxacin	>16	>16	≤0.015 – >16	14.9 / 85.1	10.8 / 85.1
Imipenem	0.12	0.12	≤0.03 – 0.5	100.0 / 0.0	100.0 / 0.0
Amoxicillin/clavulanate	16	32	4 – >32	13.5 / 16.2	- / 86.5
Piperacillin/tazobactam	8	64	1 – >64	87.8 / 8.1	71.6 / 12.2
Tigecycline[b]	0.12	0.25	0.06 – 1	100.0 / 0.0	100.0 / 0.0
Enterobacteriaceae (23)[c]					
Ceftazidime avibactam	0.06	0.25	≤0.03 – 2	- / -	- / -
Ceftazidime	0.06	0.5	≤0.03 – >32	91.3 / 8.7	91.3 / 8.7
Ciprofloxacin	0.03	>16	≤0.015 – >16	78.3 / 17.4	69.6 / 21.7
Imipenem	0.25	0.5	0.06 – 2	95.7 / 0.0	100.0 / 0.0
Amoxicillin/clavulanate	4	>32	1 – >32	65.2 / 26.1	- / 34.8
Piperacillin/tazobactam	2	8	0.12 – >64	95.7 / 4.3	95.7 / 4.3
Tigecycline[b]	0.25	4	0.12 – 4	87.0 / 0.0	78.3 / 13.0
P. aeruginosa (23)					
Ceftazidime avibactam	2	8	0.5 – 8	- / -	- / -
Ceftazidime	4	32	1 – 32	87.0 / 13.0	87.0 / 13.0
Ciprofloxacin	16	>16	0.12 – >16	26.1 / 65.2	26.1 / 73.9
Imipenem	1	8	0.06 – 16	87.0 / 8.7	87.0 / 8.7
Piperacillin/tazobactam	16	>64	2 – >64	78.3 / 21.7	65.2 / 34.8
Other non-fermentative Gram-negative bacilli[d] (6)					
Ceftazidime avibactam	4	-	0.5 – 16	- / -	- / -
Ceftazidime	4	-	2 – >32	- / -	- / -
Ciprofloxacin	0.12	-	0.06 – >16	- / -	- / -
Imipenem	0.06	-	≤0.03 – >32	- / -	- / -
Amoxicillin/clavulanate	4	-	1 – >32	- / -	- / -
Piperacillin/tazobactam	0.06	-	≤0.03 – >64	- / -	- / -
Tigecycline[b]	0.25	-	0.12 – 8	- / -	- / -

[a]Criteria as published by the CLSI [2011] and EUCAST [2011]. EUCAST provides a resistant only breakpoint for the aminopenicillins to allow the user to determine whether an organism is susceptible or intermediate depending on dosing, route of administration and whether the infection is systemic or affects the urinary tract only. Therefore, only the percentage resistant are presented in the table for amoxicillin/clavulanate; [b]US-FDA breakpoints were applied [Tygacil Product Insert, 2010]; [c]Includes: *Citrobacter freundii* (1 strain), *Citrobacter koseri* (1 strain), *Enterobacter aerogenes* (3 strains), *Enterobacter cloacae* (1 strain), *Klebsiella oxytoca* (1 strain), *K. pneumoniae* (11 strains), *Morganella morganii* (1 strain), *Proteus mirabilis* (2 strains), and *Providencia stuartii* (2 strains); [d]Includes: *A. baumannii* (2 strains), *Acinetobacter junii* (3 strains), and *Stenotrophomonas maltophilia* (1 strain)

Results

- Table 1 shows the overall distribution of isolates by region and country.
- The most common pathogen isolated was *E. coli* (78.2%) followed by other *Enterobacteriaceae* (9.7%), *P. aeruginosa* (9.7%) and other non-fermentative Gram-negative bacteria (2.5%).
- Table 2 shows the cumulative frequency distribution of ceftazidime avibactam MIC values for Gram-negative pathogens isolated, and Table 3 shows the activity of ceftazidime avibactam and comparator antimicrobial agents tested.
- *E. coli*
  - All ceftazidime avibactam MIC values (≤0.5 µg/mL) were below the CLSI or EUCAST susceptible breakpoint criteria for ceftazidime alone (CLSI: susceptible at ≤4 µg/mL and EUCAST: susceptible at ≤1 µg/mL).
  - 39.8% (74/186) of the *E. coli* were ESBL-phenotypes and the ceftazidime avibactam $MIC_{50/90}$ values were 0.12/0.25 µg/mL, which was 2-fold higher than non-ESBL strains (0.06/0.12 µg/mL). Ceftazidime avibactam activity was comparable to imipenem ($MIC_{50/90}$, 0.12/0.12 µg/mL).
  - The MIC_{90} values for all *E. coli* for the β-lactamase inhibitor combinations piperacillin/tazobactam and amoxicillin/clavulanate were 4/16 µg/mL and 16/16 µg/mL, respectively. For the ESBL-phenotype the respective MIC_{90} values were 8/64 µg/mL and 16/32 µg/mL.
  - Only 40.3% (75/186; CLSI) or 35.5% (66/186; EUCAST) of all *E. coli* were ciprofloxacin-susceptible.
- Other Enterobacteriaceae
  - Ceftazidime avibactam was the most potent antimicrobial with $MIC_{50/90}$ values of 0.06/0.25 µg/mL followed by ceftazidime at 0.06/0.5 µg/mL and imipenem at 0.25/0.5 µg/mL, respectively.
- *P. aeruginosa*
  - Imipenem and ceftazidime avibactam were the two most potent antimicrobials with $MIC_{50/90}$ values of 1/8 µg/mL and 2/8 µg/mL, respectively.
  - Ceftazidime avibactam was 4-fold more active than ceftazidime alone and all ceftazidime avibactam MIC values (≤8 µg/mL) were below the CLSI or EUCAST susceptible breakpoint criteria for ceftazidime alone (CLSI and EUCAST; susceptible at ≤8 µg/mL).
  - Ciprofloxacin susceptibility was 26.1% (6/23).

Conclusions

- Ceftazidime avibactam has potent activity against *E. coli*, the most common cause of cUTI (including those harboring ESBL enzymes), and other Enterobacteriaceae.
- Ceftazidime avibactam also has potent activity against the MDR uropathogen *P. aeruginosa*.
- Overall, ceftazidime avibactam was a potent β-lactam-β-lactamase inhibitor combination with potential for treating cUTI caused by common MDR uropathogens, as documented by these results for a Phase II clinical trial conducted in five nations.

Selected references

Clinical and Laboratory Standards Institute (2009). *M07-A8. Methods for dilution antimicrobial susceptibility tests for bacteria that grow aerobically; approved standard: eighth edition.* Wayne, PA: CLSI.

Clinical and Laboratory Standards Institute (2011). *M100-S21. Performance standards for antimicrobial susceptibility testing: 21st informational.* Wayne, PA: CLSI.

Drawz SM, Bonomo RA (2010). Three decades of beta-lactamase inhibitors. *Clin Microbiol Rev* 23: 160-201.

Endimiani A, et al. (2009). *In vitro* activity of NXL104 in combination with beta-lactams against *Klebsiella pneumoniae* isolates producing KPC carbapenemases. *Antimicrob Agents Chemother* 53: 3599-3601.

EUCAST (2011). Breakpoint tables for interpretation of MICs and zone diameters. Version 1.3, January 2011. Available at: http://www.eucast.org/clinical_breakpoints/. Accessed: March 18, 2011.

Livermore DM, et al. (2008). NXL104 combinations versus Enterobacteriaceae with CTX-M extended-spectrum beta-lactamases and carbapenemases. *J Antimicrob Chemother* 62: 1053-1056.

Stachyra T, et al. (2009). *In vitro* activity of the β-lactamase inhibitor NXL104 against KPC-2 carbapenemase and Enterobacteriaceae expressing KPC carbapenemases. *J Antimicrob Chemother* 64: 326-329.

Wagenlehner FME, Naber K (2006). Treatment of bacterial urinary tract infections: Presence and future. *Eur Urol* 49: 235-244.

Wagenlehner FME, et al. (2009). Urinary bactericidal activity of doripenem versus that of levofloxacin in patients with complicated urinary tract infections or pyelonephritis. *Antimicrob Agents Chemother* 53: 1567-1573.

This study at JMI Laboratories was supported by an Educational/Research grant from AstraZeneca, and JMI Laboratories received compensation fees for services in relation to preparing the abstract/poster, which was funded by AstraZeneca. Poster typesetting and printing was provided by Prime Medica Ltd, Knutsford, Cheshire UK, Funded by AstraZeneca. Presented at the 51st Interscience Conference on Antimicrobial Agents and Chemotherapy, September 17–20, 2011, Chicago, IL, USA.

In Vitro Activity of Ceftazidime Avibactam (CAZ104) Against Pathogens Collected During a Phase II Complicated Urinary Tract Infection (cUTI) Clinical Trial

DJ Biedenbach, ML Konrardy, HS Sader, RK Flamm, RN Jones

JMI Laboratories, North Liberty, Iowa, USA

Presented at the 51st Interscience Conference on Antimicrobial Agents and Chemotherapy, September 17–20, 2011, Chicago, IL, USA.