Activity of ACHN-490 Against Complicated Urinary Tract Infection (cUTI) Pathogens From the United States and Europe

F1-843

DJ BIEDENBACH, RN JONES, ES ARMSTRONG, JB AGGEN, GH MILLER
JMI Laboratories, North Liberty, IA, USA; Achaogen Inc., South San Francisco, CA, USA

ICAAC 2009
JMI Laboratories
North Liberty, IA, USA
www.jmilabs.com
319.665.3370, 319.665.3371
douglas-biedenbach@jmilabs.com

AMENDED ABSTRACT

Background: ACHN-490 is a neoglycoside, a next-generation aminoglycoside, in clinical development. This study determined the activity of ACHN-490 against species that cause complicated urinary tract infections (cUTIs) worldwide.

Methods: Most isolates were collected from medical centers in the United States (USA) and Europe (EU; 17 countries) from urine or blood culture with the source of bacteremia documented as a UTI. Strains were susceptibility (S) tested against ACHN-490, gentamicin (GEN), amikacin (AMK) and 10 comparators by CLSI broth microdilution. Pathogens (no. 196) included: *E. coli* (EC), *Klebsiella* spp. (KSP), *Enterobacter* spp. (EBS), *Citrobacter* spp. (CBS), *P. mirabilis* (PM), *M. morganii* (MM), *P. aeruginosa* (PSA), *S. aureus* (SA) and *S. saprophyticus* (SSAP).

Results: MIC_{90} values for ACHN-490 ranged from 0.5 to 2 µg/ml among EC, KSP, EBS and CBS. Higher MIC_{90} values were observed for PM and MM (4 - 8 µg/ml); the highest was PSA (MIC_{90}, 32 µg/ml). ACHN-490 was very active against SSAP with $MIC_{50/90}$ values of ≤0.25 µg/ml. SA isolates, including 47.8% oxacillin-resistant (R) strains, had a MIC_{90} of 2 µg/ml. Overall S rates to GEN and AMK were 84.2 and 93.9%. ACHN-490 inhibited 89.3% of strains at ≤4 µg/ml and and 98.5% of strains at ≤16 µg/ml. Staphylococcal S rates to ciprofloxacin (CIP), pip/tazo (P/T), nitrofurantoin and trim/sulfa (T/S) were 68.9, 73.3, 100 and 100%, respectively. S rates for CIP, P/T and T/S against enteric pathogens were 76.4, 87.4 and 68.5%, respectively. PSA isolates were only 50.0% S to CIP and 79.2% S to P/T.

	Cumulative % inhibited at ACHN-490 MIC (µg/ml)							
Organism/ (no. tested)	≤0.25	0.5	1	2	4	8	16	≥32
E. coli (21)	0.0	19.0	61.9	100.0	-	-	-	-
Klebsiella spp. (23)	21.7	95.7	100.0	-	-	-	-	-
Enterobacter spp. (20)	0.0	70.0	100.0	-	-	-	-	-
Citrobacter spp. (20)	0.0	70.0	90.0	95.0	100.0	-	-	-
P. mirabilis (23)	0.0	0.0	4.3	39.1	87.0	95.7	100.0	-
M. morganii (20)	0.0	0.0	5.0	60.0	90.0	100.0	-	-
P. aeruginosa (24)	0.0	0.0	0.0	0.0	33.3.	66.7	87.5	100.0
S. aureus (23)	0.0	21.7	69.6	95.7	100.0	-	-	-
S. saprophyticus (22)	90.9	95.5	100.0	-	-	-	-	-

Conclusions: ACHN-490 is active against pathogens that are the leading causes of cUTIs, even in the presence of mechanisms causing R to current front-line antimicrobial agents.

INTRODUCTION

Enterobacteriaceae are the most common uropathogens and the most prevalent species often have acquired resistance mechanisms including Amp-C and extended spectrum β-lactamase (ESBL) enzyme production. In addition to resistance to β-lactam agents, resistance to fluoroquinolones and trimethoprim/sulfamethoxazole (TMP/SMX) has been increasing to signficant levels among the Gram-negative enteric species. Multi-drug resistance is common among staphylococci and non-fermentative Gram-negative bacilli such as *Pseudomonas aeruginosa*, which are also considered to be significant (top ten) causes of urinary tract infections (UTI). It is important to provide adequate antimicrobial therapy to patients with uncomplicated, often recurrent, UTI and complicated UTI which can become severe and lead to urosepsis.

Aminoglycosides (AG) target the A-site of the 30S ribosome and have a broad-spectrum of antimicrobial activity, including species commonly associated with UTI. The most commonly prescribed AG agents utilized for the treatment of both Gram-positive and –negative bacterial pathogens in the USA include gentamicin, tobramycin and amikacin. Resistance mechanisms to these and other AGs include inactivation of the drugs by aminoglycoside modifying enzymes (AMEs), decreased permeability and/or upregulated efflux and less commonly, ribosomal alteration. The clinical importance of AMEs is significant as the encoding genes can be disseminated by plasmids or transposons. The AMEs include acetyltransferases, adenylyltransferases and phosphotransferases. This study determined the activity of the neoglycoside, or next generation aminoglycoside, ACHN-490 against nine species that are among the most common causes of UTI (complicated and uncomplicated) worldwide.

MATERIALS AND METHODS

A total of 196 isolates were selected from a recent collection of strains isolated from patients having UTI. The source of the isolates included urine (46 isolates) and bloodstream from patients in which the primary diagnosis was UTI (BSI/UTI; 150 isolates). Isolates were collected from 54 medical centers in the United States (24 states), Latin America (four countries) and Europe (11 countries). One strain was from Taiwan. Among the tested isolates, 83.7% were cultured during 2007-2009; the remaining samples from years prior to 2007 were needed to fulfill the targeted numbers of strains for some species. All isolates were identified to species level by at least two clinical microbiology laboratories including a reference laboratory (JMI Laboratories, North Liberty, Iowa, USA).

Isolates (number) included Enterobacteriaceae as follows: *E. coli* (21), *Klebsiella* spp. (23), *Enterobacter* spp. (20), *Citrobacter* spp. (20), *Proteus mirabilis* (23) and *Morganella morganii* (20). *Staphylococcus aureus* (23), including both methicillin-resistant (11; MRSA) and -susceptible (12; MSSA) strains. *Staphylococcus saprophyticus* (22) and *P. aeruginosa* (24) isolates were also included in this investigation.

Isolates were tested by reference CLSI broth microdilution methods per M07-A8 [2009] using panels manufactured by JMI Laboratories. Quality control (QC) strains *E. coli* ATCC 25922, *P. aeruginosa* ATCC 27853 and *S. aureus* ATCC 29213 were included each day that clinical isolates were tested. Colony counts were tested on all QC strains with inoculum concentrations ranging from 3.1 X 10^5 to 1.1 X 10^6 CFU/ml. Interpretive criteria and QC ranges for comparator agents used in this study were those published in the CLSI M100-S19 (2009) document. Broth microdilution MIC testing (96-well panel) was performed using doubling dilution ranges for ACHN-490, gentamicin, levofloxacin, tigecycline, doripenem, piperacillin/tazobactam, ciprofloxacin, amikacin, TMP/SMX, nitrofurantoin, ceftriaxone, cefoxitin and oxacillin.

RESULTS

- With all UTI pathogens combined (Table 1), ACHN-490 (MIC_{90}, 8 µg/ml) was more potent than gentamicin (MIC_{90}, ≥ 32 µg/ml) and amikacin (MIC_{90}, 16 µg/ml). Amikacin had better overall activity (93.9% susceptibility) than gentamicin (84.2%) at current CLSI breakpoints.
- ACHN-490 (MIC_{90}, 4 µg/ml) was more potent than gentamicin (MIC_{90}, >16 µg/ml) and amikacin (MIC_{90}, 8 µg/ml) against Enterobacteriaceae (Table 2).
- Similar potency was noted for ACHN-490 across four Enterobacteriaceae species tested in this study with MIC_{50} and MIC_{90} values ranging from 0.5 – 1 µg/ml and 0.5 – 2 µg/ml, respectively for *E. coli*, *Klebsiella* spp., *Enterobacter* spp. and *Citrobacter* spp.
- Susceptibility to gentamicin/amikacin was 81.0/95.2, 95.7/95.7, 85.0/100.0 and 85.0/95.0% for *E. coli*, *Klebsiella* spp., *Enterobacter* spp. and *Citrobacter* spp., respectively.
- Higher MIC_{50} and MIC_{90} values were observed for ACHN-490 when tested against *P. mirabilis* (4 and 8 µg/ml) and *M. morganii* (2 and 4 µg/ml) compared to the other Enterobacteriaceae species. Based on the MIC_{50}, ACHN-490 was two- to four-fold less active than gentamicin against these two species but showed activity equal to amikacin. Gentamicin susceptibility rates were 82.6 to 85.0% and amikacin susceptibility rates were 95.7 to 100% against *P. mirabilis* and *M. morganii*, respectively.
- No significant difference was noted in the activity of ACHN-490 against MSSA compared to MRSA (Table 3). In contrast, the potency of amikacin was reduced four- to eight-fold among the MRSA (MIC_{50}, 16 µg/ml) isolates compared to MSSA (MIC_{50}, 4 µg/ml).
- Potent activity was observed for ACHN-490 (MIC_{90}, ≤0.25 µg/ml), gentamicin (MIC_{90}, 0.25 µg/ml) and amikacin (MIC_{90}, 1 µg/ml) when tested against *S. saprophyticus* isolates (Table 3). Susceptibility was >90% for the two marketed aminoglycoside agents.
- Using on-scale MIC_{50} values, the rank order of potency among the aminoglycosides against *P. aeruginosa* was gentamicin (2 µg/ml) > amikacin (4 µg/ml) > ACHN-490 (8 µg/ml), as illustrated in Table 4. However, the gentamicin susceptibility rate at ≤4 µg/ml was only 58.3% compared to 87.5% for amikacin (≤16 µg/ml) against this species.

Table 1. Activity of ACHN-490, gentamicin and amikacin when tested against 196 urinary tract pathogens.

	Cumulative % inhibited at MIC (µg/ml):						
Antimicrobial agent	≤0.5	1	2	4	8	16	≥32
ACHN-490	40.8	58.2	75.5	89.3	95.4	98.5	100.0
Gentamicin	45.4	69.4	83.7	84.2[a]	85.7	86.7	100.0
Amikacin	9.7	19.4	56.1	82.1	89.3	93.9[a]	100.0

a. CLSI susceptible breakpoint.

Table 2. Comparison of the in vitro activity of ACHN-490 and selected antimicrobial agents tested against Enterobacteriaceae species (127 strains).

Antimicrobial agent	MIC_{50}	MIC_{90}	Range	% susceptible/ resistant[a]
ACHN-490	1	4	≤0.25 – 16	- / -
Gentamicin	1	>16	0.25 – >16	85.8 / 13.4
Amikacin	2	8	1 – >64	96.9 / 1.6
Ceftriaxone	≤0.25	>32	≤0.25 – >32	83.5 / 13.4
Cefoxitin	8	>8	≤1 – >8	57.5 / 42.5
Doripenem[b]	≤0.06	≤0.06	≤0.06 – 1	98.4 / -
Piperacillin/tazobactam	2	32	≤0.5 – >64	87.4 / 7.1
Ciprofloxacin	≤0.03	>4	≤0.03 – >4	76.4 / 22.0
Levofloxacin	≤0.06	8	≤0.06 – >8	78.7 / 15.8
Tigecycline[b]	0.5	2	0.12 – 8	96.1 / 0.8
Trim/sulfa[c]	≤0.5	>2	≤0.5 – >2	68.5 / 31.5
Nitrofurantoin	64	>128	≤16 – >128	37.8 / 48.0

a. Criteria as published by the CLSI [2009]. A dash indicates that no susceptibility breakpoints have been established by the CLSI or the FDA.
b. US-FDA breakpoints were applied [Product Inserts].
c. Trimethoprim/sulfamethoxazole.

Table 3. Comparison of the in vitro activity of ACHN-490 and selected antimicrobial agents tested against *Staphylococcus* spp. (45 strains).

Organism (no. tested) Antimicrobial agent	MIC_{50}	MIC_{90}	Range	% susceptible/ Resistant[a]
Oxacillin-susceptible *S. aureus* (12)				
ACHN-490	1	2	0.5 – 2	- / -
Gentamicin	0.5	1	0.25 – 1	100.0 / 0.0
Amikacin	4	4	2 – 8	100.0 / 0.0
Ciprofloxacin	0.5	>4	0.5 – >4	66.7 / 33.3
Levofloxacin	0.25	>8	0.25 – >8	66.7 / 33.3
Tigecycline[b]	0.12	0.12	≤0.06 – 0.25	100.0 / -
Trim/sulfa[c]	≤0.5	≤0.5	≤0.5	100.0 / 0.0
Nitrofurantoin	32	32	≤16 – 32	100.0 / 0.0
Oxacillin-resistant *S. aureus* (11)				
ACHN-490	1	2	0.5 – 4	- / -
Gentamicin	0.5	2	0.25 – >16	90.9 / 9.1
Amikacin	16	32	2 – 64	54.5 / 9.1
Ciprofloxacin	>4	>4	0.5 – >4	18.2 / 81.8
Levofloxacin	>8	>8	0.25 – >8	18.2 / 81.8
Tigecycline[b]	0.12	0.25	≤0.06 – 0.25	100.0 / -
Trim/sulfa[c]	≤0.5	≤0.5	≤0.5	100.0 / 0.0
Nitrofurantoin	≤16	32	≤16 – 32	100.0 / 0.0
S. saprophyticus (22)				
ACHN-490	≤0.25	≤0.25	≤0.25 – 1	- / -
Gentamicin	≤0.12	0.25	≤0.12 – 16	90.9 / 4.5
Amikacin	≤0.5	1	≤0.5 – 2	100.0 / 0.0
Oxacillin	0.5	1	≤0.25 – >2	4.5 / 95.5
Ciprofloxacin	0.5	0.5	0.12 – >4	95.5 / 4.5
Levofloxacin	0.5	1	0.25 – >8	95.5 / 4.5
Tigecycline[b]	0.25	0.25	0.12 – 0.25	- / -
Trim/sulfa[c]	≤0.5	≤0.5	≤0.5 – 1	100.0 / 0.0
Nitrofurantoin	32	32	≤16 – 32	100.0 / 0.0

a. Criteria as published by the CLSI [2009]. A dash indicates that no susceptibility breakpoints have been established by the CLSI or the FDA.
b. US-FDA breakpoints were applied [Tygacil Product Insert, 2005].
c. Trimethoprim/sulfamethoxazole.

Table 4. Comparison of the in vitro activity of ACHN-490 and selected antimicrobial agents tested against *P. aeruginosa* (24 strains).

Antimicrobial agent	MIC_{50}	MIC_{90}	Range	% susceptible/ resistant[a]
ACHN-490	8	32	4 – >32	- / -
Gentamicin	2	>16	1 – >16	58.3 / 37.5
Amikacin	4	32	2 – >64	87.5 / 8.3
Ceftriaxone	32	>32	8 – >32	4.2 / 41.7
Cefoxitin	>8	>8	>8	- / -
Doripenem	1	>8	0.12 – >8	70.8 / -
Piperacillin/tazobactam	8	>64	4 – >64	79.2 / 20.8
Ciprofloxacin	0.25	>4	≤0.03 – >4	50.0 / 50.0
Levofloxacin	1	>8	0.25 – >8	50.0 / 45.8
Tigecycline	8	>8	4 – >8	- / -
Trim/sulfa[b]	>2	>2	2 – >2	8.3 / 91.7
Nitrofurantoin	>128	>128	>128	- / -

a. Criteria as published by the CLSI [2009]. A dash indicates that no susceptibility breakpoints have been established by the CLSI or the FDA.
b. Trimethoprim/sulfamethoxazole.

CONCLUSIONS

- ACHN-490 exhibited better potency compared to gentamicin and amikacin when tested against several Gram-positive and –negative pathogens commonly isolated from patients with UTIs, including antimicrobial-resistant subsets.
- Higher ACHN-490 MICs were noted with species of Proteae than other Enterobacteriaceae.
- ACHN-490 exhibited excellent activity against both MSSA and MRSA isolates. *S. saprophyticus* is a well known Gram-positive pathogen associated with UTI in younger females and excellent potency (MIC_{90}, ≤0.25 µg/ml) was observed for ACHN-490 against this species.
- ACHN-490 had lower baseline potency than gentamicin, but gentamicin resistance among *P. aeruginosa* isolates was high (37.5%). This resulted in three times fewer ACHN-490 MICs >16 mg/ml than occurred with gentamicin
- Based on these in vitro microbiology results, ACHN-490 is a promising new agent for the treatment of UTI.
- Although no effort was made to select resistant isolates for this study, the susceptibility rates to common UTI therapeutic agents are low overall, emphasizing the need for a new agent.

REFERENCES

1. 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.
2. Clinical and Laboratory Standards Institute. (2009). *M100-S19, Performance standards for antimicrobial susceptibility testing, 19th informational supplement.* Wayne, PA., CLSI.
3. Jana S, Deb JK (2006). Molecular understanding of aminoglycoside action and resistance. *Appl Microbiol Biotechnol.* 70:140-150.
4. Shaw KJ, Rather PN, Hare RS, Miller GH (1993). Molecular genetics of aminoglycoside resistance genes and familial relationships of the aminoglycoside-modifying enzymes. *Microbiol Rev.* 57:138-163.
5. Vakulenko SB, Mobashery S (2003). Versatility of aminoglycosides and prospects for their future. *Clin. Microbiol. Rev.* 16:430-450.
6. Yamane K, Wachino J, Doi Y, Kurokawa H, Arakawa Y (2005). Global spread of multiple aminoglycoside resistance genes. *Emerg. Infect. Dis.* 11:951-953.