

Article

***In Vitro* Activities and Inoculum Effects of eftazidime-Avibactam and Aztreonam-Avibactam against Carbapenem-Resistant *Enterobacterales* Isolates from South Korea**

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Supplemental Table S1. Antimicrobial susceptibility of carbapenem-resistant *E. coli* and *K. pneumoniae* isolates to seven antimicrobial agents (n=81).

Antimicrobial agent	Inoculum size	No. of isolates (cumulative %) with indicated MICs (µg/mL)														MIC (µg/mL)		S n (%) ^a
		0.06	0.125	0.25	0.5	1	2	4	8	16	32	64	128	256	≥512	MIC ₅₀	MIC ₉₀	
CAZ	Standard					1 (1.2)	1 (2.5)	1 (3.7)	1 (4.9)		2 (7.4)	1 (8.6)	10 (21.0)	17 (42.0)	47 (100)	≥512	≥512	3 (3.7)
	High					1 (1.2)			1 (2.4)			1 (3.6)	1 (4.8)	4 (9.9)	73 (100)	≥512	≥512	1 (1.2)
CAZ-AVI	Standard				2 (2.5)	13 (18.5)	25 (49.4)	13 (65.4)	6 (72.8)	7 (81.5)	1 (82.7)				14 (100)	4	≥512	59 (72.8)
	High					6 (7.4)	11 (21.0)	17 (42.0)	13 (58.0)	17 (66.7)	2 (69.1)	1 (70.4)	5 (76.5)	4 (81.5)	15 (100)	8	≥512	47 (58.0)
ATM	Standard					4 (4.9)	1 (6.2)		1 (7.4)			3 (11.1)	2 (13.6)	8 (23.5)	62 (100)	≥512	≥512	5 (6.2)
	High					2 (2.5)	1 (3.7)			1 (4.9)		1 (6.2)	1 (7.4)	3 (11.1)	72 (100)	≥512	≥512	3 (3.7)
ATM-AVI	Standard	3 (3.7)	1 (4.9)	27 (38.3)	20 (63.0)	15 (81.5)	3 (85.2)	6 (92.6)	2 (95.1)	2 (97.5)	1 (98.8)		1 (100)			0.5	4	NA ^b
	High		2 (2.5)	14 (19.8)	10 (31.1)	2 (34.6)	10 (46.9)	4 (51.9)	3 (55.6)	3 (59.3)	19 (82.7)	2 (85.2)	1 (86.4)	8 (96.3)	3 (100)	4	256	NA ^b
MEM	Standard			3 (3.7)	3 (7.4)	2 (9.9)	4 (14.8)	4 (19.8)	12 (34.6)	15 (53.1)	11 (66.7)	12 (81.5)	4 (86.4)	11 ^c (100)		16	≥256	8 (9.9)
	High				2 (2.5)	1 (3.7)	5 (9.9)	2 (12.3)	10 (24.7)	13 (40.7)	9 (51.9)	9 (63.0)	10 (75.3)	20 ^c (100)		32	≥256	3 (3.7)
CST	Standard			19 (23.5)	51 (86.4)			2 (88.9)	2 (91.4)	3 (95.1)			3 (98.8)	1 ^c (100)		0.5	8	70 (86.4)
	High																	
TGC	Standard		2 (2.5)	9 (13.6)	9 (24.7)	18 (46.9)	14 (64.2)	17 (85.2)	7 (93.8)	2 (96.3)	1 (97.5)	1 (98.8)		1 (100)		2	8	20 (24.7)

MIC, minimum inhibitory concentration; CAZ, ceftazidime; CAZ-AVI, ceftazidime-avibactam; ATM, aztreonam; ATM-AVI, aztreonam-avibactam; MEM, meropenem; CST, colistin; TGC, tigecycline; NA, not available.^a CLSI susceptibility breakpoints were used: ceftazidime, ≤4 µg/mL; ceftazidime-avibactam, ≤8/4 µg/mL; aztreonam, ≤4 µg/mL; meropenem, ≤1 µg/mL; 2019 EUCAST susceptibility breakpoints were used for colistin and tigecycline: colistin, ≤2 µg/mL; tigecycline, ≤0.5 µg/mL.^b Not available because no breakpoint criteria have been defined for aztreonam-avibactam.^c MIC is greater than or equal to the indicated value.

Supplemental Table S2. Ceftazidime-avibactam and aztreonam-avibactam MIC distribution according to meropenem, colistin and tigecycline susceptibility pattern in carbapenem-resistant isolates.

	Colistin susceptibility ^b		P value	Tigecycline susceptibility ^c		P value
	Susceptible (n = 70)	Resistant (n = 11)		Susceptible (n = 20)	Resistant (n = 61)	
Ceftazidime-avibactam MIC ^a						
S (≤ 8 µg/mL)	50 (71.4)	9 (81.8)	.47	14 (70.0)	45 (73.8)	.74
R (> 8 µg/mL)	20 (28.6)	2 (18.2)		6 (30.0)	16 (26.2)	
Aztreonam-avibactam MIC						
Lower MIC (≤ 8 µg/mL)	67 (95.7)	10 (90.9)	.50	19 (95.0)	58 (95.1)	.99
Higher MIC (> 8 µg/mL)	3 (4.3)	1 (9.1)		1 (5.0)	3 (4.9)	

Data are presented as the number of isolates with the corresponding percentage shown in parentheses. ^a Ceftazidime-avibactam CLSI breakpoints: S $\leq 8/4$ µg/mL, R $> 8/4$ µg/mL. ^b Colistin EUCAST breakpoints: S ≤ 2 µg/mL, R > 2 µg/mL. ^c Tigecycline EUCAST breakpoints: S ≤ 0.5 µg/mL, R > 0.5 µg/mL.

Supplemental Table S3. MIC distributions of ceftazidime-avibactam and aztreonam-avibactam for carbapenem-resistant *E. coli* and *K. pneumoniae* (n=81).

AZT/AVI, MIC (µg/mL)	CAZ-AVI MIC (µg/mL) No. of isolates (no. of CPE) with indicated MIC											Grand total (No. of CPE)
	0.5	1	2	4	8	16	32	64	128	256	>256	
0.06		1 (1)									2 (1)	3 (2)
0.125	1 (0)											1 (0)
0.25	1 (0)	8 (3)	9 (6)		1 (0)						8 (8)	27 (17)
0.5		3 (0)	9 (4)	6 (3)	1 (0)						1 (1)	20 (8)
1		1 (0)	4 (0)	5 (4)	1 (1)	2 (0)					2 (2)	15 (7)
2			1 (0)	1 (0)							1 (1)	3 (1)
4			1 (0)	1 (0)	3 (0)	1 (0)						6 (0)
8			1 (0)			1 (0)						2 (0)
16						1 (0)	1 (0)					2 (0)
32						1 (0)						1 (0)
64												—
128						1 (0)						1 (0)
Grand total (No. of CPE)	2 (0)	13 (4)	25 (10)	13 (7)	6 (1)	7 (0)	1 (0)	—	—	—	14 (13)	81

Supplemental Table S4. Resistance mechanisms of carbapenem-resistant *E. coli* (n=25) and *K. pneumoniae* (n=56).

Mechanism	<i>E. coli</i>, n (%)	<i>K. pneumoniae</i>, n (%)	Total, n (%)
Non-CP-CRE	18 (72.0)	28 (50.0)	46 (100)
ESBL	13 (52.0)	17 (30.4)	30 (65.2)
AmpC	1 (4.0)	1 (1.8)	2 (4.3)
ESBL+AmpC	0	7 (12.5)	7 (15.2)
Others	4 (16.0)	3 (5.4)	7 (15.2)
CP-CRE	7 (28.0)	28 (50.0)	35 (100)
KPC	2 (8.0)	15 (26.8)	17 (48.6)
NDM	4 (16.0)	7 (12.5)	11 (31.4)
OXA-48-like	1 (4.0)	3 (5.4)	4 (11.4)
VIM	0	1 (1.8)	1 (2.9)
Others	0	2 (3.6)	2 (5.7)

Data are presented as the number of isolates with the corresponding percentage shown in parentheses. Non-CP-CRE, non-carbapenemase-producing carbapenem-resistant Enterobacteriaceae. .

Supplemental Table S5. Antimicrobial susceptibility and positive rate of inoculum effect of carbapenem-resistant isolates according to resistance mechanism.

Species (n)	Mechanism (n)	Antimicrobial agent	Inoculum size	MIC (µg/mL)			S _n (%) ^a	No. of isolates (%) with inoculum effect
				MIC ₅₀	MIC ₉₀	Range		
<i>E. coli</i> (25)	Non-CP CRE (18)	CAZ-AVI	Standard	2	16	1 to ≥512	14 (77.8)	1/17 (5.9)
			High	4	≥512	1 to ≥512	13 (72.2)	
		ATM-AVI	Standard	0.5	32	0.06 to 128	NA	1/18 (5.6)
			High	0.5	256	0.125 to ≥512	NA	
	ESBL (13)	CAZ-AVI	Standard	2	16	1 to ≥512	11 (84.6)	1/12 (8.3)
			High	8	≥512	1 to ≥512	10 (76.9)	
		ATM-AVI	Standard	0.5	8	0.06 to 8	NA	1/13 (7.7)
			High	0.5	8	0.125 to 8	NA	
	AmpC (1)	CAZ-AVI	Standard	-	-	1	1 (100)	0
			High	-	-	1	1 (100)	
		ATM-AVI	Standard	-	-	0.5	NA	0
			High	-	-	2	NA	
	CPE (7)	CAZ-AVI	Standard	≥512	≥512	1 to ≥512	3 (42.9)	1/3 (33.3)
			High	≥512	≥512	1 to ≥512		
		ATM-AVI	Standard	0.25	2	0.06 to 2	NA	1/7 (14.3)
			High	1	32	0.125 to 2	NA	
	KPC (2)	CAZ-AVI	Standard	-	-	1 to 4	2 (100)	1/2 (50.0)
			High	-	-	4 to 8	2 (100)	
		ATM-AVI	Standard	-	-	0.5 to 1	NA	1/2 (50.0)
			High	-	-	0.06 to 32	NA	
	NDM (4)	CAZ-AVI	Standard	≥512	≥512	≥512	0	ND
			High	≥512	≥512	≥512	0	
		ATM-AVI	Standard	0.25	2	0.06 to 2	NA	0
			High	0.25	2	0.124 to 2	NA	
<i>K. pneumoniae</i> (56)	Non-CP CRE (28)	CAZ-AVI	Standard	2	16	0.5 to 32	23 (82.1)	9/28 (32.1)
			High	8	256	1 to 256	14 (50.0)	
		ATM-AVI	Standard	0.5	4	0.125 to 16	NA	22/28 (78.6)
			High	32	256	0.25 to ≥512	NA	
	ESBL (17)	CAZ-AVI	Standard	4	16	0.5 to 32	13 (76.5)	6/17 (35.3)
			High	16	128	1 to 256	6 (35.3)	
		ATM-AVI	Standard	0.5	16	0.125 to 16	NA	16/17 (94.1)
			High	32	256	2 to ≥512	NA	
	AmpC (1)	CAZ-AVI	Standard	-	-	1	1 (100)	1/1 (100)
			High	-	-	256	0	
		ATM-AVI	Standard	-	-	1	NA	1/1 (100)
			High	-	-	32	NA	
	ESBL +AmpC (7)	CAZ-AVI	Standard	2	4	1 to 4	7 (100)	1/7 (14.3)
			High	4	128	2 to 128	6 (85.7)	
		ATM-AVI	Standard	1	2	0.25 to 2	NA	4/7 (57.1)
			High	32	32	2 to 32	NA	
	CPE (28)	CAZ-AVI	Standard	4	≥512	1 to ≥512	19 (67.9)	1/19 (5.3)
			High	8	≥512	2 to ≥512	17 (60.7)	
		ATM-AVI	Standard	0.25	1	0.25 to 1	NA	14/28 (50.0)
			High	2	64	0.25 to 256	NA	
	KPC (15)	CAZ-AVI	Standard	2	≥512	2 to ≥512	12 (80.0)	1/12 (8.3)
			High	8	≥512	8 to ≥512	11 (73.3)	
		ATM-AVI	Standard	0.25	1	0.25 to 1	NA	5/15 (33.3)
			High	0.5	32	0.25 to 256	NA	
	NDM (7)	CAZ-AVI	Standard	≥512	≥512	2 to ≥512	3 (42.9)	0/3
			High	≥512	≥512	2 to ≥512	2 (28.6)	
		ATM-AVI	Standard	0.25	1	0.25 to 1	NA	3/7 (42.9)
			High	0.5	64	0.25 to 64	NA	

Non-CP-CRE, non-carbapenemase-producing carbapenem-resistant Enterobacteriaceae; CAZ-AVI, ceftazidime-avibactam; ATM-AVI, aztreonam-avibactam, NA, not available; ND, not determined.^a CLSI susceptibility breakpoints were used: ceftazidime, ≤4 µg/mL; ceftazidime-avibactam,

$\leq 8/4$ $\mu\text{g/mL}$; aztreonam, ≤ 4 $\mu\text{g/mL}$; meropenem, ≤ 1 $\mu\text{g/mL}$; 2019 EUCAST susceptibility breakpoints were used for colistin and tigecycline: colistin, ≤ 2 $\mu\text{g/mL}$; tigecycline, ≤ 0.5 $\mu\text{g/mL}$.