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In Vitro Activity of BOS-228 against Carbapenem-resistant *Enterobacteriaceae*

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INTRODUCTION

BOS-228 (formerly LYS228, Novartis) is a potent next generation monobactam antibiotic active against carbapenem resistant *Enterobacteriaceae* (CRE), a CDC Urgent Threat and WHO Critical Threat pathogen, when resistance is caused by the production of serine beta-lactamases (SBLs) and/or metallo beta-lactamases (MBLs). Monobactams are important antibiotics due to their intrinsic stability to MBLs. Existing first generation monobactams such as aztreonam have limited clinical utility against MBL-producing CRE because they are susceptible to serine BLs that are often co-expressed in clinical isolates [1]. BOS-228 was identified in a medicinal chemistry program undertaken to generate monobactams with enhanced stability to SBLs. Thus, BOS-228 is differentiated from not just monobactams, but all currently approved beta-lactams in that it represents a potential single agent therapy effective against CRE that express both SBLs and MBLs. BOS-228 has completed Phase 1 clinical trials [2]. BOS-228 was licensed to Boston Pharmaceuticals in late-2018 and will be advanced to late development clinical trials.

MATERIALS & METHODS

The MIC values of BOS-228 and comparators were determined by broth microdilution following CLSI M07-A11 guidelines [3] for 973 CRE isolates, including 850 isolates with a gene encoding a carbapenemase and 123 CRE isolates where these genes were not detected. All study organisms were clinical isolates collected in 2015 and were from community- and hospital-associated sources, distributed globally (Figure 1). Susceptibility was defined using CLSI 2019 interpretive criteria, excluding colistin for which EUCAST 2019 breakpoints were used, and tigecycline, for which FDA breakpoints were used [4-5], and EUCAST 2019 interpretive criteria [5]. Ceftazidime-avibactam was tested at a fixed concentration of 4 µg/mL avibactam. Molecular characterization of β-lactamases for genes encoding MBLs (IMP, VIM, NDM), KPC and other β-lactamases (OXA-48-like) was performed via multiplex PCR, followed by sequencing.

RESULTS SUMMARY

- BOS-228 showed potent *in vitro* activity against CRE, with $MIC_{50/90}$ values of 0.5/1 µg/mL for all CRE isolates (Table 2, Table 3).
- The MIC_{90} value was 1 µg/mL for isolates producing MBL or serine (KPC) carbapenemases, and 2 µg/mL for isolates producing OXA-48 like enzymes (Table 1).
- Based on MIC_{90} values, BOS-228 was the most potent agent tested against CRE, including MBL-positive isolates and those carrying multiple carbapenemases, regardless of carbapenemase type (Table 2, Figure 3).
- Unlike ceftazidime-avibactam, BOS-228 exhibited *in vitro* activity against MBL-producers, including NDM, IMP and VIM, inhibiting 97.5% of variants at an MIC of ≤2 µg/mL (Table 2, Figure 3).

CONCLUSIONS

- BOS-228 demonstrated potent *in vitro* activity against CRE, including KPC-, MBL-, and OXA-producing isolates.
- BOS-228 also retained activity against CRE with resistance mediated by non-carbapenemase mechanisms, presumably including permeability defects.
- BOS-228, as a novel single agent monobactam, has potential to treat *Enterobacteriaceae* infections including those caused by MBL- and SBL-expressing CRE.

REFERENCES

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ACKNOWLEDGMENTS

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RESULTS

Figure 1A. Species distribution of 850 carbapenem-resistant *Enterobacteriaceae* (CRE) producing serine carbapenemases (KPC, OXA-48-like) and metallo-β-lactamases (MBLs; NDM, IMP, VIM)

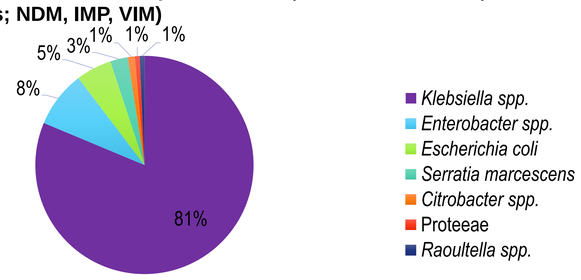


Figure 1B. Species distribution of 123 carbapenem-resistant *Enterobacteriaceae* (CRE) with no carbapenemase detected

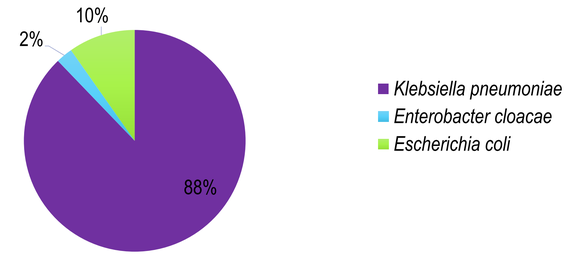
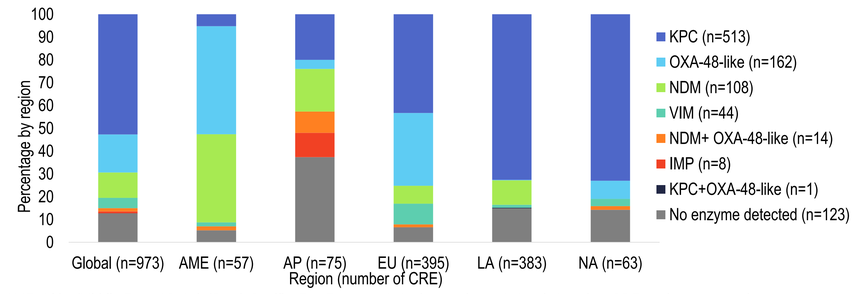


Table 1. Distribution of resistance mechanisms among carbapenem-resistant *Enterobacteriaceae* (CRE) by species

Organism	Carbapenemase detected								
	KPC	KPC+OXA	NDM	NDM+OXA	IMP	OXA	VIM	none	Total
Citrobacter freundii	6						1		7
Citrobacter koseri	2								2
Enterobacter aerogenes	6		1						7
Enterobacter asburiae	1						1		2
Enterobacter cloacae	16		22		3	2	18	3	64
Enterobacter kobei					1				1
Escherichia coli	17		7			19	2	12	57
Klebsiella oxytoca	11				1	6	1		19
Klebsiella pneumoniae	441	1	71	13	3	128	15	108	780
Proteus mirabilis			2						2
Providencia stuartii			3				1		4
Raoultella ornithinolytica						3			3
Raoultella planticola						3			3
Serratia marcescens	13		2	1		1	5		22
Total	**513**	**1**	**108**	**14**	**8**	**162**	**44**	**123**	**973**

Figure 2. Distribution of resistance mechanisms among carbapenem-resistant *Enterobacteriaceae* (CRE) by region



AME, Africa/Middle East; AP, Asia/South Pacific; EU, Europe; LA, Latin America; NA, North America; CRE, carbapenem-resistant *Enterobacteriaceae*; +, isolates contain both enzymes

Figure 3. BOS-228 MIC distribution against carbapenem-resistant *Enterobacteriaceae* (CRE) with different resistance mechanisms

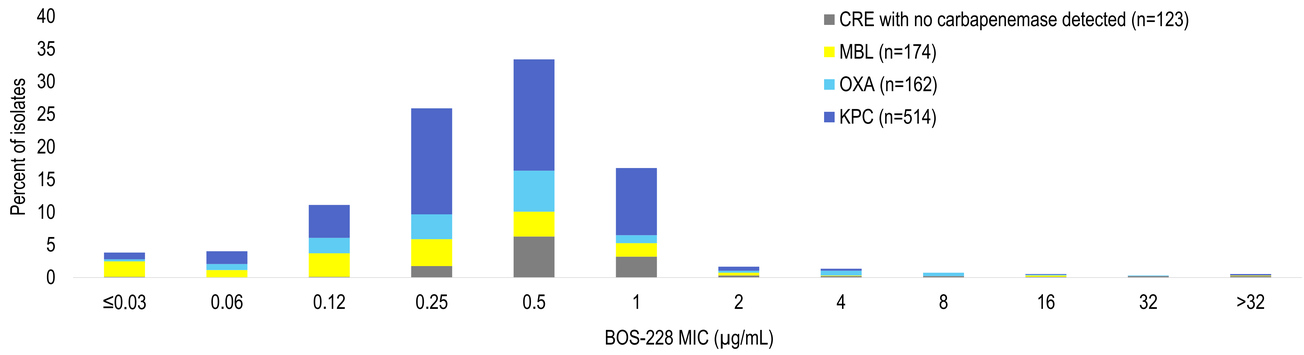


Table 2. *In vitro* activity of BOS-228 and comparator antimicrobials against 850 carbapenemase-producing CRE

Organism, Genotype (n)	Antimicrobial	%S (CLSI)	%S (EUCAST)	MIC_{50}	MIC_{90}	Range
Carbapenemase-producing CRE[a] (850)	BOS-228	na	na	0.5	1	≤0.03 - 64
	Amikacin	66.8	51.4	8	>64	0.25 - >64
	Ceftazidime-avibactam	79.5	79.5	1	>64	≤0.03 - >64
	Colistin	79.9	79.9	0.25	16	0.12 - >32
	Ertapenem	0.6	0.6	32	>64	0.5 - >64
	Meropenem	6.6	13.7	32	>64	0.5 - >64
	Tigecycline	(93.9)[j]	41.3	1	2	0.06 - 16
CRE, IMP[b] (8)	BOS-228	na	na	nc	nc	≤0.03 - 1
	Amikacin	87.5	75.0	nc	nc	1 - >64
	Ceftazidime-avibactam	0	0	nc	nc	64 - >64
	Colistin	100	100	nc	nc	0.12 - 0.5
	Ertapenem	0	0	nc	nc	1 - 8
	Meropenem	37.5	50.0	nc	nc	1 - 16
	Tigecycline	(87.5)[j]	50.0	nc	nc	0.12 - 4
CRE, KPC[c] (513)	BOS-228	na	na	0.5	1	≤0.03 - 64
	Amikacin	66.7	48.7	16	32	0.25 - >64
	Ceftazidime-avibactam	99.6	99.6	1	2	≤0.03 - 16
	Colistin	79.3	79.3	0.25	16	0.12 - >32
	Ertapenem	0	0	64	>64	1 - >64
	Meropenem	2.3	8.0	32	>64	0.5 - >64
	Tigecycline	(94.5)[j]	43.1	1	2	0.06 - 8
CRE, KPC+OXA[d] (1)	BOS-228	nc	nc	nc	nc	0.5 - 0.5
	Amikacin	nc	nc	nc	nc	>64 - >64
	Ceftazidime-avibactam	nc	nc	nc	nc	1 - 1
	Colistin	nc	nc	nc	nc	32 - 32
	Ertapenem	nc	nc	nc	nc	>64 - >64
	Meropenem	nc	nc	nc	nc	64 - 64
	Tigecycline	nc	nc	nc	nc	1 - 1
CRE, NDM[e] (108)	BOS-228	na	na	0.25	1	≤0.03 - 16
	Amikacin	49.1	39.8	32	>64	0.5 - >64
	Ceftazidime-avibactam	0	0	>64	>64	>64
	Colistin	86.1	86.1	0.25	16	0.12 - >32
	Ertapenem	0	0	64	>64	2 - >64
	Meropenem	0.9	1.9	64	>64	1 - >64
	Tigecycline	(93.5)[j]	37.0	1	2	0.06 - 4
CRE, NDM+OXA[f] (14)	BOS-228	na	na	0.5	1	0.25 - 2
	Amikacin	42.9	14.3	>64	>64	8 - >64
	Ceftazidime-avibactam	0	0	>64	>64	>64
	Colistin	71.4	71.4	0.5	8	0.12 - >32
	Ertapenem	0	0	>64	>64	>64 - >64
	Meropenem	0	0	>64	>64	>64 - >64
	Tigecycline	(100)[j]	28.6	1	2	0.12 - 2
CRE, OXA[g] (162)	BOS-228	na	na	0.5	2	≤0.03 - 32
	Amikacin	73.5	65.4	4	>64	0.5 - >64
	Ceftazidime-avibactam	100	100	1	2	0.12 - 8
	Colistin	76.5	76.5	0.25	16	0.12 - >32
	Ertapenem	0	0	16	>64	1 - >64
	Meropenem	21	35.2	8	64	0.5 - >64
	Tigecycline	(92.6)[j]	35.2	1	2	0.06 - 8
CRE, VIM[h] (44)	BOS-228	na	na	0.25	1	≤0.03 - 64
	Amikacin	93.2	68.2	8	16	0.5 - >64
	Ceftazidime-avibactam	4.6	4.6	>64	>64	4 - >64
	Colistin	84.1	84.1	0.25	>32	0.12 - >32
	Ertapenem	11.4	11.4	4	32	0.5 - >64
	Meropenem	13.6	27.3	8	64	1 - >64
	Tigecycline	(90.9)[j]	56.8	0.5	2	0.06 - 16

CRE, carbapenem-resistant *Enterobacteriaceae*; $MIC_{50/90}$ and range in µg/mL, %S, percent susceptible
[a] Isolates for which a gene encoding a carbapenemase was detected by PCR (KPC, IMP, NDM, VIM or OXA). Organisms include: *C. freundii* (7), *C. koseri* (2), *E. aerogenes* (7), *E. asburiae* (2), *E. cloacae* (61), *E. kobei* (1), *E. coli* (45), *K. oxytoca* (19), *K. pneumoniae* (672), *P. mirabilis* (2), *P. stuartii* (4), *R. ornithinolytica* (2), *R. planticola* (3), *S. marcescens* (22)
[b] Organisms include: *E. cloacae* (3), *E. kobei* (1), *K. oxytoca* (1), *K. pneumoniae* (3)
[c] Organisms include: *C. freundii* (6), *C. koseri* (2), *E. aerogenes* (6), *E. asburiae* (1), *E. cloacae* (16), *E. coli* (17), *K. oxytoca* (11), *K. pneumoniae* (441), *S. marcescens* (13)
[d] Organisms include: *K. pneumoniae* (1)
[e] Organisms include: *E. aerogenes* (1), *E. cloacae* (22), *E. coli* (7), *K. pneumoniae* (71), *P. mirabilis* (2), *P. stuartii* (3), *S. marcescens* (2)
[f] Organisms include: *K. pneumoniae* (13), *S. marcescens* (1)
[g] Organisms include: *E. cloacae* (2), *E. coli* (19), *K. oxytoca* (6), *K. pneumoniae* (128), *R. ornithinolytica* (3), *R. planticola* (3), *S. marcescens* (1)
[h] Organisms include: *C. freundii* (1), *E. asburiae* (1), *E. cloacae* (18), *E. coli* (2), *K. oxytoca* (1), *K. pneumoniae* (15), *P. stuartii* (1), *S. marcescens* (5)
[i] FDA interpretive criteria applied

Table 3. *In Vitro* Activity of BOS-228 and Comparator Antimicrobials Against 123 Non-carbapenemase-producing CRE

Organism, Genotype	Antimicrobial	%S (CLSI)	%S (EUCAST)	MIC_{50}	MIC_{90}	Range
Non-carbapenemase-producing CRE[a] (123)	BOS-228	na[b]	na	0.5	1	≤0.03 - >64
	Amikacin	74	67.5	4	>64	0.5 - >64
	Ceftazidime-avibactam	100	100	2	4	0.12 - 8
	Colistin	91.1	95.1	0.5	2	0.12 - >32
	Ertapenem	0	0	8	32	1 - >64
	Meropenem	30.1	66.7	2	8	0.06 - 32
	Tigecycline	(92.7)[c]	31.7	1	2	0.12 - 8

CRE, carbapenem-resistant *Enterobacteriaceae*; $MIC_{50/90}$ and range in µg/mL, %S, percent susceptible
[a] Isolates for which genes encoding carbapenemases were not detected by PCR (KPC, IMP, NDM, VIM or OXA). Organisms include: *E. cloacae* (2), *E. coli* (12), *K. pneumoniae* (108)
[b] not available
[c] FDA interpretive criteria applied