

In Vitro Activity of Sulbactam-Durlobactam in Combination with Other Antimicrobial Agents

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Abstract

Background: Sulbactam-durlobactam (SUL-DUR) is a β-lactam/β-lactamase inhibitor combination being developed for treatment of infections due to *Acinetobacter baumannii-calcoaceticus* complex (ABC), including multidrug-resistant and carbapenem-resistant strains. Durlobactam (ETX2514) is a diazabicyclooctane β-lactamase inhibitor with potent activity against Ambler classes A, C and D serine β-lactamases. Sulbactam is a β-lactam with antibacterial activity limited to only a few species, including ABC. Here we report *in vitro* studies using checkerboard assays that were conducted to test for interactions between SUL-DUR and other antibacterial and antifungal agents.

Methods: Susceptibility testing was performed using broth microdilution according to CLSI guidelines for SUL-DUR and twelve other antimicrobial agents (eleven antibacterial and one antifungal) against ten *A. baumannii* clinical isolates and CLSI quality control (QC) strains. Subsequently, combinations of SUL-DUR and 12 antimicrobial agents were tested in checkerboard assays. By comparing the MIC of each agent alone to the MIC in combination, a series of fractional inhibitory concentrations (FIC) were determined to generate a mean FIC, which represents any drug interactions. Synergy was defined as an FIC of ≤0.5, additivity/indifference was defined as an FIC of >0.5-4 and an FIC of >4 was used to define antagonism.

Results: The mean FIC values for combinations with SUL-DUR ranged between 0.2 and 2.5. No antagonism was observed between SUL-DUR and any of the 12 antimicrobial agents tested against any isolates. There were a few instances of synergy, but these were strain- and drug-dependent. Additivity or indifference was the prevailing type of interaction observed between SUL-DUR and the other antimicrobial agents tested.

Conclusions: *In vitro* checkerboard assays with ten clinical isolates of *A. baumannii* found additivity or indifference when SUL-DUR was tested in combination with representatives of the major classes of antimicrobial agents. No instances of antagonism between SUL-DUR and 12 other antimicrobial agents were found. These results suggest that, if approved, SUL-DUR could be co-dosed with other antimicrobial agents with no expected interactions.

Introduction

The Gram-negative organisms collectively named the *Acinetobacter baumannii-calcoaceticus* complex (ABC) have emerged as serious pathogens¹. The ABC complex includes *A. baumannii*, *A. nosocomialis*, *A. pittii* and *A. calcoaceticus*. *A. baumannii* is considered the most clinically important species of the complex due to its association with nosocomial outbreaks. Globally, the susceptibility of ABC to all antimicrobial agents has declined over the last 20 years².

Sulbactam-durlobactam (SUL-DUR) recently completed a Phase 3 clinical trial for the treatment of infections caused by carbapenem-resistant ABC organisms. Sulbactam (SUL) is an approved β-lactamase inhibitor (BLI) with antibacterial activity against *Acinetobacter* spp. due to its inhibition of PBP3, an enzyme required for cell wall biosynthesis³. However, degradation of SUL by the β-lactamases present in most contemporary ABC isolates limits its clinical use. Durlobactam (DUR) is a diazabicyclooctane BLI with potent activity against class A, C and D serine β-lactamases⁴. DUR protects SUL from degradation, restoring antibacterial activity against ABC organisms.

The goal of these studies was to determine if there was antagonism, synergy or indifference when SUL-DUR was combined with other antimicrobial agents against *A. baumannii* or representative strains of other microbial species.

Strains used in this study

Strain number	Species	Encoded β-lactamases
ATCC 19606	<i>A. baumannii</i>	ADC-2-like; OXA-98
NCTC 13304 (CLSI reference strain)	<i>A. baumannii</i>	ADC-30; TEM-1; OXA-23; OXA-66
ARC2058	<i>A. baumannii</i>	ADC-3-like; OXA-259
ARC3486	<i>A. baumannii</i>	ADC-30; TEM-1; OXA-66; OXA-72
ARC3488	<i>A. baumannii</i>	ADC-76; OXA-68; OXA-235-like
ARC3492	<i>A. baumannii</i>	ADC-52-like; TEM-1; OXA-24; OXA-132
ARC3495	<i>A. baumannii</i>	ADC-30-like; OXA-24; OXA-109
ARC5079	<i>A. baumannii</i>	ADC-52-like; OXA-65; OXA-72
ARC5081	<i>A. baumannii</i>	ADC-80; ADC-81-like; OXA-23; OXA-94
ARC5092	<i>A. baumannii</i>	ADC-5-like; OXA-23; OXA-64
ATCC 27853	<i>P. aeruginosa</i>	AmpC; PoxB
USA100	<i>S. aureus</i>	
ATCC 90028 (CLSI reference strain)	<i>C. albicans</i>	
ARC4416	<i>E. coli</i>	AmpC; CTX-M-14; TEM-1
ARC4418	<i>E. coli</i>	AmpC; CTX-M-14; CMY-2
ARC4421	<i>E. coli</i>	AmpC; CTX-M-55; TEM-1; CMY-2
ARC4426	<i>E. coli</i>	AmpC; TEM-1; CMY-2
ATCC 700603 (CLSI reference strain)	<i>K. pneumoniae</i>	OKP-6; OXA-2; SHV-18
ARC4420	<i>K. pneumoniae</i>	SHV-11, SHV-12, DHA-1, OXA-1
ARC4490	<i>K. pneumoniae</i>	SHV-11, CTX-M-15, KPC-2, OXA-1
ARC4461	<i>E. cloacae</i>	AmpC; TEM-1
ARC4473	<i>E. cloacae</i>	SHV-5; AmpC; DHA-1; TEM-1
ARC3522	<i>C. freundii</i>	AmpC, TEM-1, CMY-65
ARC3883	<i>C. freundii</i>	CMY-48; KPC-2
ATCC 25922 (CLSI reference strain)	<i>E. coli</i>	AmpC

SUL-DUR (1:1)* versus Imipenem

Strain	Species	SUL-DUR MIC (mg/L)	Imipenem MIC (mg/L)	Mean FIC	Interpretation
ATCC25922	<i>E. coli</i>	0.5	0.125	1.27	indifference
ARC4416	<i>E. coli</i>	0.25	0.125	0.84	indifference
ARC4418	<i>E. coli</i>	0.25	0.125	0.90	indifference
ARC4421	<i>E. coli</i>	0.25	0.125	1.45	indifference
ARC4426	<i>E. coli</i>	0.125	0.125	1.04	indifference
ATCC700603	<i>K. pneumoniae</i>	2	0.125	2.78	indifference
ARC4420	<i>K. pneumoniae</i>	2	0.5	2.33	indifference
ARC4490	<i>K. pneumoniae</i>	1	0.5	1.37	indifference
ARC4461	<i>E. cloacae</i>	0.125	0.125	1.63	indifference
ARC4473	<i>E. cloacae</i>	0.25	0.25	1.60	indifference
ARC3522	<i>C. freundii</i>	0.25	0.25	0.96	indifference
ARC3883	<i>C. freundii</i>	0.5	1	0.75	indifference

*SUL-DUR was tested in a 1:1 fixed ratio because durlobactam has intrinsic activity against some *Enterobacteriales* species.

Abbreviations: *A. baumannii*, *Acinetobacter baumannii*; *P. aeruginosa*, *Pseudomonas aeruginosa*; *S. aureus*, *Staphylococcus aureus*; *C. albicans*, *Candida albicans*; *E. coli*, *Escherichia coli*; *K. pneumoniae*, *Klebsiella pneumoniae*; *E. cloacae*, *Enterobacter cloacae*; *C. freundii*, *Citrobacter freundii*.

Colistin

Strain	Species	SUL-DUR MIC (mg/L)	Colistin MIC (mg/L)	Mean FIC	Interpretation
ATCC19606	<i>A. baumannii</i>	0.5	0.25	1.5	indifference
ARC2058	<i>A. baumannii</i>	0.5	0.25	<0.53	indifference
ARC3486	<i>A. baumannii</i>	0.5	0.25	<0.53	indifference
ARC3488	<i>A. baumannii</i>	0.5	0.25	0.75	indifference
ARC3492	<i>A. baumannii</i>	0.25	0.25	1.5	indifference
ARC3495	<i>A. baumannii</i>	0.5	0.25	<0.53	indifference
ARC5079	<i>A. baumannii</i>	0.5	0.25	<0.53	indifference
ARC5081	<i>A. baumannii</i>	1	0.125	1.96	indifference
ARC5092	<i>A. baumannii</i>	0.5	>8	0.64	indifference
NCTC13304	<i>A. baumannii</i>	0.5	0.25	<0.53	indifference
ATCC27853	<i>P. aeruginosa</i>	>32	0.5	1.25	indifference

Cefepime

Strain	Species	SUL-DUR MIC (mg/L)	Cefepime MIC (mg/L)	Mean FIC	Interpretation
ATCC19606	<i>A. baumannii</i>	0.5	8	0.89	indifference
ARC2058	<i>A. baumannii</i>	1	2	0.66	indifference
ARC3486	<i>A. baumannii</i>	1	8	0.48	synergy
ARC3488	<i>A. baumannii</i>	0.5	>32	0.84	indifference
ARC3492	<i>A. baumannii</i>	0.25	8	<0.13	synergy
ARC3495	<i>A. baumannii</i>	0.5	32	0.27	synergy
ARC5079	<i>A. baumannii</i>	1.0	>32	0.69	indifference
ARC5081	<i>A. baumannii</i>	2	16	0.82	indifference
ARC5092	<i>A. baumannii</i>	1	16	0.23	synergy
NCTC13304	<i>A. baumannii</i>	0.5	16	1.03	indifference
ATCC27853	<i>P. aeruginosa</i>	>32	1	1.15	indifference

Ceftazidime-avibactam

Strain	Species	SUL-DUR MIC (mg/L)	Ceftazidime-avibactam MIC (mg/L)	Mean FIC	Interpretation
ATCC19606	<i>A. baumannii</i>	1	16	0.41	synergy
ARC2058	<i>A. baumannii</i>	0.5	4	1.25	indifference
ARC3486	<i>A. baumannii</i>	0.5	4	0.64	indifference
ARC3488	<i>A. baumannii</i>	0.5	4	0.88	indifference
ARC3492	<i>A. baumannii</i>	0.5	4	<0.28	synergy
ARC3495	<i>A. baumannii</i>	0.5	8	<0.28	synergy
ARC5079	<i>A. baumannii</i>	1	16	0.77	indifference
ARC5081	<i>A. baumannii</i>	2	64	1.02	indifference
ARC5092	<i>A. baumannii</i>	0.5	8	0.63	indifference
NCTC13304	<i>A. baumannii</i>	1	8	0.5	synergy
ATCC27853	<i>P. aeruginosa</i>	>32	1	1.23	indifference

* 4 mg/L avibactam

Imipenem

Strain	Species	SUL-DUR MIC (mg/L)	Imipenem MIC (mg/L)	Mean FIC	Interpretation
ATCC19606	<i>A. baumannii</i>	0.5	0.25	<0.53	indifference
ARC2058	<i>A. baumannii</i>	0.25	0.125	1.11	indifference
ARC3486	<i>A. baumannii</i>	1	>8	0.9	indifference
ARC3488	<i>A. baumannii</i>	0.5	2	<0.09	synergy
ARC3492	<i>A. baumannii</i>	0.5	>8	0.6	indifference
ARC3495	<i>A. baumannii</i>	0.5	>8	0.6	indifference
ARC5079	<i>A. baumannii</i>	1	>8	0.72	indifference
ARC5081	<i>A. baumannii</i>	2	8	0.87	indifference
ARC5092	<i>A. baumannii</i>	0.5	>8	0.51	indifference
NCTC13304	<i>A. baumannii</i>	0.5	>8	1.51	indifference
ATCC27853	<i>P. aeruginosa</i>	>32	1	<0.13	synergy

Ciprofloxacin

Strain	Species	SUL-DUR MIC (mg/L)	Ciprofloxacin MIC (mg/L)	Mean FIC	Interpretation
ATCC19606	<i>A. baumannii</i>	0.5	0.5	1.29	indifference
ARC2058	<i>A. baumannii</i>	1	0.125	1.5	indifference
ARC3486	<i>A. baumannii</i>	1	>4	1.07	indifference
ARC3488	<i>A. baumannii</i>	0.5	>4	1.14	indifference
ARC3492	<i>A. baumannii</i>	0.25	>4	1.28	indifference
ARC3495	<i>A. baumannii</i>	0.5	>4	1.14	indifference
ARC5079	<i>A. baumannii</i>	0.5	>4	1.57	indifference
ARC5081	<i>A. baumannii</i>	2	>4	1.14	indifference
ARC5092	<i>A. baumannii</i>	0.5	>4	1.14	indifference
NCTC13304	<i>A. baumannii</i>	1	>4	1.14	indifference
ATCC27853	<i>P. aeruginosa</i>	>32	0.25	1.25	indifference

Meropenem

Strain	Species	SUL-DUR MIC (mg/L)	Meropenem MIC (mg/L)	Mean FIC	Interpretation
ATCC19606	<i>A. baumannii</i>	0.5	1	0.56	indifference
ARC2058	<i>A. baumannii</i>	0.5	0.25	0.75	indifference
ARC3486	<i>A. baumannii</i>	0.5	>8	1.3	indifference
ARC3488	<i>A. baumannii</i>	0.5	2	0.47	synergy
ARC3492	<i>A. baumannii</i>	0.5	>8	<0.04	synergy
ARC3495	<i>A. baumannii</i>	0.25	>8	0.85	indifference
ARC5079	<i>A. baumannii</i>	0.5	>8	1.03	indifference
ARC5081	<i>A. baumannii</i>	2	>8	0.9	indifference
ARC5092	<i>A. baumannii</i>	0.5	>8	0.69	indifference
NCTC13304	<i>A. baumannii</i>	0.5	>8	0.56	indifference
ATCC27853	<i>P. aeruginosa</i>	>32	1	1.13	indifference

Amikacin

Strain	Species	SUL-DUR MIC (mg/L)	Amikacin MIC (mg/L)	Mean FIC	Interpretation
ATCC19606	<i>A. baumannii</i>	0.5	8	0.91	indifference
ARC2058	<i>A. baumannii</i>	0.5	1	0.65	indifference
ARC3486	<i>A. baumannii</i>	1	32	0.76	indifference
ARC3488	<i>A. baumannii</i>	0.5	>64	0.95	indifference
ARC3492	<i>A. baumannii</i>	0.5	64	0.5	synergy
ARC3495	<i>A. baumannii</i>	0.25	64	0.87	indifference
ARC5079	<i>A. baumannii</i>	0.5	>64	1.71	indifference
ARC5081	<i>A. baumannii</i>	2	32	0.94	indifference
ARC5092	<i>A. baumannii</i>	0.5	>64	0.37	synergy
NCTC13304	<i>A. baumannii</i>	1	0.5	0.38	synergy
ATCC27853	<i>P. aeruginosa</i>	>32	1	1.23	indifference

Linezolid

Strain	Species	SUL-DUR MIC (mg/L)	Linezolid MIC (mg/L)	Mean FIC	Interpretation
ATCC19606	<i>A. baumannii</i>	1	>16	1.14	indifference
ARC2058	<i>A. baumannii</i>	0.5	>16	1.14	indifference
ARC3486	<i>A. baumannii</i>	0.5	>16	1.07	indifference
ARC3488	<i>A. baumannii</i>	0.5	>16	1.14	indifference
ARC3492	<i>A. baumannii</i>	0.25	>16	2.14	indifference
ARC3495	<i>A. baumannii</i>	0.25	>16	1.14	indifference
ARC5079	<i>A. baumannii</i>	0.5	>16	2.00	indifference
ARC5081	<i>A. baumannii</i>	2	>16	1.14	indifference
ARC5092	<i>A. baumannii</i>	0.5	>16	1.14	indifference
NCTC13304	<i>A. baumannii</i>	0.5	>16	1.28	indifference
USA100	<i>S. aureus</i>	>32	2	1.19	indifference

Minocycline

Strain	Species	SUL-DUR MIC (mg/L)	Minocycline MIC (mg/L)	Mean FIC	Interpretation
ATCC19606	<i>A. baumannii</i>	0.5	0.13	1	indifference
ARC2058	<i>A. baumannii</i>	0.25	0.125	2.38	indifference
ARC3486	<i>A. baumannii</i>	1	2	1	indifference
ARC3488	<i>A. baumannii</i>	0.5	1	1.5	indifference
ARC3492	<i>A. baumannii</i>	0.5	2	<0.28	synergy
ARC3495	<i>A. baumannii</i>	0.25	0.5	1.11	indifference
ARC5079	<i>A. baumannii</i>	0.5	0.5	1.23	indifference
ARC5081	<i>A. baumannii</i>	2	0.25	0.88	indifference
ARC5092	<i>A. baumannii</i>	0.5	4	1	indifference
NCTC13304	<i>A. baumannii</i>	1	2	1.38	indifference