Activity of SPR206, a Polymyxin Derivative, Compared to Colistin Alone and in Combination Against Multidrug-Resistant *Pseudomonas aeruginosa* Strains

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Introduction

Background: The emergence of multidrug-resistant (MDR) P. aeruginosa, has forced clinicians to resort to polymyxin antibiotics (polymyxin B and colistin (COL), previously discarded due to harmful adverse effects associated with their use (1).

Motivation: Despite their resurgence in clinical treatment, the polymyxins are continually characterized by their side effect profile (2). SPR206 is a polymyxin analogue, however the prodrug side chain has been extensively modified, decreasing the potential for adverse events. SPR206 has been shown to have reduced minimum inhibitory concentrations (MIC; MIC_{50} and MIC_{90}), values for *P. aeruginosa* when compared to COL, and other Gram-negative agents. (3).

Objective: The objective of this study was to compare the *in-vitro* activity of SPR206 to COL both alone and in combination with other Gram-negative antimicrobials against MDR *P. aeruginosa* strains through MIC susceptibility testing and time-kill experiments (TKE) .

Significance: As *MDR P. aeruginosa* infections increase patient mortality and morbidity, it is important that we are equipped with both safe and efficacious novel therapeutic options .

Methods

Bacterial strains: Fifteen carbapenem (meropenem (MEM) MIC >8mg/l) P. aeruginosa strains were evaluated using MIC susceptibility testing and TKE.

Media/ Antibiotics: COL, MEM, Fosfomycin (FOS), Amikacin (AMK), piperacillin//tazobactam (PIP/TAZ), aztreonam (AZT), ceftazidime (CAZ) were purchased commercially from Sigma Chemical Co. (St. Louis, MO, USA), Avibactam (AVI) was purchased from Fisher scientific SPR206 was obtained from its manufacturer (Spero Therapeutics Cambridge, Massachusetts)

Susceptibility Testing: MIC values were determined by broth micro-dilution in duplicate, per the current Clinical Laboratory and Standards Institute (CLSI) Guidelines for all strains. MIC testing via broth microdilution was performed for SPR-206, COL, MEM, FOS, AMK, AZT, CAZ/AVI, and PIP/TAZ. Avi was supplemented at a 4:1 ratio to CAZ.

Time-Kill Experiments:. Dual therapy and triple therapy combinations, either COL or SPR206-based, were tested against four representative strains in 24h time-kill experiments (TKE). Each antibiotic was tested at 1x the MIC, or the peak concentration, whichever was lower. A >2 log10 CFU/ml was defined as synergistic activity, and a >3log10 CFU/ml was defined as bactericidal activity.

Results

Time-Kill Analysis

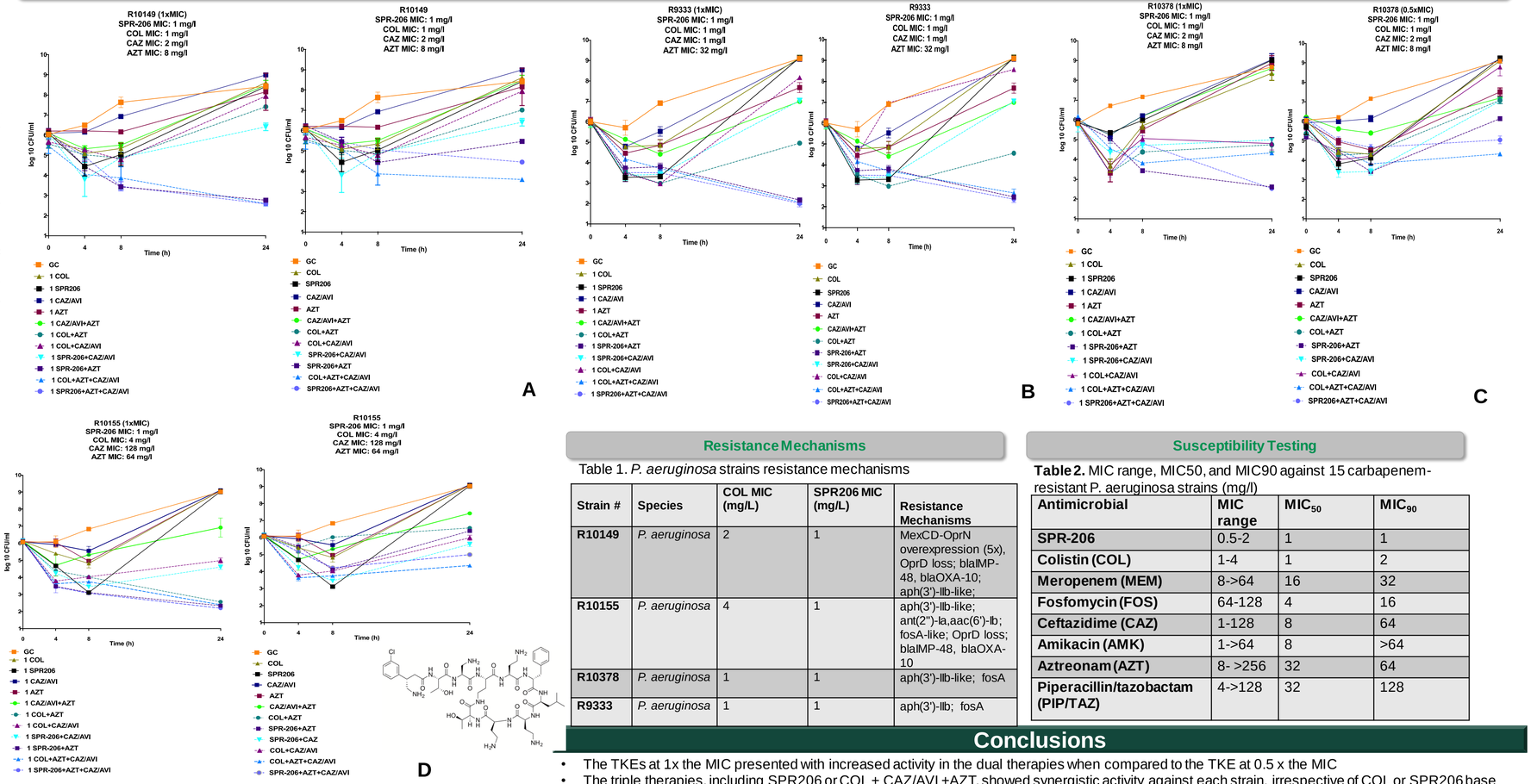

Figure 2 (A-D). Graphs at 1x and 0.5x the MIC of the Anti-Pseudomonal antimicrobials alone and in combination with SPR206 or COL against 4 strains

Resistance Mechanisms

Table 1. *P. aeruginosa* strains resistance mechanisms

Strain #	Species	COL MIC (mg/L)	SPR206 MIC (mg/L)	Resistance Mechanisms
R10149	*P. aeruginosa*	2	1	MexCD-OprN overexpression (5x), OprD loss; blaIMP-48, blaOXA-10; aph(3')-IIb-like;
R10155	*P. aeruginosa*	4	1	aph(3')-IIb-like; ant(2'')-Ia,aac(6')-Ib; fosA-like; OprD loss; blaIMP-48, blaOXA-10
R10378	*P. aeruginosa*	1	1	aph(3')-IIb-like; fosA
R9333	*P. aeruginosa*	1	1	aph(3')-IIb; fosA

Susceptibility Testing

Table 2. MIC range, MIC50, and MIC90 against 15 carbapenem-resistant P. aeruginosa strains (mg/l)

Antimicrobial	MIC range	MIC_{50}	MIC_{90}
SPR-206	0.5-2	1	1
Colistin (COL)	1-4	1	2
Meropenem (MEM)	8->64	16	32
Fosfomycin (FOS)	64-128	4	16
Ceftazidime (CAZ)	1-128	8	64
Amikacin (AMK)	1->64	8	>64
Aztreonam (AZT)	8- >256	32	64
Piperacillin/tazobactam (PIP/TAZ)	4->128	32	128

Conclusions

- The TKEs at 1x the MIC presented with increased activity in the dual therapies when compared to the TKE at 0.5 x the MIC
- The triple therapies, including SPR206 or COL + CAZ/AVI +AZT, showed synergistic activity against each strain, irrespective of COL or SPR206 base and concentration tested
- At 1x the MIC the SPR206 + AZT dual therapy presented with bactericidal activity against each strain
- Further research is warranted to solidify the role of SPR206 in the current antibiotic armamentarium

References

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Conclusions

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