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Contemporary In Vitro Synergy Rates for Aztreonam Combined with Newer Fluoroquinolones and ß-Lactams Tested Against Gram-Negative Bacilli

RN JONES, H HUYNH, PR RHOMBERG, TR FRITSCHE, HS SADER
The JONES Group/JMI Laboratories, North Liberty, IA

The JONES Group/JMI Laboratories
North Liberty, IA, USA
www.jmilabs.com
319.665.3370, fax 319.665.3371
ronald-jones@jmilabs.com

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AMENDED ABSTRACT

Background: Aztreonam (ATM) has been commonly used in various combinations to enhance spectrum and produce potential synergy (SYN). Although well studied in vitro over 10 years ago, ATM combination SYN testing has been poorly documented for newer or commonly used agents against contemporary isolates.

Methods: All MIC tests (alone or in combination) used NCCLS broth microdilution methods (M7-A6) in checkerboard tray designs. ATM was combined with ciprofloxacin (CIP), gatifloxacin (GAT), levofloxacin (LEV), cefepime (CPM), ceftazidime (CAZ) and imipenem (IMP) at clinically relevant concentrations. Interaction categories were defined by established FIC calculations. 40 strains each of *P. aeruginosa* (PSA) and Enterobacteriaceae (ENT; 12 species; ATM MIC, 1-16 µg/ml) were tested for each antimicrobial combination (480 SYN determinations).

Results: The results follow in the table:

	% by interaction category:			
Organism/co-drug	SYN	Partial SYN	ADDITIVE	INDIFFERENT
PSA-CIP	12.5	45.0	37.5	5.0
GAT	10.0	57.5	25.0	7.5
LEV	12.5	52.5	27.5	7.5
CAZ	7.5	77.5	12.5	2.5
CPM	2.5	72.5	22.5	2.5
IMP	0.0	17.5	17.5	65.0
ENT-CIP	2.5	52.5	17.5	27.5
GAT	12.5	37.5	25.0	25.0
LEV	10.0	42.5	22.5	25.0
CAZ	5.0	80.0	7.5	7.5
CPM	12.5	52.5	17.5	17.5
IMP	32.5	55.0	10.0	2.5

No antagonism or indeterminate interactions were identified among 480 evaluable SYN tests. The overall rates of SYN or partial (P) SYN for ATM + fluoroquinolone combinations was 63.4% versus PSA, greatest for ATM + GAT (67.5%). Interaction categories varied greatly among ATM + ß-lactam combinations. ATM + CAZ or CPM versus PSA had 72.5 - 82.5% SYN or P-SYN, but ATM + IMP showed dominant indifference (65.0%). In contrast, ATM + IMP was more likely have SYN (32.5%) when tested against ENT.

Conclusions: ATM, often used as an aminoglycoside substitute, continues to demonstrate enhanced (no antagonism), but variable drug activity interactions for contemporary antimicrobial combinations tested against year 2002 clinical isolates.

INTRODUCTION

Aztreonam is a synthetic monocyclic ß-lactam which belongs to the family of monobactams and is exclusively active (like aminoglycosides) against the aerobic Gram-negative bacilli, including *Pseudomonas aeruginosa*. It is not ototoxic or nephrotoxic, and is often used as an alternative to the aminoglycosides in a variety of clinical situations. Combination therapy can be used to expand the antimicrobial spectrum, to prevent the emergence of resistant mutants, to minimize toxicity, and to obtain synergistic antimicrobial activity. Because of its focused spectrum of activity (aerobic Gram-negative bacteria), aztreonam is frequently used in therapeutic combinations with other antimicrobial agents.

The objectives of this study was to evaluate the in vitro interactions between aztreonam and ciprofloxacin or gatifloxacin or levofloxacin or cefepime or ceftazidime or imipenem at clinically relevant concentrations when tested against selected *P. aeruginosa* and Enterobacteriaceae strains.

MATERIALS & METHODS

Bacterial strains. A total of 40 *P. aeruginosa* and 52 Enterobacteriaceae (12 species) strains collected for the SENTRY Antimicrobial Surveillance Program in 2002 were selected for this study. All *P. aeruginosa* strains were tested against aztreonam in combination with three fluoroquinolones or three broad spectrum ß-lactams. A group of 40 strains were selected among the 52 Enterobacteriaceae to be tested against aztreonam in combination with the fluoroquinolones, while another group (also 40 strains) from the same collection was selected to be tested against aztreonam in combination with the ß-lactams. The strains were initially chosen based upon the aztreonam MIC (1 - 16 µg/ml) and additional on-scale results with the co-drug.

Among the tested Enterobacteriaceae, 12 strains produced an extended-spectrum ß-lactamase (ESBL), five strains produced a chromosomally derepressed ampC cephalosporinase, and eight strains were resistant to fluoroquinolones (ciprofloxacin MIC, ≥4 µg/ml).

Antimicrobial susceptibility and synergism testing. Synergism tests were performed in 96-well broth microdilution trays containing two antimicrobial agents in two-fold dilutions dispensed in a checkerboard format. Each tray design contained the following antimicrobial agents (MIC range tested in parenthesis) in combination with aztreonam (32 - 0.5 µg/ml): ciprofloxacin (16 - 0.016 µg/ml), gatifloxacin (16 - 0.016 µg/ml), levofloxacin (16 - 0.016 µg/ml), cefepime (32 – 0.03 µg/ml), ceftazidime (32 - 0.03 µg/ml), and imipenem (16 - 0.03 µg/ml). Standard antimicrobial powders were supplied by respective manufacturers and the six designs of test panels were prepared by TREK Diagnostic Systems Inc. (Cleveland, OH). The quality control strains used for all tests were *Esherichia coli* ATCC 25922 and *P. aeruginosa* ATCC 27853.

The interpretations of the antimicrobial combination interactions were based on the following criteria: synergy was the decrease in the MIC of each agent of ≥ four-fold; partial synergy was the decrease of one drug of ≥ four-fold and a decrease of two-fold for the other agent; additive was a two-fold reduction in the MIC of both agents; indifference was all interactions not meeting the criteria listed above and not having antagonism where a MIC increase of ≥ four-fold for each drug would be observed in combination; and the fractional inhibitory concentration (FIC) of each agent was also calculated by dividing MIC of the drug in combination by the MIC of the drug alone. The sum of both FICs (∑FIC = FIC of drug A + FIC of drug B) in each well was used to classify the combination interactions of antimicrobial agents as synergistic (∑FIC, ≤0.5), partially synergy (∑FIC, >0.5 and <1), additive (∑FIC, 1), indifferent (∑FIC, >1 and <4) and antagonism (∑FIC, ≥4).

RESULTS

Table 1. Drug interaction (synergy) studies for aztreonam combined with three fluoroquinolones and three other ß-lactams when tested by broth checkerboard method against 80 isolates of *P. aeruginosa* and various Enterobacteriaceae.[a]

	Co-drug (occurrences [%])					
Organism/category[b] (no. tested)	CIP	GAT	LEV	CAZ	CPM	IMP
P. aeruginosa (40)						
Synergy	5(12.5)	4(10.0)	5(12.5)	3(7.5)	1(2.5)	0(0.0)
Partial synergy	18(45.0)	23(57.5)	21(52.5)	31(77.5)	29(72.5)	7(17.5)
Additive	15(37.5)	10(25.0)	11(27.5)	5(12.5)	9(22.5)	7(17.5)
Indifferent	2(5.0)	3(7.5)	3(7.5)	1(2.5)	1(2.5)	26(65.0)
Enterobacteriaceae (40)						
Synergy	1(2.5)	5(12.5)	4(10.0)	2(5.0)	5(12.5)	13(32.5)
Partial synergy	21(52.5)	15(37.5)	17(42.5)	32(80.0)	21(52.5)	22(55.0)
Additive	7(17.5)	10(25.0)	9(22.5)	3(7.5)	7(17.5)	4(10.0)
Indifferent	11(27.5)	10(25.0)	10(25.0)	3(7.5)	7(17.5)	1(2.5)

a. The strains were selected from a group of 52 strains, which included: *Citrobacter freundii* (four strains), *E. coli* (nine strains), *Enterobacter aerogenes* (three strains), *E. cloacae* (seven strains), *Hafnia alvei* (one strain), *Klebsiella oxytoca* (one strain), *K. pneumoniae* (four strains), *Morganella morganii* (five strains), *Pantoea agglomerans* (one strain), *Proteus mirabilis* (10 strains), *Providencia stuartii* (one strain) and *Serratia marcescens* (six strains).

COMMENTS

- Synergy or partial synergy were detected for the majority of strains for all aztreonam combinations evaluated, except for aztreonam plus imipenem against *P. aeruginosa* (17.5% partial and 0.0% synergy).
- The combinations with the highest frequency of synergism or partial synergy against *P. aeruginosa* were aztreonam with ceftazidime or cefepime. The majority of strains showed partial synergism for both combinations (77.5 and 72.5% respectively), and 97.5% of strains showed at least an additive effect.
- The fluoroquinolones combinations showed synergy or partial synergy against 57.5 to 67.5% of *P. aeruginosa* strains, and >90% of strains showed at least an additive effect.
- Against Enterobacteriaceae, the occurrence of synergistic or partial synergistic interactions was higher among the aztreonam/ß-lactam combinations (65.0 - 87.5%) compared to the aztreonam/fluoroquinolone combinations (50.0 - 55.0%).
- Aztreonam with cefepime showed at least an additive effect against 82.5% of the Enterobacteriaceae strains.
- Among the 24 combinations tested against ESBL-producing Enterobacteriaceae, 20 (83.3%) resulted in synergy or partial synergy; three (12.5%) resulted in an additive effect and only one strain (4.2%) resulted in an indifferent interaction (aztreonam with cefepime for a *Proteus mirabilis* isolate).
- The results were very similar among the three aztreonam/fluoroquinolone combinations evaluated with synergy or partial synergy documented against approximately one-half of the strains tested (50.0 - 55.0%), additive affect against 17.5 to 25.0% of strains, and an indifferent effect in 25.0 to 27.5% of strains.
- The highest rate of synergism/partial synergism was observed for aztreonam with imipenem when tested against Enterobacteriaceae (87.5%); while the lowest rate of favorable interactions was detected for the same combination against *P. aeruginosa* (17.5%).

CONCLUSIONS

- The synergy results of aztreonam combinations were very favorable for the majority of contemporary strains evaluated in this study and antagonism was not detected among 480 tests.
- The interaction between aztreonam and imipenem may vary significantly according to bacterial species and should be further evaluated with a larger and more diverse collection of strains.
- The results of this investigation are very encouraging with respect to potential safe clinical applications of combined therapy of aztreonam with six commonly used drugs among fluoroquinolone or ß-lactam classes of antimicrobials.

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