Efficacy of the extended spectrum β-lactamase inhibitor, AAI101, combined with β-lactams in murine models of systemic Gram-negative infection

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Introduction
AAI101 is a novel zwitterionic extended-spectrum β-lactamase inhibitor (BLI) belonging to the penicillanic acid sulfone class, currently in Phase I trials.

A major challenge confronting antibacterial chemotherapy is the emergence and global dissemination of extended-spectrum β-lactamases (ESBLs). These enzymes deactivate β-lactam antibiotics (including penicillins, cephalosporins, and monobactams) before they can access their targets in the periplasmic space or inner wall zone, thereby restricting treatment options for infections attributable to ESBL-producing pathogens.

Development of BLIs such as tazobactam (Tazo) has helped preserve the clinical value of β-lactam antibiotics by protecting them against hydrolysis. For treatment of community-onset sepsis, clinicians often prefer third-generation cephalosporins (e.g. ceftriaxone [Cro]); for patients with a high-burden of healthcare-associated factors or febrile neutropenia, cefepime (Fep) or piperacillin (Pip)/Tazo is preferred.¹ However, the efficacy of these treatments is being compromised by appearance and spread of aggressive β-lactams not susceptible to existing BLIs.²

This study was designed to assess the efficacy in mice of AAI101 vs. Tazo in combination with Pip, Cro, or Fep for treatment of systemic infections caused by ESBL-producing Enterobacteriaceae.

Materials and Methods

- ESBL-producing Enterobacteriaceae were obtained from St. John’s Medical College Hospital, Bangalore, India, and from the Sassoon General Hospital affiliated with the B. J. Medical College, Pune, India; ESBL production was confirmed by broth microdilution and disc diffusion as previously described by the CLSI.³
- Broth microdilution minimum inhibitory concentrations (MICs) (mg/L) were obtained for β-lactams, β-lactams + AAI101 4 mg/L, and β-lactams + Tazo 4 mg/L, respectively, using CLSI protocols, as follows:
  - *Escherichia coli* MRO 10006, Pip: >128, 1, 1
  - *E. coli* MRO 10007, Pip: >128, 0.5, 1
  - *Klebsiella pneumoniae* MRO 11008, Pip: >128, 4, 4
  - *Citrobacter freundii* MRO 12301, Cro: >64, 0.06, 0.25
  - *C. freundii* MRO 12301, Fep: 16, 0.03, 0.03.
- Immunocompetent female Swiss albino mice (18-22 g, obtained from a GLP-certified breeding facility at Orchid Chemicals & Pharmaceuticals, Ltd., Chennai, India; 5-6 animals per cohort) were injected i.p. with lethal doses of bacteria (6-82 x LD₅₀) suspended in physiological saline containing 5% (v/v) hog gastric mucin.⁴

Results

- At 0.5 h post-infection, single doses of Pip, Cro, or Fep, ± AAI101 or Tazo at an antibiotic/BLI (w/w) ratio of 4:1, were administered s.c. (5 mL/kg of body weight).
- Animals were monitored twice daily for 7 days, and drug doses corresponding to ED₅₀s for each treatment group were calculated by the method of Reed & Muench.⁵

<table>
<thead>
<tr>
<th>Strain</th>
<th>ED₅₀ (mg/kg)</th>
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<tbody>
<tr>
<td><em>E. coli</em> MRO 10006</td>
<td>Pip alone, &gt;128 Pip/AAI101 4:1, 42.2 Pip/Tazo 4:1, 53.8</td>
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<tr>
<td></td>
<td>Pip/AAI101 4:1, 24.9 Pip/Tazo 4:1, 58.8</td>
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<tr>
<td><em>K. pneumoniae</em> MRO 11008</td>
<td>Pip alone, not determined Pip/AAI101 4:1, 34.9 Pip/Tazo 4:1, 99.5</td>
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<tr>
<td><em>C. freundii</em> MRO 12301</td>
<td>Cro alone, &gt;40 Cro/AAI101 4:1, 2.3 Cro/Tazo 4:1, 22.5</td>
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<td>Pip alone, &gt;10 Pip/AAI101 4:1, 1.7 Pip/Tazo 4:1, 7.1</td>
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Conclusions

- When administered with Pip, Cro, or Fep, AAI101 proved more efficacious than equiproportionate ratios of Tazo at improving survival in mice treated with lethal doses of ESBL-producing Enterobacteriaceae.
- AAI101, in combination with an established β-lactam, is a promising new therapeutic modality for the treatment of infections caused by ESBL-producing Enterobacteriaceae.

References
3. Clinical and Laboratory Standards Institute, 2014. ²⁰th Informational Supplement M100-S24. CLSI, Wayne, PA, USA.

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