A Novel β-lactamase Inhibitor (Durlobactam, DUR) and β-Lactams Enhance Susceptibility Against Multidrug-Resistant (MDR) Mycobacterium abscessus (Mab)

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Background

- Mab is a MDR nontuberculous mycobacterium that causes invasive pulmonary infections in patients with structural lung disease. Mab harbors a chromosomally encoded class A β-lactamase, BlaMab, able to hydrolyze penicillins, most cephalosporins and carbapenems.
- L,D- and D,D-transpeptidases (L,D-TP and D,D-TP, respectively) shape peptidoglycan (PG) synthesis and contribute to cell wall structure.
- Select combinations of β-lactams that inhibit L,D-TP and D,D-TPs and BlaMab are desirable as they can potentially improve treatment outcomes.
- Durlobactam (DUR) is a novel DBO β-lactamase inhibitor (BLI) with broad-spectrum activity against Ambler class A, C, and D β-lactamases (Figure 1).
- Here, we investigated the mechanism of action and efficacy of DUR alone and combined with select β-lactams in restoring susceptibility of Mab to β-lactam antibiotics.

Methods

Methods

Minimum inhibitory concentrations (MICs) of cefuroxime (CEF), imipenem (IMI) and amoxicillin (Amox) with or without DUR were determined using microdilution. Approximately 5 x 10⁶ colony-forming units (CFU) per milliliter were inoculated into Middlebrook 7H9 broth supplemented with 10% (vol/vol) oleic albumin dextrose catalase and 0.05% (vol/vol) Tween 80. When more than 2 drugs were combined, Amox was added at fixed concentration of 8 μg/mL to serial dilutions of CEF-DUR or IMI-DUR in a 1:1 ratio. Mab isolates were incubated with test agents at 30°C for 48 h, and MIC was defined as lowest antibiotic concentration that prevented visible bacterial growth. Reaction intermediates in the inactivation pathway of BlaMab, L,D-TP and D,D-TPs with DUR were captured using mass spectrometry (QTOF-MS).

Table: MIC50 and MIC90 of 100 Mab clinical strains against DUR alone and in combination with Amox, CEF and IMI

<table>
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<th></th>
<th>DUR</th>
<th>Amox</th>
<th>Amox/DUR (1:1)</th>
<th>CEF</th>
<th>CEF/DUR (1:1)</th>
<th>CEF/DUR + Amox 8 μg/mL</th>
<th>CEF/amox 8 μg/mL</th>
<th>IMI</th>
<th>IMI/DUR</th>
<th>IMI/DUR + Amox 8 μg/mL</th>
<th>IMI/Amox (1:1)</th>
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<td>8</td>
<td>≤0.06</td>
<td>4</td>
<td>2</td>
<td>2</td>
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<tr>
<td>MIC90</td>
<td>8</td>
<td>≥256</td>
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<td>0.25</td>
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DUR, CEF (Cefuroxime), Amox (Amoxicillin), Imipenem (IMI)

Figure 2: Mass spectrometry of BlaMab, L,D-TP and D,D-TPs incubated with DUR

Figure 1: Chemical composition of DUR

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References

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Conclusion

We demonstrate that a novel DBO BLI, DUR, is an active agent with potent intrinsic activity against BlaMab and Mab L,D-TPs and D,D-TPs.

We hypothesize that DUR improves β-lactam activity by protecting against the hydrolytic activity of BlaMab and by targeting multiple steps in PG synthesis.

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