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Poster

## [P25-2] P25-2: Anti-infective drugs (2): Beta-lactams

Chair: Veronique Stove, Belgium

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### [P25-2-1] An UHPLC-HR-MS assay for beta-lactams in bronchoalveolar lava fluid

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Keywords: Piperacillin, Tazobactam, Meropenem, Intensive care unit, Epithelial lining fluid

#### Background

There is a growing interest in therapeutic drug monitoring of  $\beta$ -lactam antibiotics for critically ill patients. Tissue concentrations may be a more reliable estimate for efficacy. For pneumonia patients in intensive care, antibiotics in epithelial lining fluid can be assayed to estimate lung tissue concentrations. Sampling is done by taking bronchoalveolar lavage fluid (BALF). We developed a method for analyzing meropenem and piperacillin/tazobactam in BALF.

#### Methods

We used a UHPLC-HR-MS method on a Thermo Fisher Q-Exactive Hybrid Quadrupole-Orbitrap mass spectrometer for the assay of the  $\beta$ -lactam antibiotics meropenem and piperacillin and for  $\beta$ -lactamase inhibitor tazobactam. Deuterated and <sup>13</sup>C, <sup>15</sup>N labeled internal standards were used. Fifteen L of sample was added to 100L internal standard in acetonitrile. After vortexing for 3 min at 1400rpm and 9°C and centrifugation for 5 min at 14800 rpm, the supernatant was transferred to an autosampler vial for injection. Analysis was performed using an Accucore Phenylhexyl column 100\*2.1mm with a 2.6m particle size. One L of sample was injected. The autosampler temperature and column temperature were 10°C and 40°C respectively. Mobile phase A consisted of 3mM ammonium formate and 0.1% formic acid in water, MF B of 2mM ammonium formate and 0.1% formic acid in 50/50 (v/v) methanol/acetonitrile. The gradient started at 98% A and 2% B (flow rate 0.4mL/min), switched to 5% A after 3 minutes, maintained for 4 minutes, then switched back to starting conditions. Acquisition was in full scan, from m/z 50 to 680.

#### Results

Method validation was based on the EMA Guidelines. The method was selective for meropenem, piperacillin and tazobactam. Carry-over was within the predefined limits. A within-run bias between 2.64% and -14.93% and a CV between 1.27% and 6.51% were found. Between-run bias was -7.98-11.93%, accuracy 1.58%-9.07%. The range of the calibration curves was 0.25-250mg/L for meropenem and piperacillin, 0.125-125mg/L for tazobactam. At room temperature, meropenem was stable in BALF for 12h, piperacillin for 24h, tazobactam for at least 4 days.

#### Conclusions

A sensitive and specific UHPLC-HR-MS method was developed and validated for 2  $\beta$ -lactam antibiotics and a  $\beta$ -lactamase inhibitor in BALF.