P1090
Poster Session IV
Enterobacteriaceae: resistance and fitness

INFLUENCE OF BICARBONATES ON SENSITIVITY OF DIAGNOSTIC TESTS FOR OXA-48-PRODUCING ENTEROBACTERIACEAE

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Objectives: Resistance to carbapenems has been reported all over the world. Its most important mechanism is the production of carbapenem-inactivating beta-lactamases (carbapenemases) which hydrolyse the amide bond of the ?beta-lactam ring in diverse beta-lactam molecules. The OXA-48-type carbapenemases have spread extensively in Middle East, North Africa and India. In Europe, OXA-48-type-producing *Enterobacteriaceae* (including variants OXA-48, -162, -163, -181, -204, -232, -244, -245, -247) have been found in many countries, often due to imports from endemic regions, and their regional spread was reported for example in Spain, France, Belgium, UK or Ireland.

Previous studies showed that the most important factor of the activity of OXA-type beta-lactamases is the carboxylated lysine residue in the active site. Therefore, we tested the effect of bicarbonates on OXA-48-type producers in order to check whether this might increase the sensitivity of cultivation media used for the detection of these organisms and improve the sensitivity of the MALDI-TOF MS meropenem hydrolysis assay.

Methods. The influence of bicarbonates on carbapenem MICs against OXA-48-producing *Enterobacteriaceae* was tested. We also checked whether the addition of bicarbonates to liquid media supplemented with meropenem may facilitate the selective enrichment of various carbapenamase producers in cultures. Furthermore, the sensitivity of carbapenemase confirmation by MALDI-TOF MS meropenem hydrolysis and spectrophotometric hydrolysis assays upon the addition of ammonium bicarbonate was examined.

Results. The addition of sodium bicarbonate significantly increased MICs of ertapenem and meropenem for OXA-48 producers. Furthermore, liquid media supplemented with sodium bicarbonate and meropenem were reliable for the selective enrichment of carbapenemase producers. The presence of ammonium bicarbonate in buffers used in the spectrophotometric and MALDI-TOF MS carbapenemase detection increased the sensitivity of these assays.

Conclusions. Our results demonstrate that bicarbonates in media or reaction buffers can enhance the sensitivity of screening methods and diagnostic tests for carbapenemase producers. Moreover, we hypothesize that the categorization of OXA-48-producing clinical isolates to susceptible/intermediate/resistant categories based on the standard susceptibility testing may be inaccurate. Based on the data, we also propose that for clinical studies describing an effect of carbapenems in carbapenemase-producing organisms, the detailed data on their beta-lactamase content should be always provided.

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