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P1348 Hidden carbapenem-resistance in OXA-48 and ESBL-positive *Escherichia coli*

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The purpose of this study was to report the identification OXA-48 carbapenemase in seven ESBL positive *E. coli* clinical isolates, susceptible to carbapenems by disk - diffusion and E-test method, but with a borderline MIC value of ertapenem (1 mg/L). Unexpectedly, three of seven isolates were shown to harbour A/C plasmid, previously associated with VIM or NDM MBLs. This finding prompted us to analzye the mechanisms of reduced susceptibility to ertapenem.

Seven ertapenem non-susceptible *E. coli* originated from different anatomic sites were analyzed. The isolates were collected over 12 months period from patients hospitalized in GH Slavonski Brod, Slavonia region, Croatia. The susceptibility to a wide range of antibiotics was determined by disk-diffusion and broth microdilution method according to EUCAST. The transferability of cefotaxime resistance and reduced susceptibility to meropenem was determined by conjugation. Carbapenemases and extended-spectrum β-lactamases were phenotypically detected. The presence of bla_{CARB} (bla_{KPC}, bla_{VIM}, bla_{IMP}, bla_{NDM}, bla_{OXA-48}) and bla_{ESBL} genes (bla_{SHV}, bla_{TEM}, bla_{CTX-M}) was determined by PCR and sequencing. PCR-based replicon typing (PBRT) was applied to determine the plasmid content of the tested isolates. PFGE genotyping of XbaI-digested genomic DNA was performed with CHEF-DRIII system.

Isolates showed a high level of resistance to most of the antibiotics and were susceptible to colistin, amikacin, tigecycline, and fosfomycin. Despite susceptibility to ertapenem in disk-diffusion and E-test, dilution test yielded borderline MICs of 1 mg/L. Hodge and Carbapenem Inactivation Method test were positive indicating the production of carbapenemase.

All isolates transferred cefotaxime and meropenem resistance to *E. coli* recipient strain. The isolates were shown to possess group 1 of CTX–M β-lactamases and TEM–1 in addition to OXA-48. Bla_{OXA-48} genes were preceeded by IS*1999* whereas IS*Ecp* was found upstream of $bla_{CTX-M-15}$ genes in three isolates. PFGE analysis revealed two clusters and containing subgroups of highly related isolates.

This report points to the need for determination of carbapenem MICs in *E. coli* ESBL positive isolates and additional testing for all isolates with borderline ertapenem MIC defined by EUCAST. Unlike previous studies in Croatia, the dissemination of OXA-48 in Slavonia region was associated with highly related isolates, belonging into two clusters.