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Colistin-resistant metallo-beta-lactamase-producing Klebsiella pneumoniae isolates in a tertiary care Greek university hospital

Ephthalia Priavali*[1], Constantina Gartzonika[1], Georgia Vrioni[2], Leda Politi[3], Andromachi Giannaki[1], Dimitrios Papamichail[1], Hercules Sakkas[4], Georgia Kapnisi[1], Athanassios Tsakris[2], Stamatina Levidiotou[1]

[1]*Department of Microbiology, Faculty of Medicine, University of Ioannina*

[2]*Medical School, University of Athens; Microbiology*

[3]*Medical School, University of Athens; Department of Microbiology*

[4]*University of Ioannina, Medical School, Microbiology Department; Microbiology*

Background: Colistin is a backbone component of combination antimicrobial regimens for serious infections caused by carbapenemase-producing *Klebsiella pneumoniae* strains (CP-Kp), which are among the most challenging multidrug-resistant (MDR) pathogens spreading worldwide. Thus, the emergence of colistin resistance limits treatment options and represents a global threat. The aim of this study was to asses the incidence of metallo-β-lacamase (MBL)-producing *K. pneumoniae* isolates over a 4.5-year period, as well as their resistance rates to colistin and tigecycline, which are included in last-resort antimicrobials.

Material/methods: Between January 2010 and June 2014, a total of 614 consecutive non-repetitive, carbapenem-nonsusceptible *K. pneumoniae* isolates, recovered from clinical samples or surveillance cultures, taken as part of standard care of patients hospitalized at the 750-bed University Hospital of Ioannina. Demographic data collected from patient's medical records. Identification and susceptibility testing were performed using the Vitek 2 system (bioMerieux, France). MICs of carbapenems, colistin and tigecycline were additionally determined using Etest (bioMerieux) following CLSI, EUCAST criteria and US FDA recommendations, respectively. Carbapenemase production was screened with the modified Hodge test and the EDTA/phenylboronic acid combined disk test using meropenem as substrate. Acquired β-lactamase genes were detected by PCR assays and sequencing analysis.

Results: During the study period, 150 of 614 (24.4%) carbapenem-nonsusceptible *K. pneumoniae* isolates were phenotypically suspected of metallo-β-lactamase production. Molecular characterization revealed that among the 150 phenotypically MBL-positive isolates, 117 (78%) carried the bla_{NDM-1} gene, which has been introduced in our institution in 2011, and 33 (22%) carried the bla_{VIM-1} gene. All genotypically MBL producers displayed MDR phenotypes. As colistin-resistant (COL-R) were categorized 25 (16.7%) isolates (MIC range 8-32 μg/ml), retrieved mainly from urine (64%) and blood cultures (16%), followed by wound (12%) and surveillance rectal (8%) swabs. The majority of them (52%) were recovered from patients in medical wards, while 36% were recovered from ICUs and 12% from surgical wards. The colistin resistance rate was approximately the same between bla_{VIM-1} and bla_{NDM-1} positive isolates (18.2% and 16.2%, respectively). The COL-R, bla_{NDM-1}-producing isolates, were detected the last 2.5 years of the study period, following the emergence of this new carbapenemase, and co-harbored $bla_{CTX-M-15}$, bla_{TEM-1} and bla_{OXA-1} genes. Sixty per cent of patients with COL-R isolates were male and 64% were older than 65 years. None exhibited a pan-resistant phenotype. The attributable mortality rate was 16% (4 patients, all bla_{NDM-1} positive). Tigecycline resistance was detected among 9 NDM-1-producing isolates (MIC range 8-16 μg/ml).

Conclusions: NDM-1 carbapenemase was found to predominates among MBL producers. The spread of NDM-1-producing *K. pneumoniae* was associated, with the emergence of colistin resistance during the latter study period. Continuous surveillance, prudent use of colistin and effective implementation of infection control measures are urgent in our setting in order to preserve this salvage treatment option.