**TITLE:** MOLECULAR EPIDEMIOLOGY AND ANTIMICROBIAL RESISTANCE PROFILE OF KPC-PRODUCING *Enterobacter cloacae* COMPLEX RECOVERED FROM RECREATIONAL COASTAL WATERS IN RIO DE JANEIRO

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## ABSTRACT:

Enterobacter cloacae complex commonly cause nosocomial infections. KPC production in these species is frequent not only in clinical isolates, but also among environmental ones. Previously, we described wide dissemination of carbapenemase-producing Enterobacteriaceae in recreational coastal waters over one year (2013-2014), among which KPC-producing Enterobacter cloacae complex was predominant. In this study, we aimed at evaluating their antimicrobial susceptibility profile, occurrence of resistance genes and their clonal variability. A total of 61 isolates was studied, harboring either  $bla_{KPC-2}$  (59) or  $bla_{KPC-26}$  (2) alleles. Highest resistance rates were observed for imipenem and ceftriaxone (95%), followed by amoxicillinclavulanate (93%), meropenem (92%), aztreonam (85%), cefepime (61%), ceftazidime (47%), gentamicin (33%), trimethoprim-sulfamethoxazole (15%), amikacin (10%) and ciprofloxacin (5%). Antimicrobial resistance determinants detected in addition to  $bla_{KPC}$  included  $bla_{GES}$  (5), bla<sub>CTX-M-2-like</sub> (1), bla<sub>CTX-M-8-like</sub> (9), bla<sub>CTX-M-14-like</sub> (6), bla<sub>TEM-like</sub> (18), bla<sub>SHV-like</sub> (17), qnrA (20), qnrB (4), qnrS (1), and qnrVC (17). Genes encoding 16S rRNA methyltransferases and transferable resistance to polymyxins were not detected. Of notice, we found isolates carrying up to five  $(bla_{KPC-2} + bla_{CTX-8-like} + bla_{CTX-14-like} + qnrA + bla_{SHV})$  and up to four  $(bla_{KPC-2} + bla_{CTX-8-like} + bla_{CTX-14-like})$ + qnrA) different resistance genes in waters unsuitable and suitable for recreation, respectively. A total of 51 distinct RAPD profiles were found and four samples were nontypable. Curiously, four isolates carrying bla<sub>KPC</sub>, qnrA and bla<sub>SHV</sub> and showing undistinguishable RAPD profiles were recovered from three different beaches, from samples collected over 100days interval. We also observed RAPD clones carrying almost identical repertoire of resistance genes, except for the predominantly environmental gene qnrVC whose presence was not ubiquitous in all clonally-related isolates. Our findings suggest that coastal waters are not only important reservoirs of antimicrobial-resistant threats, but also matrices where bacteria may increase their repertoire of resistance determinants. Persistent clones identified will be further studied to address whether they are constantly disposed in coastal waters through sewer contamination or if they are able to remain viable and grow on these aquatic matrices. Role of pollution in this scenario will also be evaluated.

**Keywords:** *Enterobacter cloacae* complex, carbapenemase, *bla*<sub>KPC</sub>, recreational water.

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