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P-175 - CARBAPENEM-RESISTANT KLEBSIELLA PNEUMONIAE CLINICAL ISOLATES HARBOURING SIMILAR CONJUGATIVE PLASMIDS

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Background

Klebsiella pneumoniae is a major cause of health care associated infections, with high incidence in urinary tract infections, pneumonia, septicaemia and soft tissue infections. The occurrence of multidrug-resistance in this pathogen seriously increases the severity of the associated infections and the increasing occurrence of carbapenem resistance has become evident in the last years. Often, this phenotype is associated with the carbapenemase encoding, plasmid borne *bla_{KPC}* gene. The comparison of plasmids among isolates is a common approach to infer the mechanisms of spread of antibiotic resistance genes, herein used to infer *bla_{KPC}* dissemination in clinical isolates.

Method

Carbapenem-resistant *K. pneumoniae* isolates from clinical samples were characterized in terms of phenotypic and genotypic characteristics and number of plasmids. Plasmids further characterized were those that could be transferred by conjugation to a selected receptor. Conjugation assays were carried using clinical carbapenem-resistant *K. pneumoniae* isolates as donors and an environmental *K. pneumoniae* as recipient. Transconjugants were selected in the presence of tetracycline (16 mg/L) and meropenem (0.25 mg/L) and further characterized for resistance phenotypes, specific genotypes and plasmid profiles. The number and sizes of plasmids were compared based on pulsed field gel electrophoresis and optical DNA mapping (ODM). ODM was also used for plasmid profiling.

Results & Conclusions

The clinical carbapenem-resistant *K. pneumoniae* isolates were resistant to different classes of antibiotics (beta-lactams, aminoglycosides, quinolones, cephalosporins, sulphonamides and carbapenems) and susceptible to tetracycline. Among the 2-4 plasmids observed in a single isolate, in general, only one could be transferred by conjugation. These plasmids had sizes ranging from 50 kbp to 140 kbp and all contained the gene *bla_{KPC}*. In transconjugants, this gene was observed to be associated with the replicon types FIA and N, and to the acquisition of resistance to meropenem, cephalothin and ceftazidime. Ciprofloxacin resistance was not observed to be transferred. Based on ODM analysis it was possible to confirm a high similarity between the conjugative plasmids from different clinical isolates.

The combination of ODM and conjugation assays gave evidence for the spread of *bla_{KPC}* via horizontal gene transfer among clinical isolates and the potential of dissemination also for environmental strains. The association of specific replicon types in *K. pneumoniae* to conjugative plasmids harbouring the *bla_{KPC}* gene is of interest for predicting how the spread of carbapenem resistance occurs.

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