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ePoster Viewing

Molecular bacterial typing methods

Genetic relatedness of *Pseudomonas aeruginosa* isolated from cystic fibrosis patients attending a clinic at a tertiary academic hospital in Pretoria, South Africa

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Background: Cystic fibrosis (CF) is an inherited recessive genetic disease caused by a mutation in the cystic fibrosis transmembrane regulator (*CFTR*) gene, encoding for a protein responsible for chloride and sodium transport. The dysfunction of this protein results in an ionic imbalance and subsequent dehydration of the mucus, impaired mucociliary clearance, innate immune dysfunction and consequent bronchiectasis. As a result the removal of lung pathogens, such as *Pseudomonas aeruginosa*, is impaired and these pathogens are able to colonise the lungs. In South Africa, there is limited data available regarding the molecular characteristics and genetic relatedness of *Pseudomonas aeruginosa* in CF.

Material/methods: Multi-site samples from cough swabs, sputum and nasal swabs were collected from patients attending a CF clinic in Pretoria, South Africa. Specimens were processed routinely by the Diagnostic Division of the Department of Medical Microbiology (UP/NHLS) using culture and the VITEK 2 system (bioMérieux, France) and were additionally cultured onto selective media [ChromID *P. aeruginosa*, (bioMérieux, France)]. All presumptive *P. aeruginosa* isolates were confirmed using matrix-assisted laser desorption ionisation-time of flight (MALDI-TOF) (Bruker Daltonics, USA). Pulsed field gel electrophoresis (PFGE), BOX-PCR and ERIC-PCR assays were performed on all the *P. aeruginosa* isolates to determine their genetic relatedness. This was followed by multi-locus sequence typing (MLST) on selected isolates.

Results: A total of 23 *P. aeruginosa* isolates were collected from five patients. Eleven of the *P. aeruginosa* (48%) isolates were untypeable using PFGE and the remaining twelve isolates (52%) could be grouped into two pulsotypes (with four outliers) using a similarity coefficient of 80%. Pulsotype A constituted 50% (6/12) of the isolates, whereas pulsotype B constituted 17% (2/12) of the isolates and the outliers made up the remaining 33% (4/12). Two isolates were untypeable using ERIC-PCR and the remaining 21 isolates (91%) could be divided into four groups (with five outliers) using a similarity coefficient of 70%. The majority of the isolates belonged to group I [30% (7/23)], followed by group III [17% (4/23)], group IV [13% (3/23)] and by group II [9% (2/23)], while the outliers made up 22% (5/23). The BOX-PCR had two untypeable isolates and the remaining 21 isolates (91%) formed one group [19/23 (82%)] with two [2/23 (9%)] outliers using a similarity coefficient of 70%. The

MLST analysis showed three previously reported sequence types (STs): ST395, ST554, ST1062 and a new sequence type, ST2225.

Conclusions: This study showed a genetically diverse *P. aeruginosa* population with limited patient to patient spread. Most of the sequence types identified in this study were uncommon and have previously been reported sporadically. Similarly, this was the first reported instance of ST1062 in CF and a new sequence type, ST2225, which has not been identified before.