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Paper Poster Session

Antibacterial resistance: the animal and food production part of one health

Characterization of antimicrobial resistance in *Pseudomonas* spp. isolates from healthy animals and children

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Background: *Pseudomonas* spp. colonise diverse terrestrial and aquatic habitats, and certain species are important opportunistic pathogens causing plant and human diseases. The significance of *Pseudomonas* spp. is associated with its high resistance to antimicrobials. Most *Pseudomonas aeruginosa* studies had focused on clinical human strains, but studies on healthy humans or animals are scarce or null.

The aim of this study was to characterise *Pseudomonas* spp. recovered from faecal samples of healthy animals and children.

Material/methods: A total of 167 *Pseudomonas* spp. isolates were recovered from healthy animals (n=133) and healthy children (n=34). Clonal relationship among isolates was studied by *SpeI*-PFGE and MLST. Susceptibility to 12 antipseudomonal agents was established by disc diffusion method (EUCAST, 2015). Metallo-beta-lactamases (MBL) and extended spectrum-beta-lactamases (ESBL) phenotypes were determined by double-disc-synergy method. The presence and characterization of integrons were carried out by PCR and sequencing. Virulence markers (*exoU*, *exoS*, *exoY*, *exoT*, *exoA*, *lasA*, *lasB*, *aprA*, *rhlAB*, *rhlI*, *rhlR*, *lasI*, *lasR*) were detected in *P. aeruginosa* strains by PCR. The OprD alterations were analysed by PCR, sequencing and comparison with the sequence of the *P. aeruginosa* PAO1 control strain.

Results: A total of 109 different PFGE patterns were detected among the 167 recovered isolates. According to different PFGE and phenotypes, 113 *Pseudomonas* spp. isolates, which belonged to the following species: *P. aeruginosa* (42), *Pseudomonas* sp. (30), *P. putida* (14), *P. lundensis* (5), *P. fragi* (5), *P. fluorescens* (4), *P. protegens* (3), *P. reactans* (2), *P. koreensis* (1), *P. mendocina* (1), *P. plecoglossicida* (1), *P. brassicacearum* (1), *P. pseudoalcaligenes* (1), *P. baetica* (1); *P. fulva* (1) and *P. cedrina* (1), were further characterized in this study. The resistance percentages detected were: 70.8% ticarcillin, 42.5% aztreonam, 12.4% cefepime, 9.7% ceftazidime, 7.9% piperacillin-tazobactam, 7.9% meropenem and 1.8% imipenem. All isolates were susceptible to netilmicin, gentamicin, amikacin, tobramycin and ciprofloxacin. Neither ESBL nor MBL phenotypes were found. A high diversity of virulence markers was detected among the 42 *P. aeruginosa*, being those 21.4% *exoU* - positive ones recovered only from healthy children. One *P. aeruginosa* strain showed an insertion sequence truncating its *lasR* gene. A high variety of alterations in OprD protein was detected. *P. aeruginosa* strains belonged to a wide variety of sequence type (ST), such as ST27, ST560 and ST1711 previously described in clinical animal strains. Additionally, new STs were identified in this study, as ST1732, ST2096 and ST2194.

Conclusions: *Pseudomonas* spp. isolates showed low levels of antimicrobial resistance, with exception of resistance levels to ticarcillin and aztreonam. Moreover, many *P. aeruginosa* strains from children amplified *exoU* gene, being a major risk factor in clinical outbreak. ST found in clinical animal strains (ST27, ST560 and ST1711) have also been identified in our *P. aeruginosa* strains.