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Abstract (poster session)

The active efflux system MexXY/OprM may be activated by different mutations in clinical Pseudomonas aeruginosa

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Objectives: Constitutive overproduction of the pump MexXY/OprM (XY/M+) results in an increased resistance to aminoglycosides (AGs),ciprofloxacin (Cip), and cefepime (Fep) in P. aeruginosa. To date, three types of in vitro mutants upregulating the mexXY operon have been identified. While the agrZ mutants harbor mutations in a repressor gene (mexZ) controlling operon mexXY, those of the agrW1 type exhibit various defects in the ribosomal machinery all associated with overexpression of a gene of unknown function, PA5471, whose product ultimately induces mexXY transcription. Finally, in the agrW2 mutants, the mexXY operon is activated following alteration of a two-component regulatory system, named ParRS, also involved in carbapenem and colistin resistance. The goal of this study was to determine the prevalence of these three types of mutants in a collection of XY/M+ clinical strains. Methods: 93 XY/M+ strains were selected from our laboratory collection and analysed by Clondiag chips to investigate their clonal relatedness. Their susceptibility to antibiotics was determined and interpreted according to EUCAST recommendations. RT-qPCR was used to measure specific gene expressions. Amino acid substitutions in protein MexZ were mapped to the 3D structure of TtgR, a MexZ homolog in P. putida. Results: Forty four (77.2%) agrZ mutants, 7 (12.3%) agrW1 mutants, and 5 (8.8%) agrW2 were identified among the 57 clonally distinct XY/M+ strains. One additional isolate belonged to both the agrW1 and agrW2 types. In average, the agrW mutants were 2-fold more resistant to AGs than the agrZ ones. These latters were found to harbour various mutations inactivating gene mexZ or introducing single amino acid changes in the DNA binding-, the dimerization-, or the signal transduction- domain of repressor MexZ. Two agrW1 mutants exhibited single point allelic alterations in the genes encoding 23S rRNA, and another one a 7 nucleotide deletion in the leader peptide of PA5471. The 4 remaining agrW1 strains displayed a delayed exponential growth phase suggesting defects in protein synthesis. Finally, all the agrW2 mutants contained amino acid variations in sensor protein ParS. Conclusions: This study demonstrates that clinical strains of P. aeruginosa may use different regulatory circuitries to overproduce the MexXY/OprM pump and become multidrug resistant. Though less prevalent than the agrZ mutants, the agrW strains are more resistant to AGs.