

OMP Proteomic analysis of Benzalkonium Chloride and Ciprofloxacin adapted Biofilm cells

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Adaptive resistance to antimicrobials has been widely reported in planktonic studied through phenotypic characterization and proteomic analysis. Concerning biofilm adaptation, the response of biofilm-entrapped cells to chemical stress conditions is not yet well studied. There is evidence that proteins involved in oxidative stress response, cell envelope synthesis, as well as in synthesis of EPS become up- or down-regulated in biofilms, indicating that these altered phenotypes might contribute to antimicrobial tolerance. This work aimed to examine whether exposure of *Pseudomonas aeruginosa* biofilms to benzalkonium chloride (BC) and ciprofloxacin (CIP) could induce an adaptive response in bacteria. This was attained by inspection of proteome alterations of the outer membrane (OMP) in biofilm cells. Biofilms were formed in 6-well plates for 24h being after submitted to the presence of 0.9 mM BC and 6.0 ug/ml CIP, during 13 days. The obtained biofilm-cells were separated and the OMP extracted. Protein patterns were analysed by 2-DE and gels by the SameSpot software. Biofilm-proteome showed that *P. aeruginosa* adaptation to BC promoted the down-regulation of 36 OMP and the up-regulation of only one. OMP 2DE of *P. aeruginosa* adapted to CIP revealed the down-regulation of 29 OMP. Six OMPs were changed in common by both antimicrobials, revealing a possible similar stress response. Proteins identification is in progress. This study highlighted that there might be an OMP regulation when bacteria within biofilms are submitted to chemical adaptation. This particular response to the environment can be one of the causes of the well-known biofilm resistance phenotype. Acknowledgments:IBB-CEB, FCT(PTDC/SAUESA/64609/2006;SFRH/BD/31065/2006)