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Membranome analysis of benzalkonium chloride adapted Planktonic and Biofilm cells

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Adaptive resistance to antimicrobials has been widely reported through phenotypic characterization and proteomic analysis. Concerning biofilm adaptation, the response of biofilm-entrapped cells to chemical stress conditions is not yet well-studied. Membrane proteins play key roles in cell life cycles and are central in resistance mechanisms. The outer membrane (OMP) is an important compartment where metabolites circulate, constituting more than half of all known drug targets.

This work aimed to examine whether antimicrobial adaptation of planktonic and sessile *Pseudomonas aeruginosa* to benzalkonium chloride (BC) could induce a membranome alteration. This was inspected by proteome alterations of the OMP of planktonic and biofilm cells. Adaptation was attained by continuous exposure to increasing doses of the antimicrobial. Planktonic adaptive resistance was induced by subculturing *P. aeruginosa* in TSB supplemented with increasing concentrations of BC. Biofilms were formed for 24h being after submitted to maturation in the presence of BC. Biofilm and planktonic non- and adapted cells were collected and the OMP extracted. Protein patterns were analysed by 2D and gels by the SameSpot software.

Results showed that the protein contents of biofilm cells are differentiated from the suspended counterparts. The 2D pattern of adapted biofilm cells is also different from the adapted planktonic bacteria, revealing that the response to BC is different if cells are free floating or in sessile state.

This study highlighted that there might be different membranome regulation when bacteria are submitted to chemical adaptation. This particular response to the environment can be one of the causes of the well-known biofilm resistance phenotype.