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ADAPTIVE RESPONSE OF SINGLE AND BINARY BIOFILMS FORMED IN THE PRESENCE OF BENZALKONIUM CHLORIDE

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In actual situations bacteria can live nicely in hostile environments in part due to its ability to live in biofilms and to regulate gene expression as an adaptive response to a variety of stresses. This altered profile, compared to planktonic counterparts, as well as the interactions amongst the several strains existing within the biofilm seems to be responsible for the gradual loss of susceptibility to antimicrobials. The presence of biofilms

to biofilm-associated infections. The aim of this work was to examine whether exposure of *P.aeruginosa* and *E.coli* to an antimicrobial agent - benzalkoniun chloride (BC) - during single and binary biofilm formation could induce an adaptive response in bacteria and further evaluate the biofilm behaviour after aggression with the same agent. Biofilms were formed in microtiter plates for 5 days in presence of 0.9 mM of BC, being afterwards submitted to BC attack (1 mM). Biofilms were characterized by total mass, through crystal violet, metabolic activity, through XTT method, and number of cells. The data showed that both single and binary biofilms formed in the presence of BC have clearly more mass, less activity and slight less number of cells. These events could be sign of a different metabolic state of the cells entrapped in the biofilms previously exposed to the surfactant. The simultaneous growth of both species seems not prejudice or favour binary biofilm development. After BC attack, biofilms developed in the absence of the surfactant suffered mass and activity reduction and had fewer cells, probably due to the toxic effect of the product. Conversely, when the biofilms were developed in the presence of BC, the attack leads to biofilms with more mass and activity, being however the number of cells reduced. This discrepancy may be due to the loss of culturability of bacteria in solid media. These facts emphasise that BC is not effective against biofilms developed in its presence. It can be concluded that the presence of BC during single and binary sessile growth seems to induce an adaptive resistance of *P.aeruginosa* and *E.coli* biofilms. This biofilm features, resulting from an adverse situation, can play a significant role in the selection of resistant pathogenic bacteria and be on the basis of recalcitrant biofilms, contaminating medical surfaces and causing nosocomial infections.

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