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### **Isolation of novel bacteriophages for the control of *P. aeruginosa* biofilms**

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*Pseudomonas aeruginosa* is one of the most common gram-negative bacterium involve in nosocomial infections and, worryingly, it frequently shows a low antibiotic susceptibility. Additionally, *P. aeruginosa* has an inapt ability to adhere to surfaces and form virulent biofilms which are particularly difficult to eradicate. In this way, there is a need to develop new antimicrobial agents as an alternative to antibiotherapy and bacteriophages (phages) appear as one attractive solution for this problem. This work describes the isolation and characterization of novel phages and their application for planktonic and biofilm cell control. 17 different phages were isolated from hospital effluents and were tested against 35 antibiotic multi-resistant clinical strains provided by the São Marcos Hospital (Braga). Four of these phages, showing broad lytic spectra, were selected and their efficacy against planktonic cells was studied. Despite the superior lytic spectra exhibited by the selected phages, two of them were not efficient against their hosts and therefore were not chosen for biofilm control experiments. Meanwhile, the other 2 phages (phages phiIBB-PAA2 and phiIBB-PAP21), well capable of causing a great biomass reduction of planktonic cells, were tested against 24 hours old biofilms using different multiplicities of infection (MOI). Both phages caused an approximately 2 log reduction of biofilm-cells, already after 2 hours and the reduction was further enhanced after 6 hours of biofilm treatment, independently of the MOI. The main dissimilarity between the two phage-host systems concerns the biofilm-cell resistance to the phages. In brief, biofilm-cells of *P. aeruginosa* PAO1, the host of phage phiIBB-PAP21, acquired resistance to the phage and consequently an increase on the amount of cells after 24 hours of infection was observed. On the other hand, phage phiIBB-PAA2 continued to destroy the biofilms of *P. aeruginosa* ATCC 10145 and there were no evidences of cells becoming resistant, even after 24 hours of infection. This work shows that the two selected phages are well capable of controlling biofilms; however short treatment prevents the emergence of phage resistant hosts.

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