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EFFECT OF TEMPERATURE ON BACTERIOPHAGE INFECTION OF *PSEUDOMONAS FLUORECENS* CELLS VS. BIOFILMS

S. Sillankorva¹, I. Sutherland², R. Oliveira¹, J. Azeredo¹; ¹Universidade do Minho, Braga, PORTUGAL, ²Edinburgh University, Edinburgh, UNITED KINGDOM.

Bacteriophages can be seen as good alternative biofilm control agents due to their high specificity, efficacy against biocide resistant bacteria and because they are innocuous to the environment. This study was focused on bacteriophage control of *Pseudomonas fluorescens* biofilms. These biofilms are difficult to eradicate by the current chemical treatments and contribute to the spoilage of dairy industry products due to the proteolytic activity of some *Pseudomonas fluorescens* strains. The effect of infection temperature, as well as the influence of bacterial and biofilm growth temperature on bacteriophage infection, was investigated. *Pseudomonas fluorescens* biofilms and suspensions of this microorganism were grown and infected at 4°C, 26°C and 37°C. The rate of cell lysis after bacteriophage infection was evaluated by ATP release. This method proved to be suitable when dealing with biofilm infection, because the determination of CFU is difficult to perform. The results revealed that the optimum temperature of infection of planktonic cells and biofilms is 26°C. At this temperature, bacteriophage øs1, at a multiplicity of infection (MOI) of 0.5 infected both planktonic cells and biofilms grown at 26°C causing a biomass reduction of about 85% (in both cases). The rate of log cell reduction was about 1.2 times greater for planktonic cells than for biofilms and the rate of bacteriophage growth was greater for suspended cells. This indicates that bacterial cells in planktonic form are more sensible to bacteriophage infection than biofilm-like bacteria; nevertheless

biofilms were successfully eradicated in these conditions. When biofilms and planktonic cells were grown at 4°C and 37°C and infected at 26°C (the optimal temperature of phage infection) the rate of cell lysis and bacteriophage growth was greatly reduced. In this case, the rate of cell growth overcame the rate of bacteriophage infection, thus after 60 min of bacteriophage infection no biomass reduction was observed. When cells are grown at 4°C and 37°C, probably the specific receptors for bacteriophage binding are not expressed at the cell surface. This constitutes a drawback on bacteriophage biofilm control that requires further investigations