Disclosing the complexity involved in phage-biofilm interaction: the case study of a *Sep1virus* phage infecting *S. epidermidis*

Luís D. R. Melo, Graça Pinto, Fernando Oliveira, Angela França, Diana Vilas Boas, Carina Almeida, Sanna Sillankorva, Nuno Cerca, Joana Azeredo

CEB - Centre of Biological Engineering, Universidade do Minho, 4710-057 Braga, Portugal
Group: BIOFILM | Line: Health Biotechnology and Bioengineering

Staphylococcus epidermidis is a major causative agent of nosocomial infections, mainly associated with the use of indwelling devices, on which this bacterium forms structures known as biofilms. Due to biofilms high tolerance to antibiotics, virulent bacteriophages have been suggested as novel anti-biofilm therapeutic agents. In this study, we used the *S. epidermidis*-specific phage phiIBB-SEP1 (SEP1) [1] and evaluated its activity against biofilms. Despite its broad host spectrum and high activity against exponential phase cells, the same was not observed for cells encased in a biofilm structure. To understand the underlying factors impairing SEP1 inefficacy against biofilms, we tested this phage against distinct bacterial populations. Interestingly, SEP1 was able to infect late stationary-phase (dormant), persister and biofilm-released cells, suggesting that the inefficacy for biofilm control resulted from the biofilm structure. To demonstrate this hypothesis, SEP1 activity was tested against clusters of cells from scraped biofilms resulting in a 2 orders-of-magnitude reduction in the number of viable cells, after six hours of infection. Additionally, LIVE/DEAD staining allowed the observation that stationary-phase cells responded to phage addition, as determined by the increase in SYBR medium fluorescence intensity, which can be related with an increase on the cell metabolic activity.
These are promising results, since the rare feature presented by this phage of infecting cells with reduced metabolic activity allied with its ability to infect persister and biofilm-released cells, suggest its use as anti-biofilm agent when combined with enzymatic (dispersin B) or mechanic debridement (sharp).

References

[1] Melo, L. D. *et al.* Isolation and characterization of a new *Staphylococcus epidermidis* broad-spectrum bacteriophage. *J Gen Virol* **95**, 506-515, doi:10.1099/vir.0.060590-0, 2014.