Disinfection Food Safety VBNC cells Staphylococcus aureus

Bacteriophage treatment after chemical disinfection to tackle *S. aureus* VBNC cells adhered to food contact surfaces

Pilar Teixeira ^{1,2}, Inês Silva ^{1,2}, Joana Azeredo ^{1,2}, Luis D. R. Melo ^{1,2}

1. CEB - Centre of Biological Engineering, University of Minho, 4710-057 Braga, Portugal

2. LABBELS – Associate Laboratory, Braga/Guimarães, Portugal

Correspondence: pilar@ceb.uminho.pt

Food is often contaminated during processing and packaging through contact with equipment surfaces. To avoid it, an efficient sanitation is necessary. However, several studies have demonstrated that although sanitizers reduced the levels of foodborne pathogens they might induce cells of foodborne bacteria into a viable but non-culturable (VBNC) state. We already confirmed that sodium hypochlorite and peracetic acid induce VBNC state in *E. coli* and that those VBNC cells can be detected by flow cytometry. These VBNC cells can "resuscitate" and recover their virulence resulting in a serious risk to human health. Thus, it is urgent to establishing an efficient disinfection protocol to food contact surfaces.

The main aim of this work was to develop a targeted solution for controlling risks associated with *Staphylococcus aureus* VBNC cells in food processing surfaces. For that, clinical isolates of *S. aureus* were subjected to disinfection with sodium hypochlorite (SH) and the bacteriophage LM12 was assessed as an antimicrobial agent to target the remaining VBNC cells.

The results showed that to completely eliminate 1x10⁸ CFU/ml of *S. aureus* adhered to stainless steel surfaces it is necessary a concentration of 275 mg/l of SH with a contact time of 5 minutes. A 1 log reduction is observed to 200 mg/l of SH. An increase of contact time, as expected, leads to lower concentrations needed for reduction. When phage is used as the antimicrobial agent, at contact times of 30 and 60 minutes, it was observed that *S. aureus* presented susceptibility with more than 1 log reduction.

Despite some vulnerability of bacteriophage LM12 to high concentrations of SH, we are currently exploring the application of LM12 to tackle *S. aureus* VBNC cells after disinfection of stainless steel surfaces with sodium hypochlorite.