Phage therapy

Bacteriophage are commonly referred to as phage and are defined as viruses that infect bacteria (Doss et al. 2017). Phage were discovered independently by two different scientists: Frederick Twort in 1915 and again by Félix d´Herelle in 1917 (Salmond and Fineran 2015). Phage therapy was first attempted by d´Herelle to therapeutically treat humans. The use of the bacteriophage to treat bacterial infection initially yielded impressive results. However, many problems soon appeared, such as the lack of reproducibility and the impossibility of achieving quality controls (Haq et al. 2012). In addition, the discovery and use of many chemical antibiotics further decreased interest in phage therapy research in United States. In contrast, phage therapy continued in the Soviet Union, Eastern Europe, and France. From the decade of the 80s, due to the emergence of resistance to antibiotics, some research groups focused on phage therapy as an alternative to the use of these antibiotics. (Sulakvelidze, Alavidze, and Morris 2001).

The use of phages to treat infections has been studied in several fields, showing several advantages against the use of the antibiotic. The method to isolate phages is very fast, relatively simple and inexpensive. Resistance to phage develops about ten times slower than antibiotic resistance (Parasion et al. 2014).

The phages continue replicating in the presence of the host bacterium until an important decrease of the same one (Schmelcher and Loessner 2014) , this means that less quantities of phages could be used to carry out a treatment due to the ability to self-replicate. Moreover, most phage isolated have a relatively high level of specificity for their host. Therefore, it would have an advantage in the use of chronic diseases where the microbiota is affected by the use of antibiotics. Phage therapy is also suitable for the use in humans, since phage do not infect eukaryotic cells (Parasion et al. 2014) .

Recent investigation using animal models have explore phage treatment against a range of clinically significant pathogens. When challenged with gut-derived sepsis due to *P. aeruginosa,* oral administration of phage saved 66.7% of mice from mortality compared to 0% in the control group (Watanabe et al. 2007) . A single doses of phages used as prophylaxis in a Hamster model of *C. difficile*-induced ileocecitis was sufficient

prophylaxis against infection, saving 11 of 12 mice whereas control animals receiving *C. difficile* and clindamycin died within 96h (Nale et al. 2016) .

Human trials for phage therapy have taken place for almost a century at several institutes in Eastern Europe. The Eliava Institute of Bacteriophage and the Institute of Immunology and Experimental therapy in Wroclaw, Poland, have extensively used phage in preclinical and clinical treatment of common bacterial pathogens such as *S. aureus*, *E. coli*, *Streptococcus* spp., *P, aeruginosa*, *Proteus* spp., *S, dysenteriae*, *Salmonella* spp., and Enterococcus spp. (Soothill 1992). For example, six patients with antibiotic unresponsive diabetic foot ulcers, were treated by topical administration with *S. aureus* -specific phage. One application of this phage was enough for recovery in all individuals (Fish et al. 2016). In a 1938 clinical trial, 219 patients with bacterial dysentery were treated solely with a phage cocktail consisting of a variety of phage targeting *Shigella flexneri, Shigella Shiga, E. coli, Proteus* spp., *P. aeruginosa*, *Salmonella typhi*, *Salmonella paratyphi* A and B, *Staphylococcus* spp., *Streptococcus* spp. and *Enterococcus* spp.; cocktails were administered both orally and rectally. Overall 74% of the 219 patients showed improvement or were completely relieved of symptoms (Lin, Koskella, and Lin 2017; Kutateladze and Adamia 2008).

Currently, there are no phage therapy products approved for human use in the European Union or United States. However, in the food industry, there are several commercial phage preparations used for biocontrol of bacterial pathogens that are approved by the FDA under the classification of "generally considered as safe". Evidence suggests that phage biocontrol can be an effective method for improving food safety at numerous stages in meat production and processing, for example Atterbury and colleagues (Atterbury 2009),used phages to control *C. jejuni* contamination on the surface of chicken skin. After 24 hours, the treatment with phages resulted in a 1-1.3 log reduction in *C. jejuni*. In other work, Hungaro et al. (Hungaro et al. 2013), used a mixture which contain a phage with chemicals agents (dichloroisocyanurate, peroxyacetic acid, lactic acid) to treat S. enteritidis on chicken skin. A reduction of 1-log CFU / cm^2 was showed. Phages have been used to reduce bacterial contamination in fruits, vegetables, and dairy products. For example, Levenrentz et al. (Leverentz et al. 2001), studied the phage therapy against *Salmonella* on fresh fruit.

Other examples of phage therapy are included in the literature of the following authors. Phage applications in sheep (Bach et al. 2009); phage applications in cattle (Rozema et al. 2009); In the phage application in the swine yield (Wall et al. 2010) , showed the effects of *Salmonella*-phages use in pigs during transport and holding prior to slaughter; Phage applications in poultry include a lot of literature because it is the major reservoir for *Salmonella* and Campylobacter in the world, which is responsible for causing salmonellosis and campylobacteriosis in humans, an example of this literature is include in (Connerton et al. 2004; Higgins et al. 2005; Sillankorva, Neubauer, and Azeredo 2010)

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