

317. Biocontrol of *Pseudomonas syringae* pv. *actinidiae* in kiwi plants using bacteriophages

Rute Rego^{1,2}, Diana Priscila Pires², Rafaela Monte¹, Joana Azeredo², Conceição Santos³,
Luísa Moura^{1,4}

¹*Instituto Politécnico de Viana do Castelo, Escola Superior Agrária, 4990-706 Ponte de Lima, Portugal*

²*CEB - Centre of Biological Engineering, University of Minho, Campus de Gualtar, 4710-057 Braga, Portugal*

³*Department of Biology, Faculty of Sciences of University of Porto (FCUP), LAQV-REQUIMTE, FCUP, Porto, Portugal*

⁴*CISAS, Instituto Politécnico de Viana do Castelo, 4900-347, Viana do Castelo, Portugal*

Pseudomonas syringae pv. *actinidiae* (Psa) is the causal agent of bacterial canker in kiwifruit (*Actinidia deliciosa*), leading to severe symptoms in plants and consequent considerable production and financial losses. Current methods for controlling this disease rely on the use of copper-based products and, in countries outside the European Union, in the use of antibiotics. These products, besides being considered phytotoxic, can lead to the spread of resistance to both copper and antibiotics. With the increasing need to obtain safe and effective biocontrol strategies against this pathogen, this study focused on the isolation and characterization of (bacterio)phages for the control of Psa. A collection of Psa strains and phages were isolated from branches, buds, leaves, petals, sepals, and stamens from kiwifruit plants in orchards in the North of Portugal. Phages were isolated using the enrichment procedure with Psa strains CFBP 7286 and P84 as possible hosts, and the lytic spectra of 6 selected phages were tested against the Psa collection. Three phages displaying broader host ranges (between 90 to 97% of efficacy among Psa strains) were selected for further analysis. Phage stability was studied at different temperatures (-20° to 60°C), pH (1-13) and UV light, and it was observed that the three phages were stable between -20°C and 50°C, pH range of 3 to 11 and UV light at 366 nm up to 180 min. The morphological characterization of phages was done by Transmission Electron Microscopy and the genome sequencing of these phages is ongoing. In vitro efficacy assays showed that phage 177T was able to reduce the number of CFUs after 4h of inoculation and keeping the bacterial load at low levels for up to 24h post-infection with MOI=1. In vitro efficacy tests were also carried out on kiwi leaf discs, and it was possible to detect a reduction in the bacterial load of Psa.

Altogether, our results clearly highlight the potential of phages as promising biocontrol agents against Psa, that could be used in the future to replace copper compounds and antibiotics. This work was supported by the GesPSA Kiwi: GesPSA Kiwi-Ferramenta Operacional para gestão

sustentável do cancro bacteriano (Psa) da Actinídea-NORTE-01-0247-FEDER-033647, a project funded by Fundo Europeu de Desenvolvimento Regional (FEDER) through the Programa Operacional Regional do Norte (NORTE2020).