



Characterization of three newly- isolated, broad host range or polyvalent, *Xanthomonas* sp. effective bacteriophages.

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Introduction

For effective phage biocontrol of plant diseases, caused by more than one phytopathogenic bacteria, such as bacterial spot on pepper and tomato (Fig. 1), phage cocktails could be applied. Such cocktails, containing phages with different host range, as control strategy in fighting crop diseases, is a rapidly evolving field in agro sciences. As alternative to such cocktails, phage pesticides, containing polyvalent or broad host range phages, might be applied.

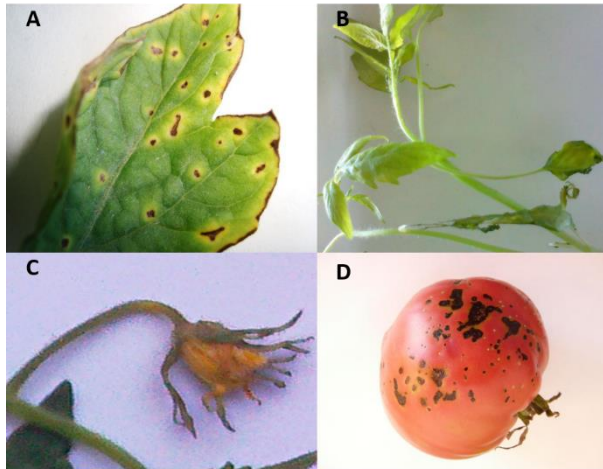


Fig. 1 Bacterial spot on tomato

Aim of the study

In this study we present, for the first time, the isolation and initial characterization of three polyvalent (broad host range) bacteriophages (SfXv124t/1, SfXv124t/2 and SfXv124t/3), effective against three phytopathogenic bacteria causing bacterial spot on tomato and pepper: *Xanthomonas vesicatoria*, *Xanthomonas euvesicatoria* and *Xanthomonas gardneri*.

Methods

Bacteriophage isolation was done by applying double agar overlay plaque assay (DAOPA) – Fig. 2.

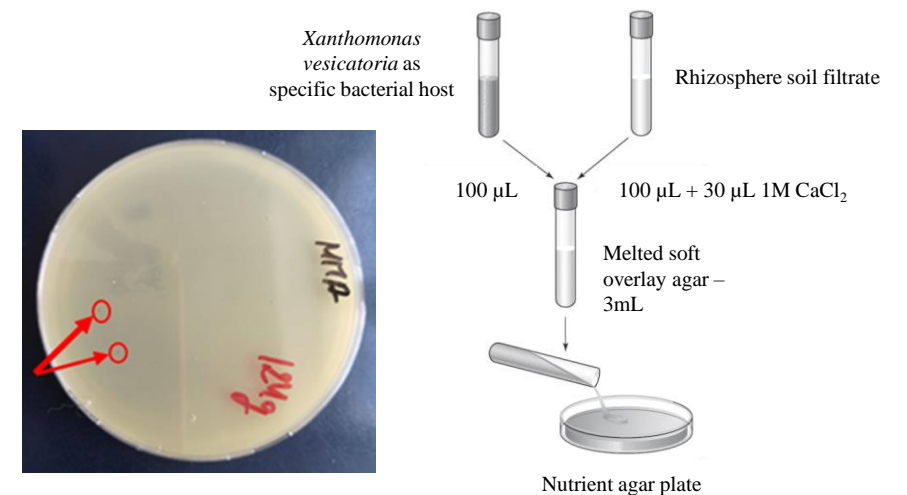


Fig. 2 DAOPA

Results

The initial characterization of the phage isolates includes: 1/ host range analysis of the three isolates, 2/ determination of the types of plaque morphology and measuring the plaques diameter, formed by the phages on solid agar plate by DAOPA and 3/ determination of the virions morphology by using TEM. Phage strains were isolated from tomato rhizosphere soil, using *Xanthomonas vesicatoria*, a strain isolated from tomato, as specific host. According to TEM and DAOPA results, there might be a *Siphoviridae* phage (SfXv124t/1) and a *Podoviridae* (SfXv124t/3) phage, among the isolates (Fig. 3 and 4).

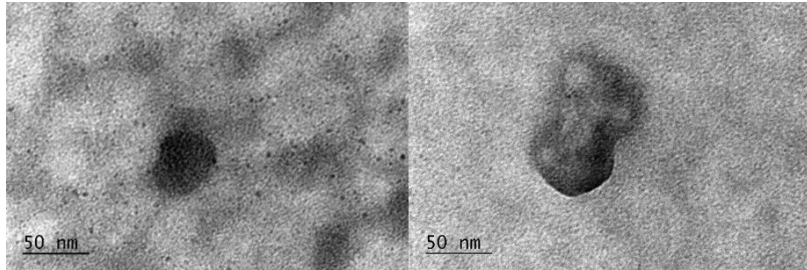


Fig. 3 TEM morphology of SfXv124t/1

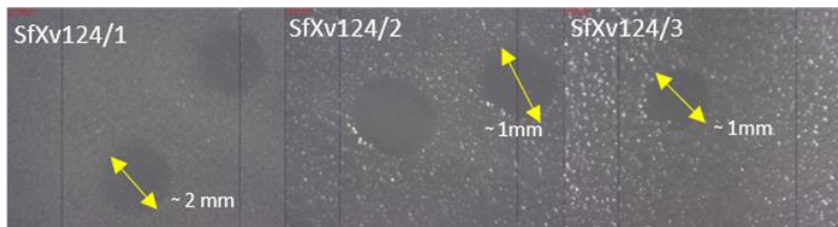


Fig. 4 Plaque morphology of three phages.

The phage host range determination was carried out using 23 phytopathogenic strains belonging to the genus *Xanthomonas* and *Pseudomonas*, as specific hosts. The results showed that phages SfXv124t/2 and SfXv124t/3 are capable to lyse two phytopathogenic bacteria: *X. euvesicatoria* and *X. vesicatoria* as opposed to SfXv124t/1 lysing three phytopathogenic bacteria: the two aforementioned and *X. gardneri*. None of the tested phages has lytic activity against *Pseudomonas syringae* pv. *tomato* strains (Fig. 5).

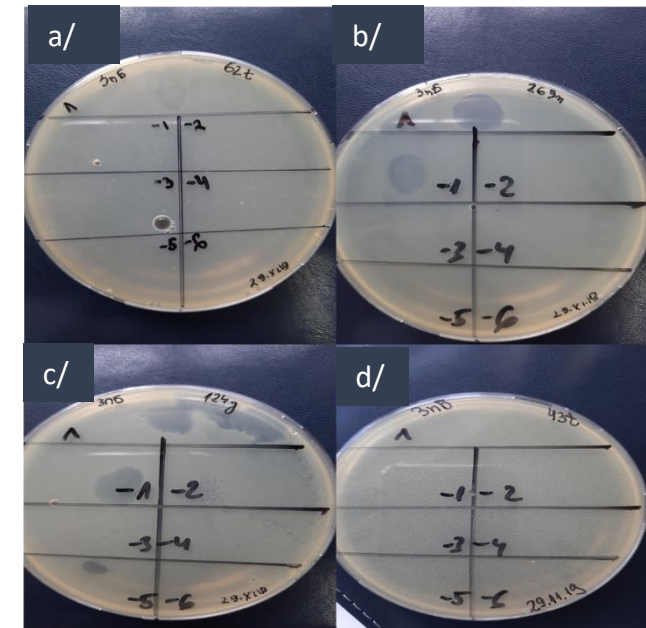


Fig. 5 Host range of the phage SfXv124t/1: lysing activity on a/ *Xanthomonas gardneri*, b/ *Xanthomonas euvesicatoria* and c/ *Xanthomonas vesicatoria*; d/ *Xanthomonas vesicatoria* resistant strain

Conclusion

The broad range of phages effectiveness showed that the newly isolated bacteriophages are polyvalent, hence - a good alternative to chemical pesticides as they are capable to destroy the all three causative agents of bacterial spot disease on tomato and pepper.

Acknowledgement

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