

## CHARACTERIZATION OF LYTIC PROPERTIES OF *ERWINIA AMYLOVORA* BACTERIOPHAGES

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*Erwinia amylovora*, as a highly harmful pathogen of fruit crops, has attracted the attention of researchers due to the need in effective means of controlling fire blight. An environmentally friendly tool to control fire blight is biological agents. Specific bacteriophages can serve as potential bioagents.

In the present study, bacteriophages were isolated from the soil samples on the territory of Belarus using *E. amylovora* 1/79Sm as an indicator bacterial culture (Germany, Spontaneous Sm-resistant mutant of 1/79, *Cotoneaster* sp., 1979): Hena1, Hena2 (Grodno Region), Roscha1, Fleur, Stepyanka (Minsk Region). Using a double agar overlay method a variety of phage negative colony morphology was observed: for Hena1 point negative colonies, for Hena2, Roscha1 - from point to 2 mm in diameter, with uneven edges, for Fleur, Stepyanka - 3-5 mm in diameter.

During the study, host range analysis was conducted using the method of spotting phage lysates (10 µl volume, phage titer  $10^7$  PFU/ml) on the bacterial culture lawn. Investigated bacterial strains were *E. amylovora* E2, L3-2, L3-6, E3, E4, *Pantoea agglomerans* 194, 197, 198, 208, 216, 219, *Pseudomonas syringae* pv. *syringae* 12.6, 14.5 (1), 19.10. Among the studied bacteriophages, the lytic profile was the narrowest for Hena1 (two sensitive *E. amylovora* strains). The largest host range was shown for Hena2 and Roscha1, for which clear lysis zones were observed on all the presented bacterial strains of *E. amylovora* and strain *P. agglomerans* 216. Due to indication of turbid lysis zones of studied bacterial strains when applying phages Fleur, Stepyanka, as well as Hena2, Roscha1 on certain *P. syringae* pv. *syringae* and *P. agglomerans* bacterial strains, further testing of positive results of sensitivity studies using Gracia method is required.

For host bacterial strain *E. amylovora* 1/79Sm and phages, the bactericidal action of phage lysates of various titers was investigated. In the study 8 tubes with 5 ml of LB broth were used, 0.1 ml of a culture of bacteria with the optical density (OD) of 1 at 600 nm was added to each tube. In 7 tubes, 0.1 ml of phage lysates of various titers (from  $10^2$  to  $10^8$  PFU/ml) was added. After overnight incubation of the tubes, the OD of the resulting suspension was measured at 600 nm. For phage Hena1 when applying a phage lysate of  $10^8$  PFU/ml titer, on average 1.9 times decrease in OD value in comparison with the control value was shown. For phage Hena2 the OD value decreased to  $0.02 \pm 0.015$ , which is on average 10 times less than the control value. For phage Roscha1 the OD value decreased to  $0.04 \pm 0.015$  and on average 5.4 times. The extent of bacterial culture lysis was proportional to the titer of three bacteriophages. For Fleur and Stepyanka, a proportional dependence of the extent of the bacterial culture lysis on the bacteriophage titer was not observed; on average 1.5-2 times and 1.5-1.65 times reduction of OD respectively.

