Construction and characterization of the deletion mutant of *slyA* gene in phytopathogenic bacteria *Erwinia amylovora*

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Key message. The regulon of global transcriptional regulator SlyA in bacteria *Erwinia amylovora* was examined. For that, the Δ slyA mutant was constructed. The mutant showed a decreased level of amylovoran production and polymyxin B resistance.

Keywords: *Erwinia amylovora*, SlyA, transcription regulation, deletion mutant, virulence factors

SlyA is a transcriptional regulator that is common for *Enterobacteriaceae* family members, but also found in other bacteria and archaea. The protein has been shown to be involved in the virulence factors regulation in some pathogens, but its role in *Erwinia amylovora* was unknown.

In this study we exam the regulon of global transcriptional regulator SlyA in bacteria *Erwinia amylovora*. The factor belongs to the MarR (multiple antibiotic resistance regulators) family that regulate a wide variety of cellular processes, including resistance to multiple antibiotics, organic solvents, oxidative-stress agents and some aromatic compounds [2].

For examination of the role of SlyA we have constructed the deletion mutant of *slyA* gene, utilizing one-step inactivation approach by Datsenko [1]. Having predicted the regulon with the bioinformatics tool we took the most expressive virulence factors to check for any changes. For swarming motility analysis, the assay with semi-solid agar was used, but despite the presence of *fliL* operon in predicted SlyA regulon no significant difference was detected. The test for cellulose production (as an auxiliary biofilm polysaccharide) has not shown the difference too. Also, the ability to amylovoran (main biofilm polysaccharide) production was measured. The mutant exhibited decreased (about 58,6%) level of exopolysaccharide secretion in comparison with wild type, whereas the complementation test showed intermediate values (about 89,8% of wild type results). The test for survivability in presence of antimicrobial agent polymyxin B also was conducted. There mutant showed the decreased survival rate. These results allow us to suggest that SlyA plays an important role in the pathogenicity of *Erwinia amylovora*.

The further research will be focused on biofilm formation study, survival ability under different conditions such as hyperosmotic stress, ROS-mediated stress, low pH and other unfavorable conditions that might be faced by bacteria in the host plant.

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