Pseudomonas spp. strains isolated from grapevine leaves modulate the inhibitory activity of the biocontrol agent Lysobacter capsici AZ78

Francesca Brescia^{1,2}, Lisa Ioriatti¹, Oscar Giovannini¹, Gerardo Puopolo¹, Ilaria Pertot^{1,3}

¹Department of Sustainable Ecosystems & Bioresources, Research and Innovation Centre, Fondazione Edmund Mach (FEM), San Michele all'Adige, Italy; ²Department of Agricultural, Food, Environmental and Animal Sciences, University of Udine, Udine, Italy; ³Center Agriculture, Food, Environment, University of Trento, San Michele all'Adige, Italy Email: francesca.brescia@guests.fmach.it

Abstract (300 word limit)*

Statement of the Problem: Bacterial biocontrol agents can improve plant heath with various mechanisms. The bacterial genus *Lysobacter* includes different species producing compounds and lytic enzymes active against phytopathogenic microorganisms and therefore can be a source of new biocontrol agents. In particular, *L. capsici* AZ78 (AZ78), isolated from tobacco rhizosphere, effectively controls the causal agent of grapevine downy mildew *(Plasmopara viticola)*, thanks to the production of antibiotics.

Since bacterial communities might modulate the antibiotic production of biocontrol agents, the aim of this research was to understand if grapevine phyllosphere bacteria could affect the antibiotic production of AZ78. To test the hypothesis we used a simplified model system with a culturable phytopathogenic oomycete (*Pythium ultimum*).

Methodology & Theoretical Orientation: 47 bacterial strains were isolated from leaves of *Vitis vinifera* L. cv. Pinot gris and Goldtraminer, identified by 16S rDNA phylogenetic analysis and their impact on AZ78's inhibitory activity was assessed *in vitro* according to the experimental design described in Figure 1.

Findings: Most of the Gram-negative bacterial isolates were γ -Proteobacteria, while the Gram-positive isolates belonged to Actinobacteria and Firmicutes. None of the isolated strains showed an inhibitory activity against *P. ultimum.* Interestingly, most of them promoted AZ78 inhibitory activity. In particular *Pseudomonas* sp. L35 increased AZ78 inhibitory activity of the 29.6 \pm 0.95%, this can be related to a change in AZ78 gene expression triggered by the presence of the strain.

Conclusion & Significance: The interaction with the natural microbiota is an important factor to be considered

in evaluating biocontrol agents efficacy, because their inhibitory activity can be affected by the microbiota itself. To gain a full picture, additional studies are necessary, taking into account the plant response, as well as considering the variation in AZ78 gene expression.

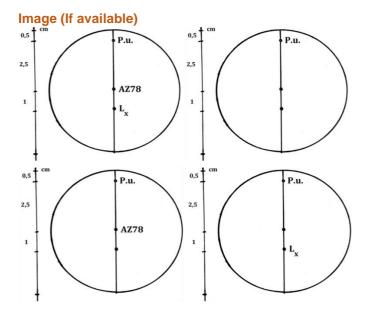


Figure 1: Evaluation of the impact of the 47 bacterial isolates (L_X) on the in vitro inhibitory activity of AZ78. The bacterial isolates and AZ78 were coinoculated on Luria Bertani Agar (LBA) at 1 cm of distance. After 48 h incubation at 25°C, 5 mm plugs of *Pythium ultimum* (P.u.) were inoculated at 2.5 cm from AZ78. After seven days incubation at 25°C, the growth area of P.u. was measured.

Recent Publications (if any-minimum 5)

- 1. De Boer W (2017) Upscaling of fungal-bacterial interactions: from the lab to the field. Current Opinion in Microbiology 37:35-41.
- 2. Panthee S, Hamamoto H, Paudel A, Sekimizu K (2016) *Lysobacter* species: a potential source of novel antibiotics. Archives of Microbiology 198:839-845.
- 3. Puopolo G, Cimmino A, Palmieri MC, Giovannini O, Evidente A, Pertot I. (2014) *Lysobacter capsici* AZ78 produces cyclo(L-Pro-L-Tyr), a 2,5-diketopiperazine with toxic activity against sporangia of *Phytophthora infestans* and *Plasmopara viticola*. Journal of Applied Microbiology 117:1168-1180.
- 4. Puopolo G, Tomada S, Pertot I (2018). The impact of the omics era on the knowledge and use of *Lysobacter* species to control phytopathogenic micro-organisms. Journal of Applied Microbiology 124:15-27.
- 5. Tyc O, de Jager VCL, van den Berg M, Gerards S, Janssens TKS, Zaagman N, Kai M, Svatos A, Zweers H, Hordijk C, Besselink H, de Boer W, Garbeva P (2017) Exploring bacterial interspecific interactions for discovery of novel antimicrobial compounds. Microbial Biotechnology 10:910-925.
- 6. Xie Y, Wright S, Shen Y, Du L, (2012) Bioactive natural products from Lysobacter. Natural Product Reports 29:1277-1287.



Image:

Biography (150 word limit)*

Francesca Brescia was born in 1991 in Italy. In 2013 she obtained her Bachelor's degree in Biology at the University of Torino. She decided to continue her studies at the University of Pavia, where in 2015 she obtained her Master's degree in Experimental and Applied Biology and, in November 2015, she gained a nine-months research fellowship about the study of the fungal community associated to different *Rubbus* species. In 2016 she started the doctoral course at the PhD school "Agricultural Science and Biotechnology" of the University of Udine at the Edmund Mach Foundation of San Michele all'Adige (TN) concerning the interactions between plant-associated bacteria and biocontrol agents in different nutrient conditions. From August 2018 to February 2019 she was a visiting PhD student at the Technical University of Vienna (Austria), where she carried out a part of her PhD project studying the compounds produced during microbial interactions.

Notes/Comments: