The effect of *Pseudomonas* sp. RU47 and phosphorus fertilization on gene abundances and activities of phosphomonoesterase in the rhizosphere of tomato

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INTRODUCTION

Low availability of phosphorus (P) in soils might be compensated by inoculation with PGPB that produce extracellular enzymes, such as acid and alkaline phosphomonoesterases.

Hypotheses

- Acid and alkaline phosphomonoesterase activities and phosphatase-encoding gene abundances are expected to be higher in rhizosphere than in bulk soil.
- Microbial inoculation with *Pseudomonas* sp. RU47 and P fertilization positively affects acid and alkaline phosphomonoesterase activities and gene abundances.

MATERIALS AND METHODS

Experimental Design

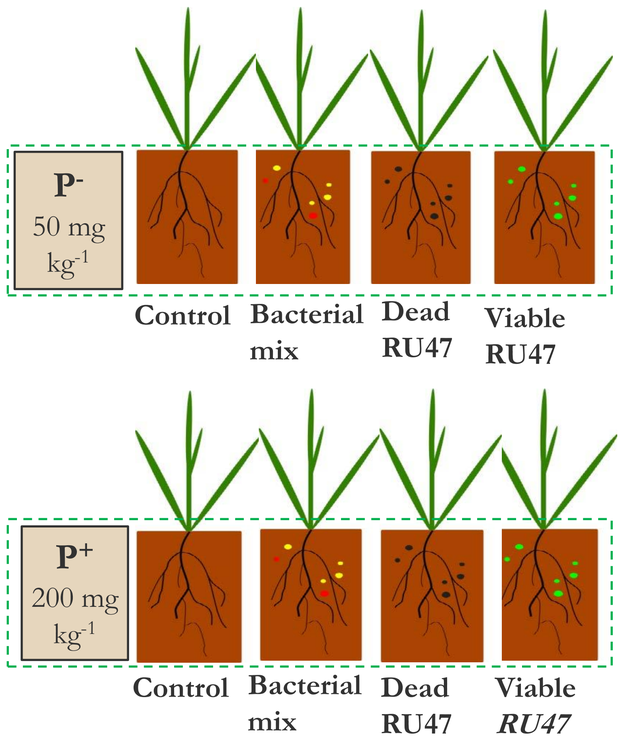
Plant Tomato (*Solanum lycopersicum* L. var. Mobil)
Duration 50 days
Conditions

- rhizoboxes (2.08 L) Luvisol (1) : sand (1)
- N-, K-, Mg- and Ca- fertilization
- greenhouse (ø 20.1 °C, ø 52.9 % humidity)

Replicates 5 (Σ 40 rhizoboxes)

Treatments

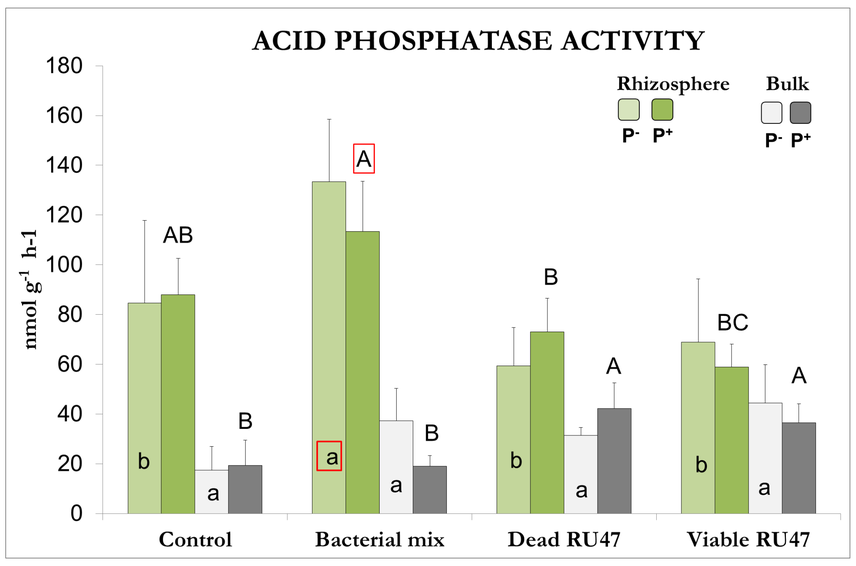
- **P fertilization**
- **PGPB inoculation**
  Bacterial mix unselectively cultivated microorganisms from soil
  Dead RU47 heat killed *Pseudomonas* sp. RU47 cells
  Viable RU47 living *Pseudomonas* sp. RU47 cells

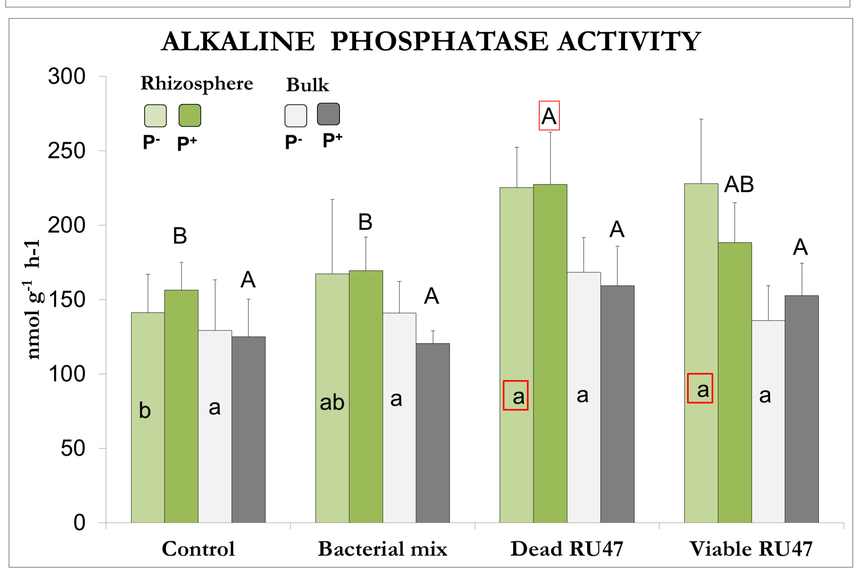


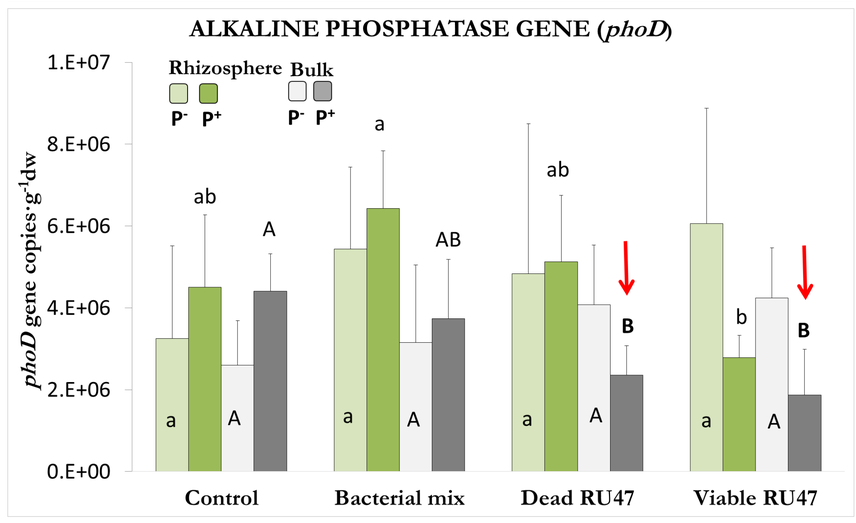
Analyses

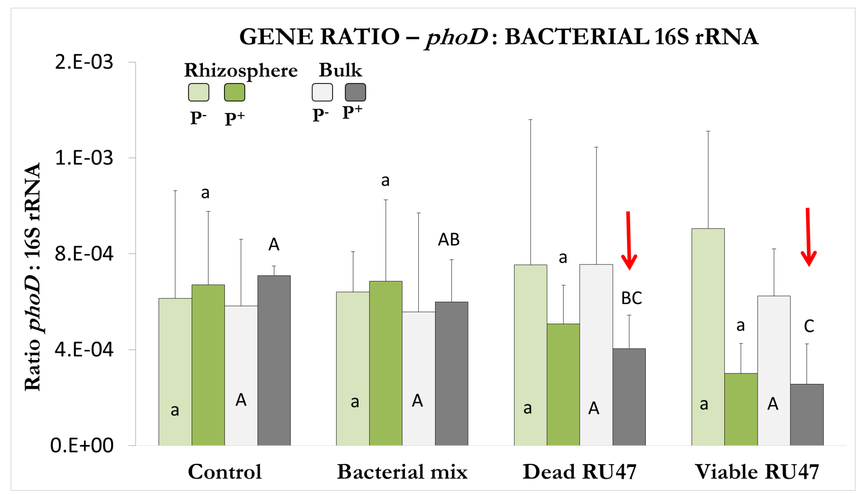
Enzyme assay (MUF) Marx et al. 2001
Quantitative PCR (16S, *phoD, phoN,* and *appA*) Bergkemper et al. 2016

RESULTS









- Higher abundance of RU47 was detected in **P+** than in the **P-** treatment 50 DAS (data not shown).
- Acid and alkaline phosphomonoesterases showed higher activities in the rhizosphere than in the bulk soil ($p < 0.05$) in all treatments.
- Dead RU47 increases significantly ($p<0.5$) alkaline phosphomonoesterase activity only in the rhizosphere probably due to nutrient addition from dead cells which stimulated enzyme production by endogenous microorganisms.
- In general, quantity of *phoD* was higher in rhizosphere than in bulk soil ($p < 0.05$).
- P fertilization together with RU 47 inoculation reduced the abundance of bacteria carrying the *phoD* gene in the rhizosphere and bulk soil.
- Relative abundance of *phoD* gene to bacterial 16S rRNA was significantly decreased in bulk soil of RU47 inoculated rhizoboxes under P fertilized rhizoboxes ($p < 0.05$).

CONCLUSION and OUTLOOK

P availability in soil might influence RU47's efficiency in P mineralization. The addition of dead or viable RU47 cells increased phosphatase activity in the rhizosphere. While the addition of dead RU47 cells might stimulate indigenous microbial activity (priming effect), increased PA by the addition of viable RU47 cells might cause by RU47 or interacting indigenous soil microbes. Since RU47 is mainly producing acid phosphatases in pure culture, we will extend our functional gene analyses to *phoN* and *appA* (acid phosphatases).

References: Bergkemper, F., Kublik, S., Lang, F., Krüger, J., Vestergaard, G., Schloter, M., & Schulz, S. 2016. Novel oligonucleotide primers reveal a high diversity of microbes which drive phosphorous turnover in soil. *Journal of Microbiological Methods*, 125, 91–97.
Marx, M.C., Wood, M., Jarvis, S.C. 2001. A microplate fluorimetric assay for the study of enzyme diversity in soils. *Soil Biol Biochem,* 33, 1633–40.

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