Decoding the sugar beet seed and rhizosphere microbiome for biocontrol of *Rhizoctonia solani*

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Introduction

Crop breeding aims to attain plants with less susceptibility to pathogens, higher productivity or better adaptation to target field environments. However, our knowledge of the connection between plant breeding and microbiome assemblage is limited. Here, we analysed the structure of the bacterial communities associated with *Rhizoctonia solani* tolerant and susceptible (Figure 1) sugar beet cultivars.

Figure 1: *R. solani* infested sugar beet plants.

1) Diversity

16S rRNA gene amplicon reads from seed and rhizosphere from four different cultivars with respect to the *R. solani* susceptibility (partially resistant, susceptible) were analysed. A comparison of the seed and rhizosphere microbial assemblage revealed a clear specification of the microbiomes, i.e. structural and phylogenetic diversification of each microhabitat (Figure 2). Shannon diversity was unexpectedly high for seeds (8.8) and highest for rhizosphere from plants cultivated in soil (13.4).

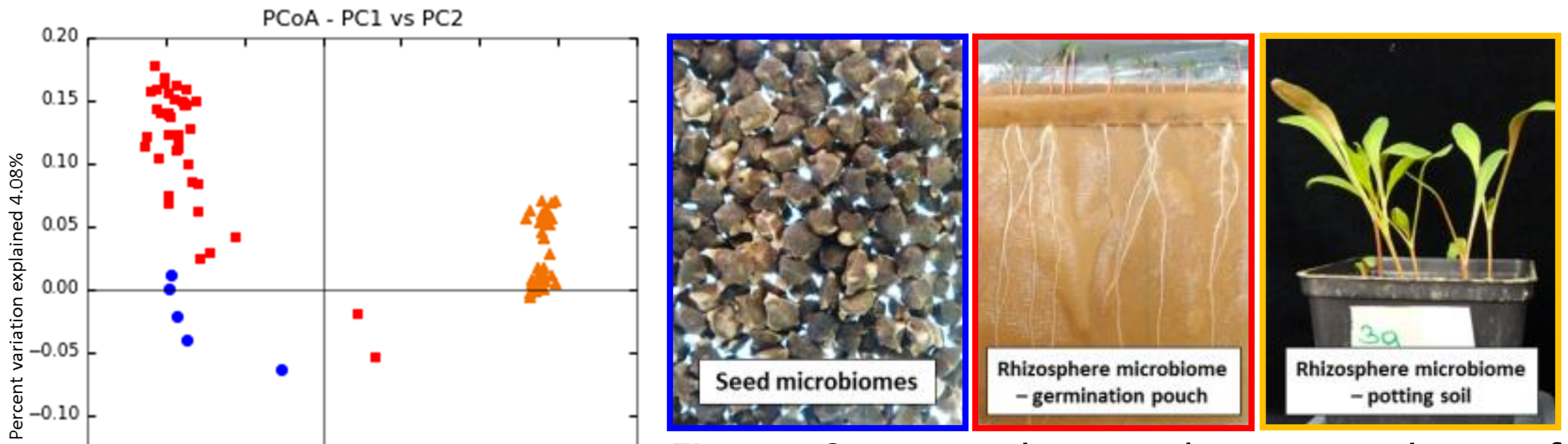

Figure 2: Principle coordinates analysis of unweighted UniFrac distances generated from the 16S rRNA sequence data obtained from the seed (blue) and rhizosphere (pouch-red, soil-orange) of the sugar beet plants.

2) Community structure

At phylum level, *Gammaproteobacteria* dominated in seeds (72.5%) and *Crenarchaeota* (34.5%) were found exclusively in the rhizosphere (Figure 3). Moreover, unweighted UniFrac-based PCoA plots of the rhizosphere microbiome showed that tolerant and susceptible cultivars formed separate clusters (P<0.001) (Figure 4).

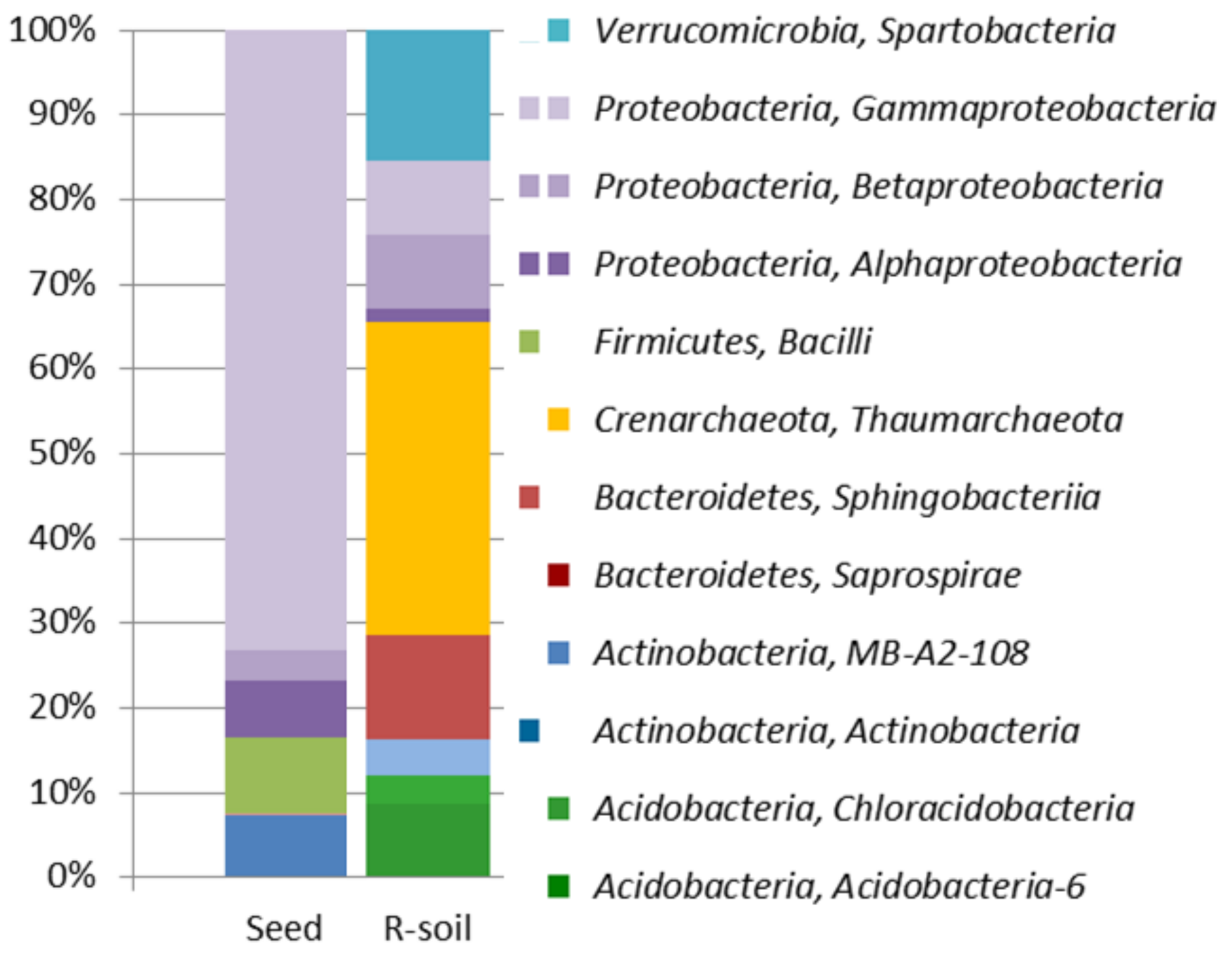

Figure 3: Taxonomic composition at class level of the seed- and rhizosphere-associated bacterial community.

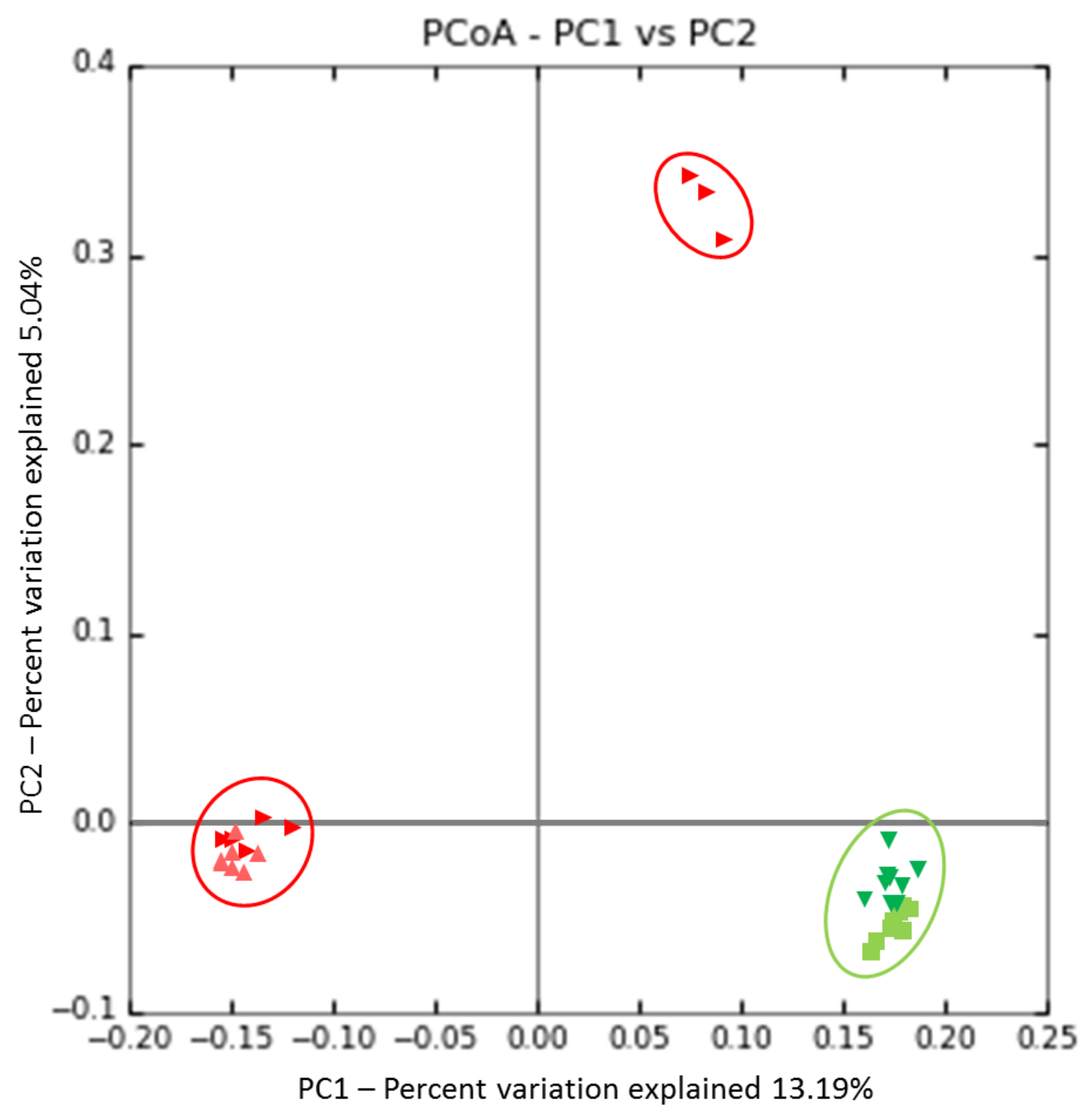

Figure 4: Principle coordinates analysis of unweighted UniFrac distances generated from 16S rRNA sequence data from the rhizosphere of the partially resistant (green) and susceptible (red) sugar beet cultivars.

3) Resistance microbiome

The results indicated that the specificity of associated microbiomes is higher with respect of the overall capacity to encounter *R. solani* than the cultivar specificity. Particularly in the rhizosphere, additional OTUs only detected in *R. solani*-tolerant cultivars were revealed by comparative analysis. Out of 2,873 OTUs in the rhizosphere assigned to *Pseudomonas*, 292 OTUs (4,415 reads) and 167 OTUs (3,676 reads) clustered together for the partially resistant and susceptible *R. solani* effect, respectively (Figure 5).

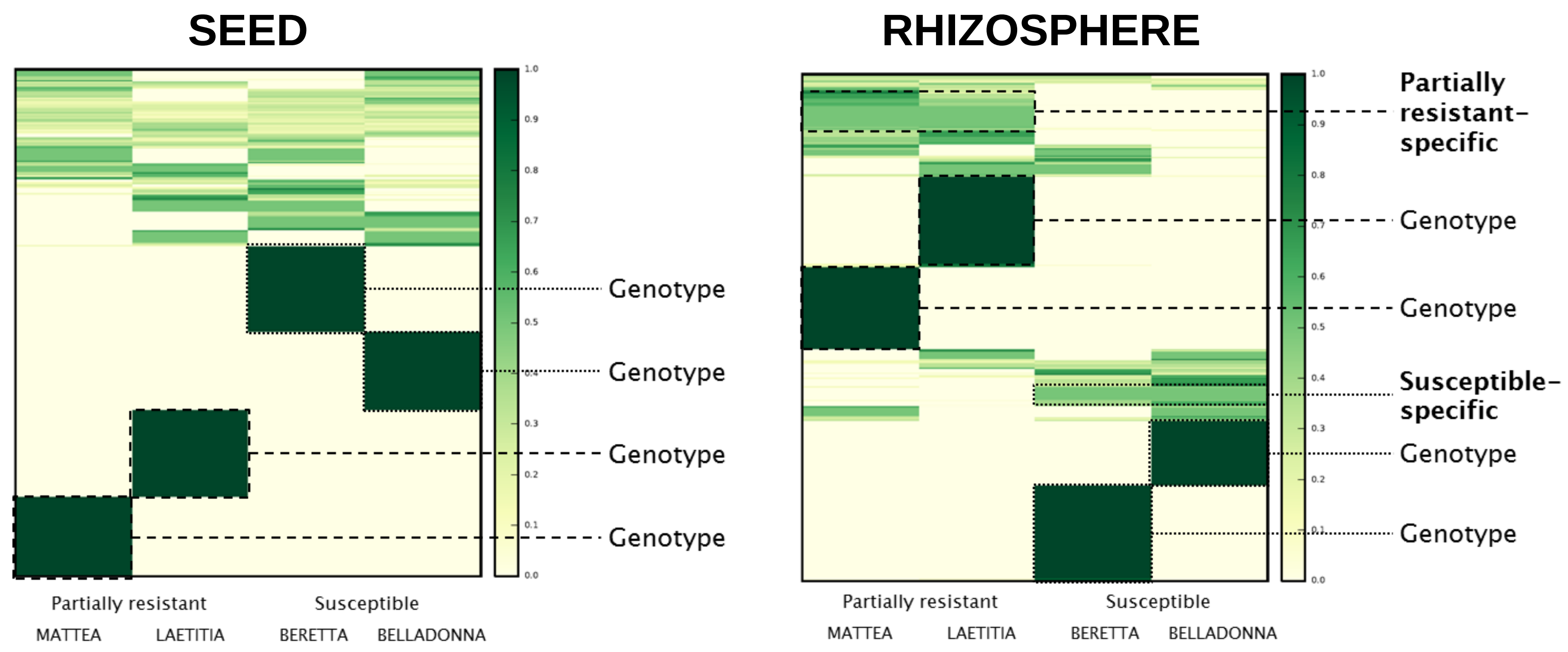

Figure 5: Abundance heatmap of significantly different key OTUs assigned to the genus *Pseudomonas*. The *R. solani* and genotype effect was indicated by dashed (partially resistant) and dotted (susceptible) lines. Each line represents a *Pseudomonas*-specific OTU.

4) Antagonistic genotype

An antagonistic *Pseudomonas* genotype was identified to be specific for all *Rhizoctonia*-tolerant cultivars, indicating a naturally occurring mechanism in acquiring beneficial microbes. The endophyte *P. poae* RE*1-1-14 was isolated as a representative strain of that genotype and compared to other phylogenetically related *Pseudomonas* strains. For example, two unlinked gene clusters consisted of three non-ribosomal peptide synthetase biosynthesis genes for poaeamide (Figure 6) were found to be specific and involved in biocontrol activity [1].

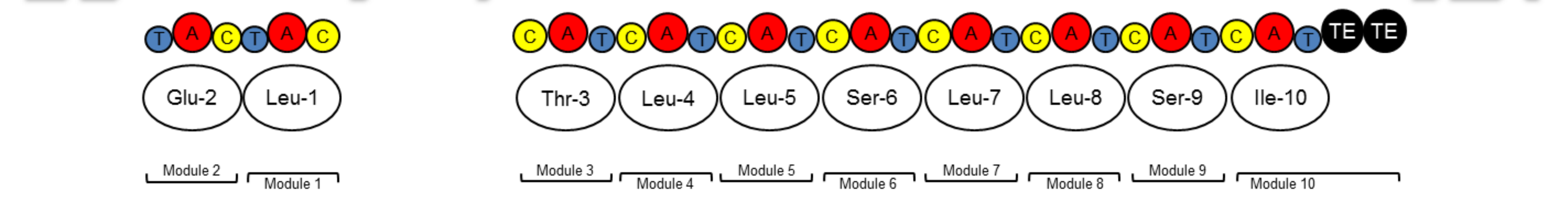

Figure 6: Organization of the poaeamide gene cluster (poaA, poaB, and poaC; black), LuxR family transcriptional regulators (green), macrolide-specific ABC-type efflux carrier (white) and hypothetical proteins (gray). (C – condensation, A – adenylation, T – thiolation, TE – thioesterification and the amino acids).

Conclusion

» Overall results reveal the impact of crop breeding on the plant-associated microbiome and implies a contribution to integrated management strategies in modern sustainable agriculture. «

Reference: [1] Zachow et al. 2015 MPMI 28(7): 800-810.

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