

Supplementary Methods

Isolation and characterization of PSB

The relative abundance of PSB was assessed by suspending 0.5 g of fresh soil from each sample in 49.5 ml of sterile water and shaking the suspension for about 2 h. Serial dilutions were prepared and tested to determine a suitable concentration of colony forming units (CFUs) for plating. The cells were grown on Pikovskaya's agar medium (PVK; Pikovskaya, 1948) at 20 °C for eight days. The PVK medium was composed of: 10 g glucose, 5 g hydroxyapatite, 0.5 g (NH₄)₂SO₄, 0.2 g NaCl, 0.1 g MgSO₄ * 7 H₂O, 0.2 g KCl, 0.5 g yeast extract, 0.002 g MnSO₄ * H₂O, 0.002 g FeSO₄ * 7 H₂O, and 15 g agar-agar in 1 L distilled water (Pikovskaya, 1948). The pH of the solution was adjusted to 7.0. If a bacterial colony dissolved hydroxyapatite present in the medium, a halo (clear zone) became visible in the otherwise milky medium. The relative abundance of PSB was calculated as the percentage of colonies that were formed by PSB over the total bacterial colonies formed. The PSB CFUs were collected and aseptically transferred into buffering solutions for sequencing analyses. Genomic DNA of bacterial colonies was extracted using the NucleoMag® Tissue kit on a KingFisher platform (Thermo Fisher Scientific, Massachusetts, USA) and diluted 100-fold with nuclease-free water. The 16S rRNA gene fragment covering variable regions V5-V8 were amplified using primers 799F and 1391R (Chelius and Triplett, 2001; Walker and Pace, 2007). PCR products were purified using the NucleoMag® 96 PCR cleanup kit and Sanger sequenced (GATC Biotech) using primer 799F.

Sequence data from the isolated cultures were processed using Geneious (Biomatters, New Zealand). Sequence similarity searches were conducted against the nr/nt nucleotide database at NCBI and NCBI RefSeq Targeted Loci Project (<https://www.ncbi.nlm.nih.gov/refseq/targetedloci/>). The partial 16S rRNA sequences were deposited in GenBank (<https://www.ncbi.nlm.nih.gov/genbank/>) under the accession numbers MW806976-MW807204. The best 20 hits were selected. Sequences with a 98% cut-off similarity were clustered into operational taxonomic units (OTUs). To this end, the name of the lowest common rank in the taxonomy was chosen. Only the non-redundant top hits were selected for taxonomic annotation. For this purpose, each 16S rRNA gene sequence was compared with all other sequences and sequences sharing

identity above 98% identity were assigned to one taxon. Names for OTU assignment were used as indicated in the NCBI database, although recently so-called environmental bacteria of the genus *Burkholderia* have been transferred to the genus *Paraburkholderia* and three species of the genera *Burkholderia* have been transferred to the new genus *Caballeronia* gen. nov. which represents a distinctive clade in phylogenetic trees (Dobritsa and Samadpour, 2016). Major phylogenetic changes were detected at the order and genus levels.

The impact of site and girdling treatment on the number of isolated PSB was tested per sampling time point by robust 2-way ANOVAs on trimmed means using the *t2way* command of the “WRS2” package (Mair and Wilcox, 2019). The impact of girdling was further tested by pairwise comparisons between the girdled and the respective control plots individually per sampling point at each site by robust 1-way ANOVAs on trimmed means using the *t1way* command of the “WRS2” package.

Relative abundance and taxonomy of PSB isolates

The relative numbers of isolated PSB did not differ significantly between the two sites (**Supplementary Table 1**). Girdling had no effect on the relative number of PSB cultures at T1 (**Supplementary Figure 3A**), while a lower proportion of PSB were isolated from the girdled plots at T2 (p-value < 0.01). The decrease was stronger at the LUE site (p-value = 0.04).

At the BBR site, 13 different genera were isolated, many of which were detected only at the girdled samples (**Supplementary Figure 3B**). Most isolates belonged to the genus *Pseudomonas*. The relative number of *Pseudomonas* isolates appeared to decrease at the samples obtained from the girdled plots, while others (i.e. *Paraburkholderia*, *Erwinia*) increased. In contrast, only 4 genera were isolated from the LUE samples and almost all cultures were identified as *Paraburkholderia* (**Supplementary Figure 3B**). Species *Pseudomonas* and *Serratia* appeared only in the LUE samples from the girdled plots and *Caballeronia* only in the control plots.

References

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