



## Positive effects of plant association on rhizosphere microbial communities depend on plant species involved and soil nitrogen level



Barbara Pivato<sup>\*</sup>, David Bru, Hugues Busset, Florence Deau, Annick Matejicek, Laurent Philippot, Delphine Moreau

Agroécologie, AgroSup Dijon, INRA, Univ. Bourgogne Franche-Comté, F-21000 Dijon, France

### ARTICLE INFO

#### Article history:

Received 26 January 2017

Received in revised form

7 June 2017

Accepted 16 June 2017

Available online 4 July 2017

#### Keywords:

Plant-microbe interaction

Rhizosphere

Plant association

Nitrogen

### ABSTRACT

How plant species associations affect the rhizosphere microbiota, in comparison to each plant species, has been overlooked. Here we investigated how bi-species plant associations affect total bacterial and N-cycling microbial communities (nitrate reducers and ammonia-oxidizers), as an example of functional communities, at low and high soil N levels. At low N level, the total bacterial abundance in the rhizosphere of the bi-species associations was significantly higher than that of either plant on its own for half of the bi-species associations. For the other associations, the bacterial abundance was not different from the corresponding mono-species cultures. The abundance of several N-cycling guilds was also enhanced by some of the bi-species associations with increases of up to 125% and no negative effects were recorded, which highlights the importance of plant diversity in agro-ecosystems.

© 2017 Elsevier Ltd. All rights reserved.

Understanding how plants are shaping microbial communities and their activities is a key for harnessing the rhizosphere microbiome and enhancing the beneficial plant-microbe interactions in agroecosystems (De Vrieze, 2015; Lareen et al., 2016). Different root exudates, such as the phytohormone salicylic acid (Lebeis et al., 2015) or the nitrification inhibitor brachialactone (Subbarao et al., 2009), and also other root traits (Cantarel et al., 2015), or N-related plant traits (Moreau et al., 2015) modulate the structure or the activity of the rhizosphere microbiome.

The vast majority of these studies focused on a single plant species (review in Bakker et al., 2013; Philippot et al., 2013). However, recent studies found plant species richness and community composition to be also important drivers of total microbial communities (e.g. microbial biomass, activity, structure) and functional communities as those involved in soil N-cycling (McGill et al., 2010; Eisenhauer et al., 2013; Le Roux et al., 2013; Strecker et al., 2015, 2016). Nevertheless, how these plant species associations affect soil microbial community structure and function, in comparison to each individual plant species, remains unclear. For example, are the abundance and structure of microbial communities in plant species associations similar to those of either individual plant species? Or can interactions between plant species

increase the abundance of rhizosphere microbial communities in comparison to either plant species on its own?

Here, we analyzed the responses of soil microbial communities to mono- and bi-species plant associations, considering four monocotyledonous species, on both total and N-cycling microbial communities at low and high soil N levels. We focused on total bacteria as well as on the nitrate reducing and ammonia-oxidizing communities due to their role in the availability of nitrate and ammonium, which are major plant nutrients.

Our hypothesis was that bi-species associations lead to a different rhizosphere condition than mono-species associations, which can result in an increased abundance of microbial communities (positive effect) than each correspondent mono-species.

A greenhouse experiment was conducted using three weed species (*Alopecurus myosuroides*, *Apera spica-venti* and *Vulpia myuros*) and one crop species (*Triticum aestivum* cv. *Caphorn*). Four plants were cultivated in each pot, in either mono-species cultures (i.e. four plants of the same species) or bi-species cultures (i.e. two plants of two species), with three replicate pots per treatment. Pots were previously filled with a Cambisol collected in a grassland from a Long Term Observatory near Lusignan site [France: 46°25'12.91"N; 0°07'29.35"E] (soil properties are presented in Supplementary information, Table 1). The pots were filled with clay beads and soil and kept at 70% of the water holding capacity for a week before the pre-germinated seedling transplantation. All

<sup>\*</sup> Corresponding author.

E-mail address: [barbara.pivato@inra.fr](mailto:barbara.pivato@inra.fr) (B. Pivato).

mono- and bi-species pots were weighted individually three times per day and automatically watered with nutrient solutions as described in Moreau et al. (2015) at two contrasted N concentrations: 0 (N-) or 14 mmol/L (N+) of nitrate, in order to reach 70% of water holding capacity in each pot. The air temperature was  $21.8 \pm 0.8$  °C, and the incident photosynthetically active radiation was  $20.3 \pm 4.8$  mol m<sup>-2</sup> d<sup>-1</sup>, with a 16 h photoperiod. Eighty-four days after seedling transplantation, the shoot part of the plants was harvested and shoot biomass per pot was determined after drying (48 h at 80 °C). The rhizosphere soil was sampled from the four root systems as a whole of each pot as described by Offre et al. (2007), and suspended in 250 ml of water in flasks instead of 100 ml. Soil DNA was extracted using the ISO-10063 procedure (Petric et al., 2011; Plassart et al., 2012). The genetic structure of rhizosphere bacterial communities was characterized by Automated Ribosomal Intergenic Spacer Analysis (A-RISA) (Ranjard et al., 2003), which exploits the variability in the length of the intergenic spacer (IGS) between the 16S and 23S sub-unit rRNA genes among different bacterial genotypes. PCR amplification of the 16S–23S intergenic spacer region in the rRNA operon was performed with a fluorescence-labeled forward primer and A-RISA-PCR fragments ranging in size from 400 to 1200 bp were next discriminated and measured by using an automated electrophoresis system. DNA purification, quantification and A-RISA fingerprinting procedures were performed as previously described (Ranjard et al., 2003).

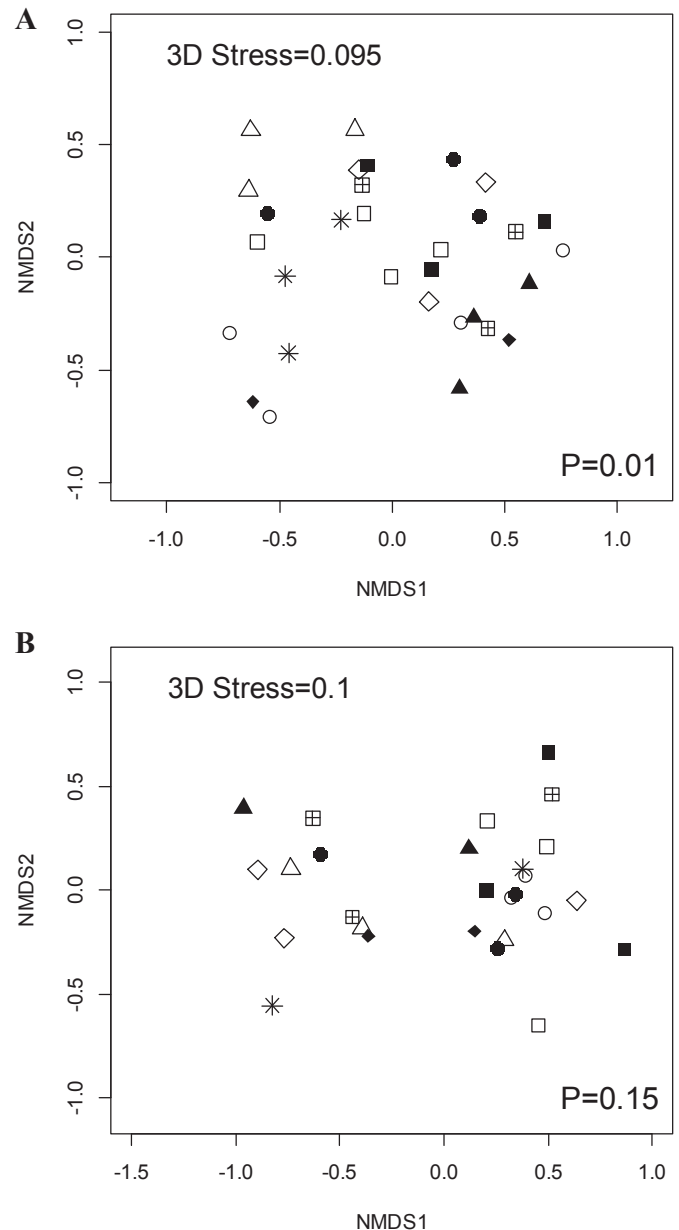
The abundances of the total bacterial and crenarchaeal communities (16S rRNA), the bacterial and Thaumarchaeal ammonia oxidizers (AOB and AOA), and of the dissimilatory nitrate reducers (NarG and NapA) were determined by quantitative PCR (qPCR) (Bru et al., 2011). Plant shoot biomass per pot and qPCR abundances were submitted to two-way and one-way ANOVA followed by Tukey test. Non-Metric Multidimensional Scaling (NMDS), with Bray-Curtis as distance matrix, followed by Permutational MANOVA (PERMANOVA; Anderson, 2001), was used to analyze the genetic structure of bacterial communities. All statistical analyses were performed using the 3.2.1 version of R (R Development Core Team, 2014). NMDS and PERMANOVA were run in Vegan package (Oksanen et al., 2012).

Shoot biomass of plants per pot were different depending both on plant association and N level (two way ANOVA with plant association and N level as factors; plant association: F-ratio = 11.02 and  $P < 0.0001$ ; N level: F-ratio = 1056.42 and  $P < 0.0001$ ; plant association  $\times$  N level: F-ratio = 9.63 and  $P < 0.0001$ ), but differences among plant associations were detected only in N+ (Supplementary information, Fig. 1), with *T. aestivum* ( $113.17 \pm 1.97$  g), [*A. myosuroides* + *T. aestivum*] ( $105.35 \pm 2.85$  g) and [*A. spica-venti* + *T. aestivum*] ( $104.43 \pm 4.13$  g) showing the highest biomass and *V. myuros* ( $47.27 \pm 3.31$  g) and [*A. spica-venti* + *V. myuros*] the lowest ( $52.1 \pm 16.74$  g). These results are in accordance with the different plant species affinities for N, with *A. spica-venti* and *V. myuros* having lower N requirements than *A. myosuroides* and *T. aestivum* (Ellenberg, 1974; Moreau et al., 2013).

A strong impact of soil N level was also observed on the genetic structure of rhizosphere bacterial communities (PERMANOVA analysis,  $P = 0.001$ ). In N- conditions only, the plant associations impacted differently the genetic structure of rhizosphere bacterial communities (Fig. 1), and this is probably due to enhanced competition for N between plant roots and rhizosphere microorganisms, as reviewed by Kuzyakov and Xu (2013).

Plant associations strongly impacted the abundances of total bacterial and N-cycling microbial communities, and this effect was even higher at N- than N+ conditions (two and one way ANOVA, Supplementary information, Fig. 2).

In order to check the effect of bi-species associations on microbial communities abundances in comparison to their respective



**Fig. 1.** Ordination plots of ARISA profiles from rhizosphere total bacterial communities, based on the Bray-Curtis similarity matrix: representation of N- (A) and N+ (B) bacterial community structure. The quality of ordination is indicated by fair level of stress values, calculated in three dimension NMDS. Non-parametric MANOVA (PERMANOVA, Anderson, 2001) on Bray-Curtis distances was used to test if bacterial community structures differed significantly according to plant association for each N level and corresponding p-values are reported at the bottom-right side of each graph.

Legend:  $\triangle$  *Alopecurus myosuroides* (Am),  $\diamond$  *Apera spica-venti* (Asv),  $\blacksquare$  *Triticum aestivum* (Ta),  $\circ$  *Vulpia myuros* (Vm),  $\blacktriangle$  [Am + Asv],  $\blacklozenge$  [Am + Ta],  $\square$  [Am + Vm],  $\bullet$  [Asv + Ta],  $\ast$  [Asv + Vm],  $\boxplus$  [Ta + Vm].

mono-species, further tests were performed on total bacterial and functional communities (Fig. 2). Significant effects were only detected at N- conditions (Fig. 2A, C, E, G and I).

In N- conditions, in half of the bi-species associations, the abundance of the total bacteria in the rhizosphere was higher than that of both corresponding mono-species (Fig. 2A). For the other bi-species associations, the abundance of the total bacterial community was similar to that obtained in the rhizosphere of at least one correspondent mono-species culture. A positive effect of bi-species associations was also observed on the abundance of the studied N-

Download English Version:

<https://daneshyari.com/en/article/5516300>

Download Persian Version:

<https://daneshyari.com/article/5516300>

[Daneshyari.com](https://daneshyari.com)