ENVIRONMENTAL METABARCODING REVEALS CONTRASTING MICROBIAL COMMUNITIES AT TWO POPLAR PHYTOMANAGEMENT SITES

Michel Chalot[a], J Foulon[a], C Zappelini[a], A Durand[a], B Valot[a], D Blaudez[b],

[a] *Université de Bourgogne Franche-Comté, Laboratoire Chrono-Environnement, Montbéliard, France*

[b] *Laboratoire Interdisciplinaire des Environnements Continentaux, UMR 7360 CNRS-Université de Lorraine, Faculté des Sciences et Technologies, Vandoeuvre-les-Nancy, France*

michel.chalot@univ-fcomte.fr

Keywords: Bacterial 16S; Fungal ITS; Illumina MiSeq sequencing; Phytomanagement; Poplar clones

Introduction

Little is known about the entirety of the microbial communities that are associated with tree roots in derelict soils. As a first step towards tree-based phytoremediation that is assisted by symbiotic fungi, we have adopted a metabarcoding approach that employs the high-throughput Illumina MiSeq platform to investigate whether the soil structure and composition and the contaminant levels have the largest effects on the diversity of bacterial and fungal phyla. To investigate potential variance caused by the site's history, we compare two contrasting contaminated areas in France. We hypothesized that the implementation of poplars would significantly and differentially shape the fungal and bacterial communities.

Methods

Sampling procedure and soil physico-chemical properties

After litter removal, the roots were collected from the upper 20 cm layer of soil from under the tree canopies. For each poplar clone, five individual trees were randomly sampled, and for each individual tree, 3 pseudo-replicates were sampled and mixed to obtain root and soil composites. Within the planted or unplanted control plots, five bulk soil samples were collected directly below the root system of the poplars (planted) or natural vegetation (unplanted) from the top 20 cm of soil.

Molecular methods

The total DNA was extracted from soil with a PowerSoil® DNA Isolation Kit. Equimolar DNA pools were created and adjusted to 10 ng/µl. The sequencing of the V3-V5 domains of 16S rRNA genes and the fungal ITS2 was performed with an Illumina MiSeq platform (Microsynth AG, Switzerland) using adequat PCR amplifications. Reads were assigned to each sample according to a unique barcode, and contigs were then

This work is licensed under a Creative Commons Attribution-NonCommercial-NoDerivatives 4.0 International License.

assigned using the Mothur pipeline. Taxonomic assignments were performed using a Bayesian approach in the Greengene database for bacteria and the UNITE database for fungi as described in Zappelini et al (2015).

Results

A soil analysis revealed that the two soils displayed contrasting physico-chemical characteristics. Diversity indices and β-diversity measures illustrated that the root microbial communities were well separated from the soil microbial communities at Leforest (L) and Pierrelaye (P) experimental sites. A detailed study of the fungal composition showed that *Ascomycota* dominated the overall fungal communities on poplar soil, the root samples at Pierrelaye, and the unplanted soil at the experimental sites. Conversely, *Basidiomycota* accounted for a much higher percentage of the fungal community in poplar root samples from the Pierrelaye site. The occurrence and dominance of the ectomycorrhizal community at Leforest but not at Pierrelaye is the major feature of our data set.

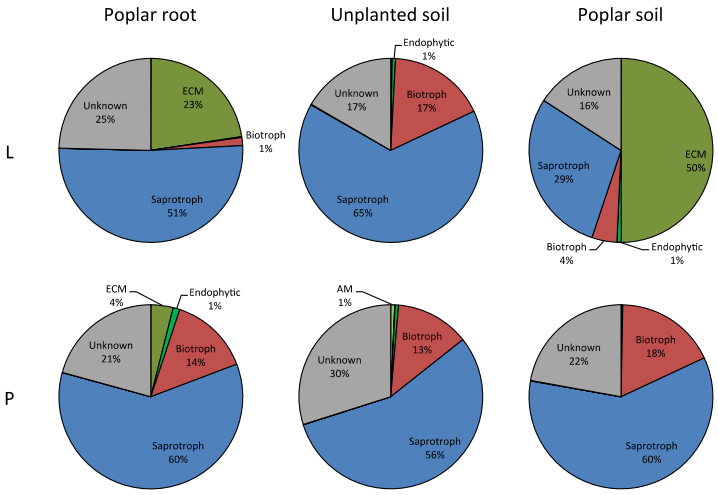

Figure 1. Relative proportions of fungal sequences assigned to major functional groups. The abundances of the various OTU groups from soil and root samples that were collected under *Populus* cv Skado or from the unplanted soil at the Leforest (L) and Pierrelaye (P) experimental sites were set to 100%, and the OTUs were classified as ectomycorrhizal (ECM), arbuscular mycorrhizal (AM), endophytic, saprotrophic or biotrophic

Conclusion

Overall, ectomycorrhizal root symbionts appeared to be highly constrained by soil characteristics at the phytomanagement sites

Reference

Zappelini, C.; Karimi B.; Foulon, J.; Lacercat-Didier L.; Maillard, F.; Valot, B., Blaudez, D. ; Cazaux, D. ; Gilbert D. ; Yergeau E. ; Greer C. ; Chalot M (2015) Diversity and complexity of microbial communities from a chlor-alkali tailings dump, *Soil Biol. Biochem*. *90*, 101–11

This work is licensed under a Creative Commons Attribution-NonCommercial-NoDerivatives 4.0 International License.