

Effect of AgNPs in the functional and structural microbial community using ARISA and next generation sequencing

Benjamin Gomez Pizarro^{a,b}, Karsten Schlich^a, Kerstin Hund-Rinke^a, and Cecilia Díaz^a

^a Fraunhofer Institute for Molecular Biology and Applied Ecology, Auf dem Aberg 1, 57392 Schmallenberg, Germany

^b Universidad Andrés Bello, Facultad de Ciencias de la Vida, Echaurren 281, Santiago, RM 8370146, Chile.

Contact: karsten.schlich@ime.fraunhofer.de

Introduction

- The effect of pollutants on soil microflora is of interest since they play a significant role in the functioning of the nutrient cycles and the retention function for pollutants
- Is it possible to identify changes in functional and structural microbial community of applied soils through the assessment of the active fraction of genetic material (RNA) and DNA, respectively?
- **Objective:** Evaluate the performance of two community fingerprinting methods to assess the effect on microbial community, in a study case using silver nanoparticles (AgNPs)

Test setup

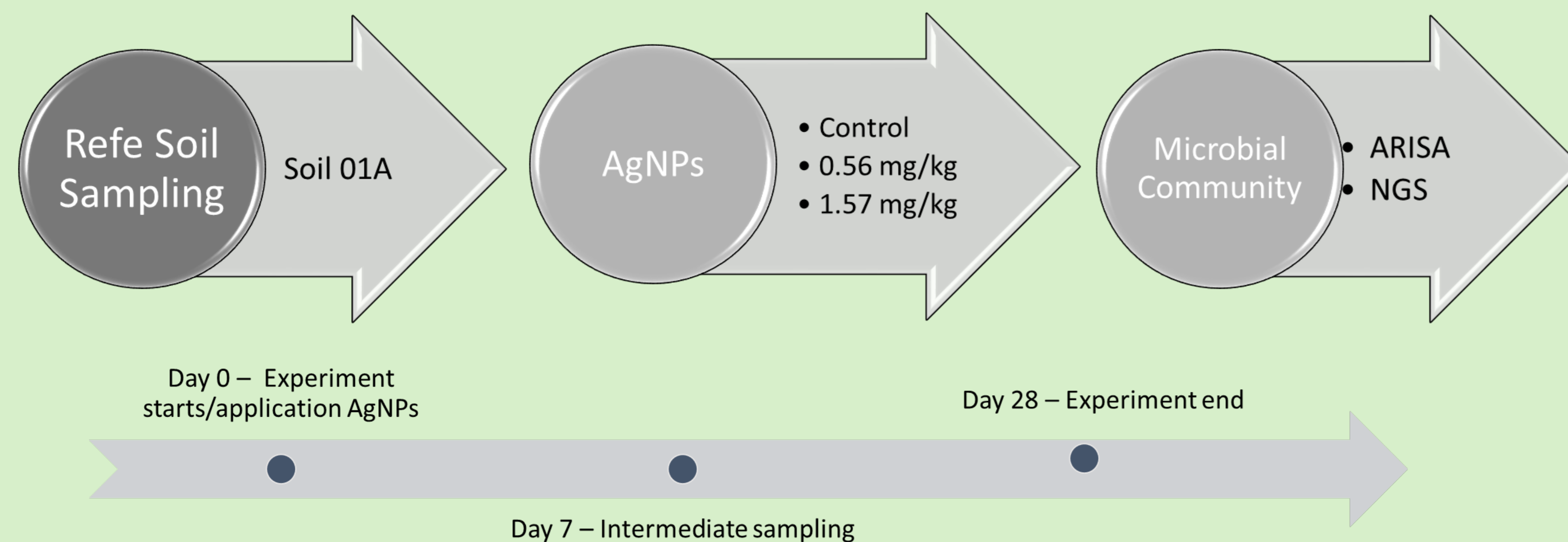


Figure 1. Test set up and sampling schedule. Samples were taking at at the beginning (day 0) and after 7 and 28 days. On each sampling DNA and RNA (reverse transcribed to cDNA before analysis) of the samples were extracted and the microbial community analyzed with ARISA-PCR and NGS.

Microbial community analysis:

- Automated Ribosomal Intergenic Spacer Aanalysis (ARISA). PCR amplification of the 16S-23S intergenic spacer region in the rRNA. ARISA-PCR fragments ranging in size from 400 to 1,200 bp were next discriminated and measured by using an automated electrophoresis system.
- Next generation sequencing (NGS): metabarcoding of the 16S rDNA (Illumina MiSeq Sequencing technology)

Results and conclusion

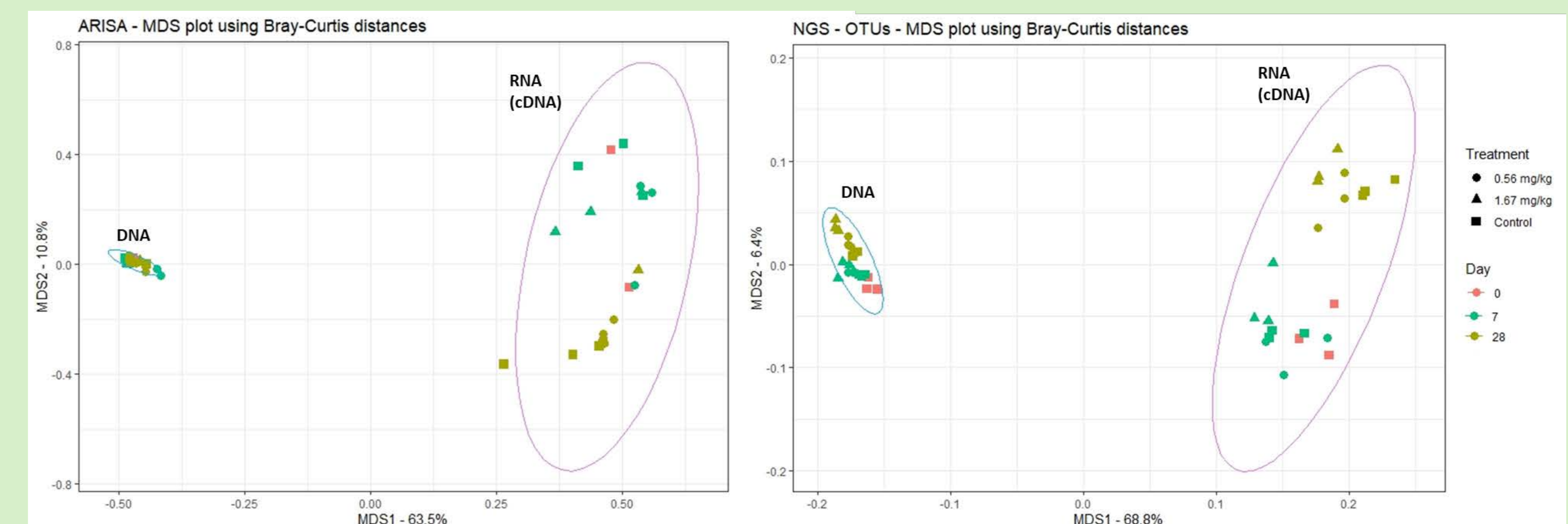


Figure 2. Metric multidimensional scaling (MDS) analysis of bacterial communities, evaluated as OTUs, using Bray-Curtis distance.

- The larger changes in community profiles were observed in the functional community (cDNA).
- Changes in the microbial community were observed during the time course of the experiment, but it was also possible to observe an effect of the AgNPs in the community.
- With both techniques it was possible to observed the mentioned effects (similar cloustering)

Recommendation: ARISA-PCR is a cheap and useful technique which is of interest to observe general effect or changes in a community, and NGS provides deeper insight and it is possible to discriminate to a deeper level between samples