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# Link between seed and root microbiota : what impact on telluric parasitism of plants ?



## Social-economic context

Winter oilseed rape (*Brassica napus*) is worldwide cultivated, mostly for its oil production and livestock feeding. It is today largely known that the massive use of chemicals on crops is deleterious for both environmental, animal and human health. On the other hand, many studies have shown the beneficial effects of microbial communities on plant growth and health. Thus, to study plant associated microbiota could be a good alternative to help reducing the use of pesticides.

## Scientific context

Plant associated microbial communities could have a positive impact on their hosts fitness, by modifying the production of primary metabolites, the flowering kinetics, or resistance against pests. To date, it is recognized that the microbial communities associated with the rhizosphere come from the surrounding soil. However, seeds are vectors for diverse microbial sets that could be transmitted over several generations, by vertical transmission. Thus, as well as the surrounding soil, seed can be considered as source of primary inoculum for the rhizosphere, and can thereby have an impact on telluric parasitism.

## Objectives

### Q1 : Does the seed microbiota participate in the roots colonisation ?

The first part of this project consists in analysing the microbial diversity of seeds harvested from several genotypes of oilseed rape cultivated in 2016 and 2017. Microbial diversity is assessed with barcoding approaches using two bacterial markers (16S and *gyrB*) and one fungal marker (ITS1). This first step will help to select two oilseed rape genotypes with contrasting microbiota compositions.

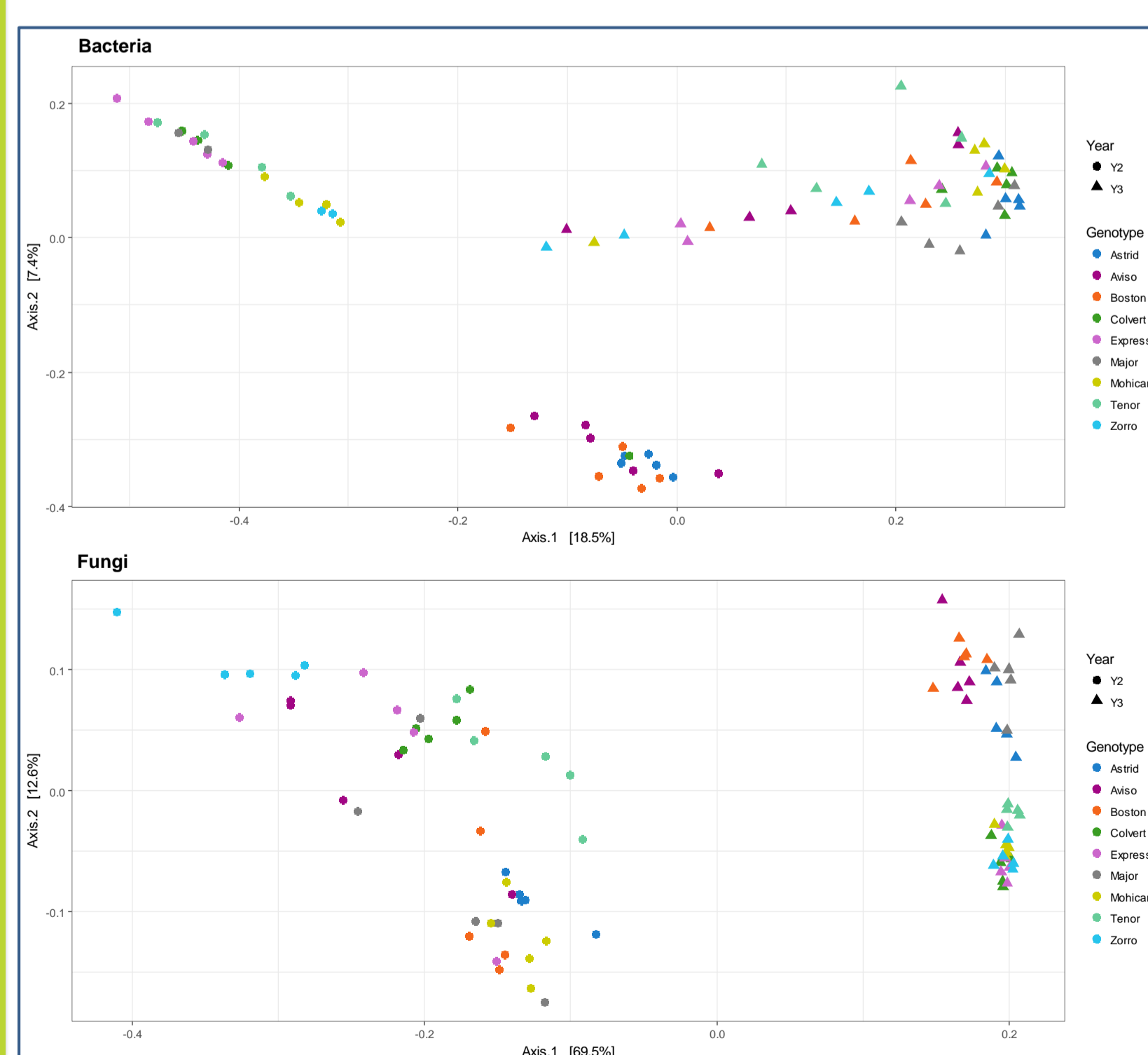
Sensitivity rate of these genotypes against two pathogens will then be estimated : *Rhizoctonia solani* causing damping-off and root rot, and *Verticillium longisporum* causing plant wilt.

For the selected genotypes, seeds harvested in 2016 and 2017 will be sown in two soils with different levels of diversity (low and high), with or without the presence of one pathogen. Expression of the disease will be measured by the observation of symptoms on roots, and pathogens will be quantified in roots by qPCR. On the other hand, taxonomic diversity of both soils and roots microbiota will be studied using barcoding approaches.

### Q2 : How and through what mechanisms does the seed promote the primary and secondary colonisation of the microbiota ?

To answer this second question, synthetic communities will be constituted to reduce assemblage complexity. For this purpose, isolations of cultivated bacteria associated with seeds will be done on agar medium. For each replicate, colonies will be randomly collected and prick out in 96-wells plates. Next, each strain will be typed with *gyrB* marker using high-throughput sequencing. These synthetic communities will then be applied on oilseed rape flowers to evaluate whether there is an internal transmission to the seed and to estimate the persistence of each strain. Metabolomics and metatranscriptomics approaches would also be considered.

## Results



The first results show that the seed microbiota is influenced by host genotype and year of production.

Figure 1 represent the differences in bacterial and fungal assemblages between each samples (one dot = one sample). These results show a high influence of the year of production on the structure of both bacterial and fungal communities regardless of the genotypes. Concerning the plant genotype, differences are highlighted : some genotypes have very close microbial structures, whereas others have more contrasted microbiota.

Other results show that there is a higher bacterial richness and a lesser fungal diversity for all genotypes in 2017 than in 2016.

Figure 1 : Bacterial and fungal community structures estimated with ordination of Bray-Curtis similarity indice with comparison between the year of production and the plant genotype.

## Perspectives

This project will help to better understand the link between the seed and the root microbiota. By selecting seed microbial communities that would play a role on recruitment of beneficial root microbiota, it would be possible to promote the plant fitness and help fighting against some pathogens.

