

Rhizosphere 2



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Rhizosphere 2

Session 10 - Functional and structural diversity - fungi and mesofauna

P-918 From oligonucleotide barcoding to development of a phyloarray for truffle species identification

Zampieri Elisa¹, Murat Claude¹, El Karkouri Khalid², Bonfante Paola¹

¹ Università degli studi di Torino biologia vegetale viale Mattioli 25 10125 Torino Italia Italy

² Unité des Rickettsies, CNRS UMR 6020, IFR-48, Faculté de Médecine, Université de la Méditerranée, 27 Boulevard Jean Moulin, 13385, Marseille cedex 05, France

Truffles are edible mycorrhizal fungi largely required by food markets for their taste and flavour. Many PCR-based studies have been focused on the identification of single truffle species. Here, we provide the first data on the development of a new molecular method to identify a relatively high number of *Tuber* species with a single experiment. The method is based on the technique of DNA barcoding, i.e., the use of a short gene sequence from a standardized region of the genome as a diagnostic "biomarker" for species (<http://www.barcoding.si.edu/>).

As a first step, the method required the identification of several species-specific oligonucleotides (or motifs), located within the internal transcribed spacer (ITS) of the rDNA repeat. The barcode has been developed for five *Tuber* spp. (*T. magnatum*, *T. melanosporum*, *T. aestivum*, *T. indicum* and *T. mesentericum*) on the bases of 526 ITS sequences available in GeneBank and allowed a reliable *in silico* identification of these *Tuber* spp.

In a second step, after a dot blot experiment one species-specific motif was selected among those designed for *T. magnatum* and *T. melanosporum*. In a blind test these two oligonucleotides allowed to identify both truffle species among Ascomycetes and Basidiomycetes DNAs. The experiments confirmed the usefulness of the technique in the identification of the two truffles.

The last step of the project involves the development of a phyloarray, which is currently under construction. Thanks to this array, where species-specific motifs are spotted, multiple *Tuber* species will be detected at the same time. This new identification tool will allow to detect truffles in complex matrix such as mycorrhizal roots and soil.

P-965 Arbuscular Mycorrhizae Fungi (AMF) associated with rhizosphere of banana systems

Vanlauwe Bernard², Ruto Laban³, Kahangi Esther³, van Asten Piet¹, Losenge Turop¹, Mnyazi Jefwa Joyce¹

¹ National Museums of Kenya Herbarium department 45166 00100 Nairobi Nairobi Kenya

² Tropical Institute of Soil Biology and Fertility-CIAT (TSBF-CIAT)

³ Jomo Kenyatta University of Agriculture and Technology (JKUAT)

⁴ International Institute of Tropical Agriculture (IITA)

A survey was undertaken in central Kenya to establish AMF in the rhizosphere of 7 tissue culture (TC) and 11 local banana cultivars. Soils were taken, and AMF spores morphologically identified and counted. The same soil samples were used to establish trap cultures with sorghum, soybean, cooking (Kibuzi) and a desert (Giant Cavendish) banana cultivar. A total of 22 AMF species (morphotypes) were present in banana systems comprising of 12 *Glomus* spp., 7 *Acaulospora* spp., 2 *Scutellospora* spp and 1 *Gigaspora* sp. Banana cultivars significantly ($p = 0.002$) affected spore abundance, with slight variations noted in the probability of occurrence of AMF species. The species richness and diversity ranged from 0-18 and 0-1.971 respectively. With the exception of Grand naine, Chinese dwarf and Lacatan, which were the least common of tissue culture banana cultivars, high species richness and diversity was generally found in the rhizosphere of all TC banana cultivars. The highest species diversity (Shannon index) was however described in two local cooking cultivars, Githumo (2.1455) and Kiganda (2.531) and desert cultivar, sweet banana (2.522). Rank abundance curve showed two most abundant *Acaulospora* spp. to have higher proportion of spores (45.9%) than the three most abundant *Glomus* spp. (29.7%). Trap cultures had total of 13 AMF species with highest species richness in Soybean (10) while sorghum and the two banana cultivars had 8 species each. Two *Glomus* spp. were specifically associated with the two banana cultivars. The highest total spore abundance was in sorghum (712) followed by soybean (571) with the least being 334 and 338 in Kibuzi and Giant Cavendish respectively. Shannon diversity index was highest for soybean (1.679) and Giant Cavendish (1.105) and low in sorghum (0.893) and Kibuzi (0.612). Renyi profile curve shows soybean to have the highest species richness and evenness. Three *Acaulospora* species account for 65.4% of the total spore abundance while only 18.1% represent the two most abundant *Glomus* spp. The total species diversity index was lower (1.955) in trap cultures compared to the field soils (2.162). The rhizosphere of TC banana cultivars is favorable for AMF. A proportion of 80% of AMF species from rhizosphere of banana systems associates with other hosts.

P-998 Effects of zeaxanthin-accumulating potatoes on fungal communities in the rhizosphere

Meincke Remo¹, Weinert Nicole², Radl Viviane³, Kania Angelika⁴, Dong Xia⁵, Neumann Günter¹, Schlöter Michael³, Smalla Kornelia², Berg Gabriele⁴

¹ Graz University of Technology Institute of Environmental Biotechnology Petersgasse 12 8010 Graz Steiermark Austria

² The Federal Biological Research Centre for Agriculture and Forestry, Institute for Plant Virology, Microbiology and Bio safety, Braunschweig, Germany

³ GSF - National Research Center for Environment and Health, Institute of Soil Ecology, Neuherberg, Germany

⁴ University of Hohenheim, Plant Nutrition, Stuttgart, Germany

⁵ Technische Universität München, Chair of Plant Breeding, Munich, Germany

⁶ Graz University of Technology, Institute of Environmental Biotechnology, Graz, Austria

To determine effects of the transgenic modified potato clones SR47 und SR48 on the fungal community a multiphase approach of cultivation-dependent methods (CFU, functional analysis of antagonists) and cultivation-independent analyses (SSCP) was chosen. To compare influence of the GM potatoes with natural variations, four commercial cultivars (Selma Désirée, Ditta, Sibü) were included. The two clones, the parental line and the four commercial cultivars were planted in six randomized plots per treatment at different locations in Germany (Roggenstein 2005, Oberviehhausen 2006). Samples were taken at three development stages (EC30, EC60 and EC90). Each sample contained root material of five plants and for the geocaulosphere material from tubers of five plants. The CFU in the rhizosphere showed no significant differences between the plants but statistically lower values in the geocaulosphere. The antagonistic activity was tested against *Rhizoctonia solani* Kühn, *Verticillium dahliae* Kleb. ELV25 and *Phytophthora infestans* (Mont.) de Bary. A lower portion of antagonists was observed for the clone SR48 than for the others at the first sampling time. However, the natural variation between the different cultivars was high. The cultivation-independent analysis of fungal communities by SSCP of the PCR-amplified ITS-region shows no differences between the parental line and the two transgenic clones. Small variations for some ribotypes were observed. Differences in community structure were observed for the sampling time in all cultivars. Altogether, cultivation-dependent and cultivation-independent analyses showed a higher variation between the different cultivars than between the parental line and the two transgenic clones.