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# OBSERVATION OF ECTOMYCORRHIZAL FUNGI (ECM) STRUCTURES IN EUCALYPTUS DEEP ROOTS (SÃO PAULO, BRAZIL)

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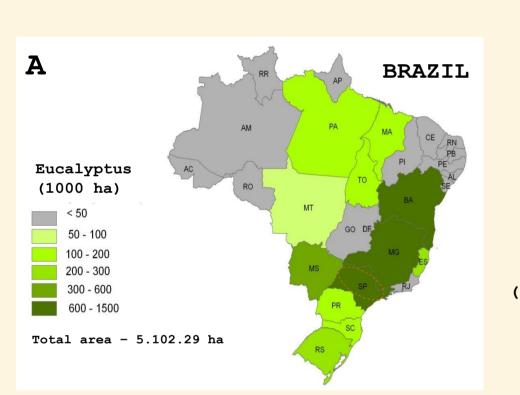
#### INTRODUCTION

Many field observations suggest that ECM contribute to a number of key ecosystem functions such as carbon cycling, nutrient mobilization from soil organic matter and soil minerals. In Brazil, the ECM form symbiotic associations with species of great economic importance, belonging to Eucalyptus genus. Furthermore, most of the studies are concentrated in the superficial soil layers and studies assessing root colonization by mycorrhizal fungi at depths > 50 cm are few. This study aimed at determining of ECM structures in deep roots of *Eucalyptus grandis* (2 to 4-years-old).

#### **MATERIALS AND METHODS**

The study was conducted in the Southeast of Brazil (Figure 1). Soil is a very deep Oxisol with low clay content (~20%). The mean annual rainfall was about 1400 mm and temperature was 19°C. Deep roots were observed using two strategies:

- a-) with minirhizotrons positioned in a deep pit to examine fortnightly the development of fine roots (≤ 2 mm) from 2011 to 2013 (site 1);
- b-) following root collection in March 2013 during the excavation of a pit and observed with a microscope (site 2).



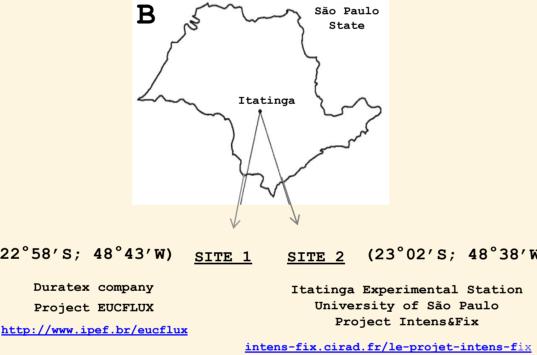




Figure 1 - Location of study sites (A and B); Permanent pit (C); Root scanner (D); Minirhizotron system (E) and (F) microscope (MZ 12.5 - LEICA).

The pictures were taken using a root scanner system at the tubes inner surface producing a 360° image of the tube-soil interface (21 x 19 cm). The images were analyzed with the software WinRHIZO TRON 2009.

Root fragments were photographed using a microscope paired with a software (Motic Images Plus 2.0). Isolated mycorrhizal apex were specifically sampled in order to better characterize the structures (analysis in progress).

#### **RESULTS AND DISCUSSION**

Observations with minirhizotrons: the sequence of images showed the complete disappearance of the mycelia networks after twelve months of observations (Figure 2). The mycorhizal roots shown in the Figure 3 has a diameter of 0.25 mm and its elongation rate was 0.37 cm per day from 16/02/12 to 12/05/12.

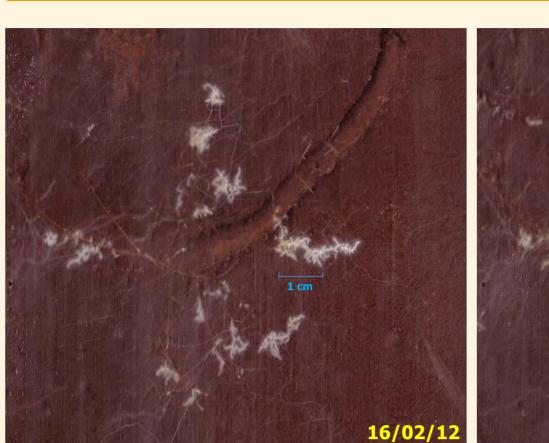
Minirhizotrons studies showed strong changes in fine root morphology, which suggested a possible mycorrhization (Figure 4). These results encouraged us to collect deep roots by excavation to confirm and to identify the fungal species able to form ECM on deep roots

## **SITE 1 - MINIRHIZOTRONS**



Figure 2 - Changes in mycelium areas (0-50 cm) over the first year of observations with the minirhizotron system.

#### **SITE 1 - MINIRHIZOTRONS**





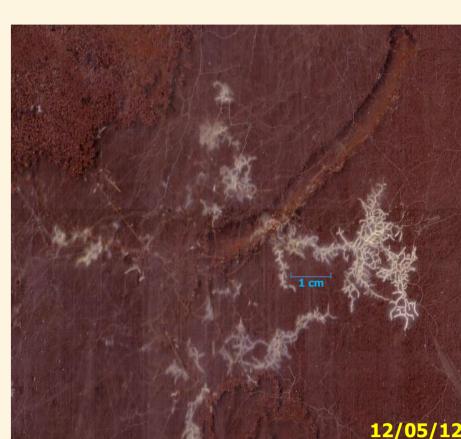
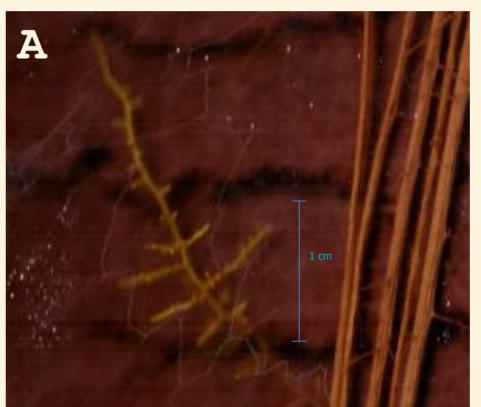
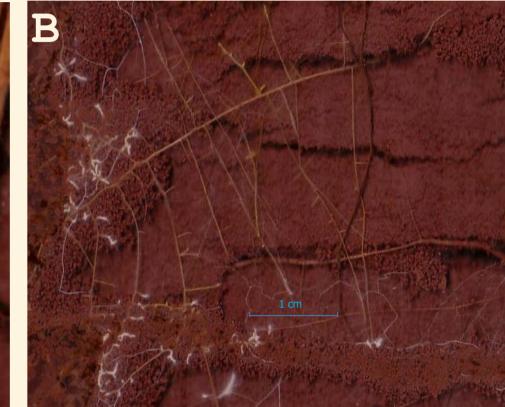


Figure 3 – Elongation of mycorrhizal roots (0-50 cm) monitored through the minirhizotron system.





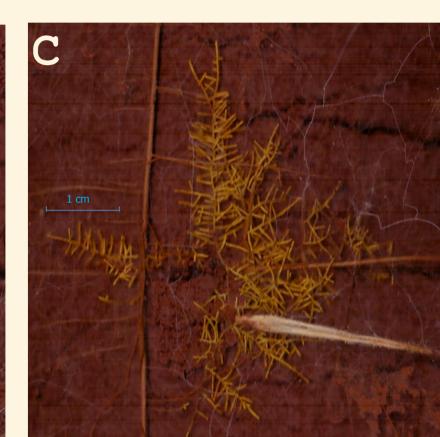


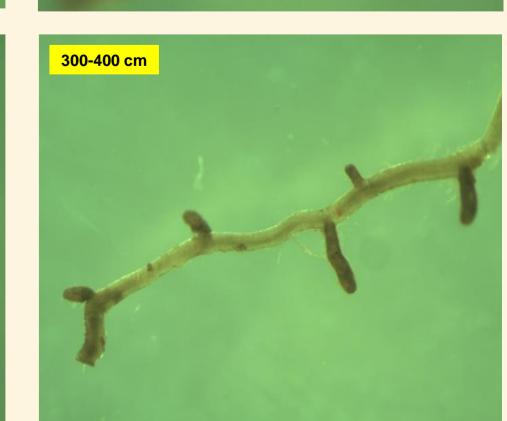
Figure 4 - Changes in deep roots between of 4-5 m (A); 2-3 m (B) and 1-2 m (C) monitored through the minirhizotron system.

Similar ECM structures shown by changes in root coloration and shapes were found from the soil surface down to a depth of 4 meters (Figure 5). This study is being complemented by DNA extractions of samples selected for comparisons of structures like ECM.

#### SITE 2 – PIT SAMPLING







**Figure 5** – Possible ECM structures photografed between 0 and 4 meters.

### **CONCLUSION**

Minirhizotrons and observation of root fragments strongly suggest the presence of very deep ECM in *E. grandis* plantations. However, molecular studies are needed to confirm these observations.