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COLLECTION OF ROOTS OF NATIVE HOSTS AND RHIZOSPHERIC SOIL

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ABSTRACT

The rhizospheric soils from eleven selected sites were collected on seasonal bases along with the available native host over a period of one year. At every selected site, the collection method employed was the same, i.e. rhizospheric soil cores from 5-10 different sites around roots of host plants were collected. The collected soil/ rhizospheric soil cores were mixed thoroughly and were sub-sampled by repeated quartering and pooling opposite quarters (Ambler and Young, 1977). Fine root system of the native host species was also separated and collected for analysis of mycorrhizal status. The host as well as the rhizospheric soil were packed in labeled polythene bags separately and were brought to the laboratory of Shankarrao Mohite College, Akluj.

The rhizospheric soil collected during the three seasons i.e. rainy, winter and summer season from all the eleven sites after bringing to the laboratory were allowed to air dry. These soil samples on air drying were then sieved through sieves of holes of 2 mm diameter and stored in labeled airtight plastic containers at 4^{o}C for further physico-chemical analysis and spore extraction.

Usually arbuscular mycorrhizal fungi infected roots were observed in the upper 0-30 cm of the soil core. For all plant roots studied for arbuscular mycorrhizal fungi, colonization root systems selected were fine, terminal and fresh/live. These "feeder roots", which provide the obligate symbiont large surface area to interact with host plants.

In small seedlings or plants, the entire root system was collected for the measurement of root colonization. Soil dispersion agents "sodium pyrophosphate" have been used to remove adhering soil particles to the fine root system. Smaller root fragments were cleaned on sieve before assaying for arbuscular mycorrhizal colonization. Then, roots were cut in 1-2 cm pieces and stored in individual vials containing FAA (formalin 1: acetic acid 1: alcohol 18).

KEYWORDS: *rhizospheric soils , arbuscular mycorrhizal fungi , obligate symbiont.*

INTRODUCTION :

Assessment of arbuscular mycorrhizal colonization:

a) Clearing and staining roots (Phillips and Hayman, 1970):

Roots samples stored in FAA were first washed with tap water repeatedly for complete removal of FAA traces and then placed in a small glass vial. These root samples dipped in 10% (w/v) KOH were then digested in autoclave for 15 min at 121^{o}C. Clearing time and temperature for digestion varied with plant species. KOH solution was then poured off and the roots were rinsed with tap water for at least 3-5 times to remove excess of KOH solution. These roots were then surface covered with 1% HCl, which was poured off after 3-5 min. 0.05% trypan blue in lactophenol stain (1:1) was added to the above processed roots for

several hours. These roots were then immersed in lactophenol for destaining. Roots were finally observed under the compound microscope for arbuscular mycorrhizal fungi colonization.

b) Calculation of the percentage of root segments colonization (Giovannetti and Mosse, 1980):

For a particular host, the total number of infected root segments or root segments infected for any type of propagule (i.e. hyphae, arbuscules, vesicles or chlamydospores etc), out of the total number of root segments (of equal size) screened was expressed as percent root colonization. The root segments were uniformly spread on a slide to view all the root segments without having to move other segments. 100 root segments were mounted into groups of 10 for fast analysis. Calculation was done using the formula as -

$$\textbf{Percent root colonization} = \frac{\text{No. of infected root pieces}}{\text{No. of root pieces screened}} \times 100$$

CONCLUSION

At every selected site, the collection method employed was the same, i.e. rhizospheric soil cores from 5-10 different sites around roots of host plants were collected.

For all plant roots studied for arbuscular mycorrhizal fungi, colonization root systems selected were fine, terminal and fresh/live.

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