Pak. J. Bot., 36(3): 659-662, 2004.

SOME OBSERVATIONS ON MYCORRHIZAE OF *LEUCAENA LEUCOCEPHALA* (LAM.) DE WIT.

A. MAHMOOD, RUBINA IQBAL, RAIHA QADRI AND SAEEDA NAZ

Department of Botany,
University of Karachi, Karachi-75270, Pakistan.

Abstract

In young roots of *Leucaena leucocephala* infection hyphae formed appresorium, sending branches in different directions to colonize the host cell. The mycorrhizal fungus was non-septate and branched. Fungal cells (Pelotons) were observed in the root cortex. Characteristic arbuscules commonly formed by VAM fungus were visible. The main hyphae branched and the branched hyphae terminated in an arbuscule. The cortical cells of the root showed large number of vesicles which were either globular or elliptical in shape.

Introduction

Many tree legumes develop a double symbiosis with both nitrogen fixing bacteria and Vesicular–Arbuscular Mycorrhizae, VAM (Barea *et al*., 1990; Wall, 2000; Qadri *et al*., 2004). VAM are advantageous to woody legumes because they increase the uptake of low mobile nutrients in the soil especially Phosphorus (Manjunath et.al.1984). The combined effect of two symbionts, the Rhizobia and VAM help legume species to mitigate nitrogen and phosphorus two of the most important nutritional deficiencies, (Barea *et al*., 1988).

Leucaena leucocephala is a fast growing woody mimosoid legume which is well known for its adaptability in arid and semi –arid lands of the world including Pakistan. The tree has gained much impetus for its multifarious uses to mankind (Anon., 1984). Roots of *Leucaena* usually carry nitrogen fixing nodules (Iqbal & Mahmood, 1992). From the survey of literature it seems that no work has been done on the mycorrhizae of *L. leucocephala* in Pakistan. In this paper occurrence and structural aspects of mycorrhizae in *L. leucocephala* are presented.

Material and Methods

For mycorrhizal studies young roots of *L. leucocephala* were collected from trees growing in the garden of the Department of Botany, University of Karachi. Roots were thoroughly washed with tap water to remove any soil particles attached with them. Young roots were cut into small pieces, fixed in F.A.A. (Formalin Acetic acid Alcohol, (5: 5: 90) and embedded in paraffin wax (m.p. 56˚C). Transverse and longitudinal sections 10-12µm thick were cut on a rotary microtome. Paraffin was removed with xylol and slides were stained with safranin and fast green (Johanson, 1940). For the estimation of the extent of mycorrhizal infection, very thin roots obtained from *L. leucocephala* trees were gently washed under tap water and cut into 1cm segments, fixed in FAA, cleared in 10% potassium hydroxide at 90^{o}C and stained with trypan blue in lectophenol as described by Philips & Hayman (1970).

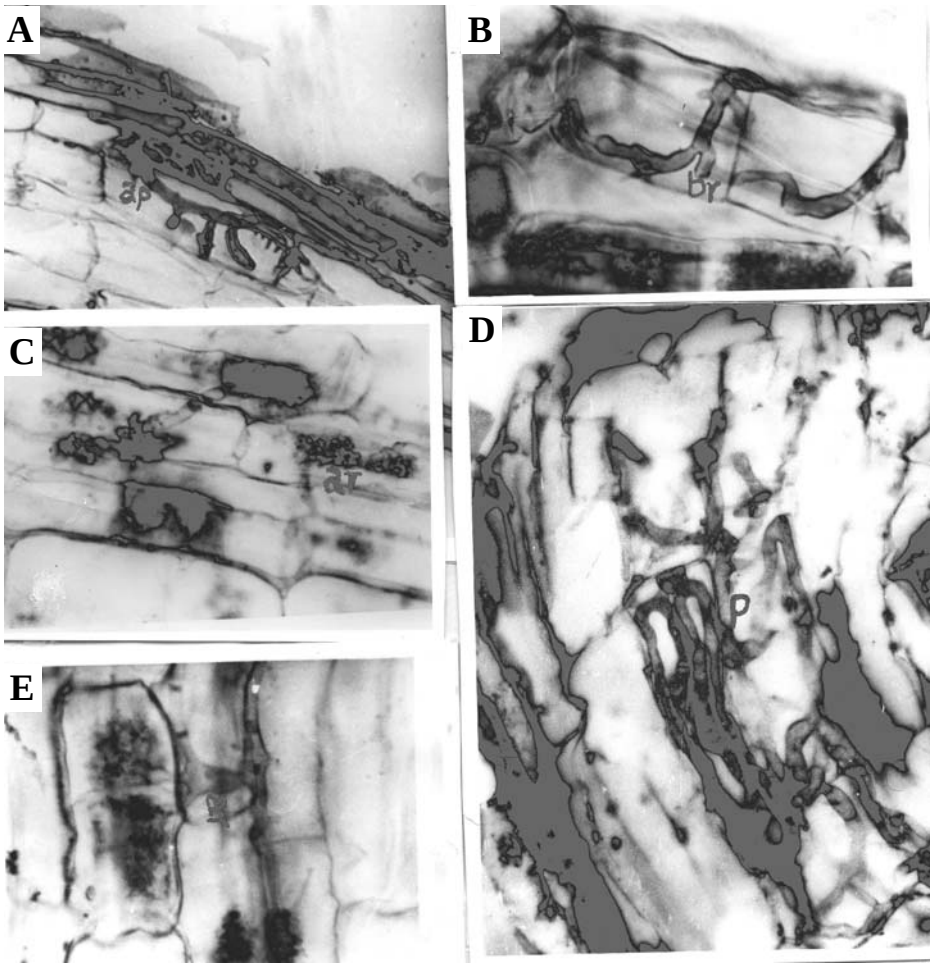

Fig. 1. Longitudinal section of roots of *Leucaena leucocephala*: A. Appresorium (ap) in the cortex X400, B. The hyphae are branched (b) X1000, C. Arbuscules (ar) X400, D. Coil of hyphae (pelotons) (p) X 400, E. A single arbuscule (ar) attached with the hyphae. X1000.

Results and Discussion

Infection hyphae forming appresorium (Fig. 1A). The appresorium ramified in different directions to colonize the host cell. The mycorrhizal fungus was non septate and branched (Fig. 1B). The diameter of the hyphae varied between 1-5 µm. Fungal cells (Pelotons) were observed in the root cortex (Fig. 1D). Characteristic arbuscules commonly formed by VAM fungus are visible in (Fig. 1C). A single arbuscule attached to hypha is shown in Fig. 1E. The main hypha branched and branch hyphae terminated in an arbuscule. The cortical cells of the root showed large number of vesicles. The vesicles were globular (Fig. 2A) and elliptical in shape (Fig. 3A). Globular type of vesicle have been reported in *Artemisia brevifolia* (Saif *et al*., 1977) and elliptical type in *Fagopyrum tataricium* (Saif & Iffat, 1976). The diameter of globular vesicles varied between 7-21µm

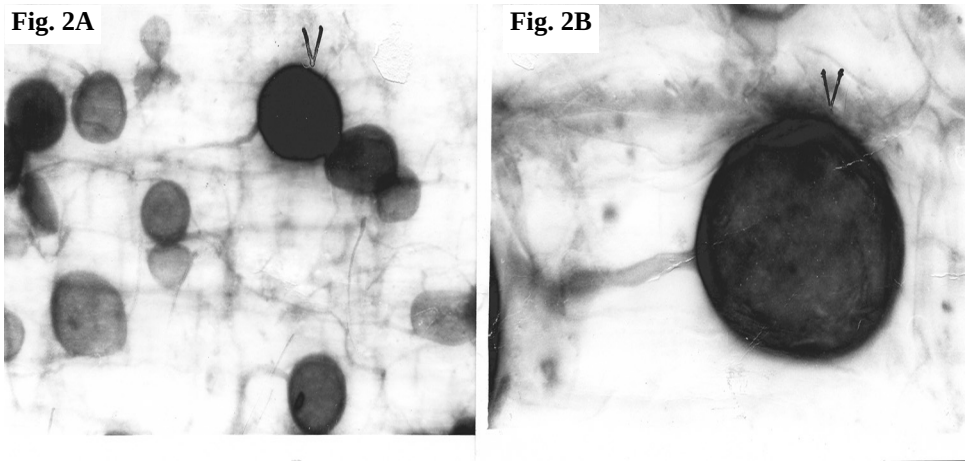

Fig. 2A. Longitudinal section of roots of *Leucaena leucocephala*: A. Globular vesicles (v) along with the hyphae. X400.

Fig. 2B. A single enlarged globular vesicle (v) showing open ended connection with the hyphae. X 2000.

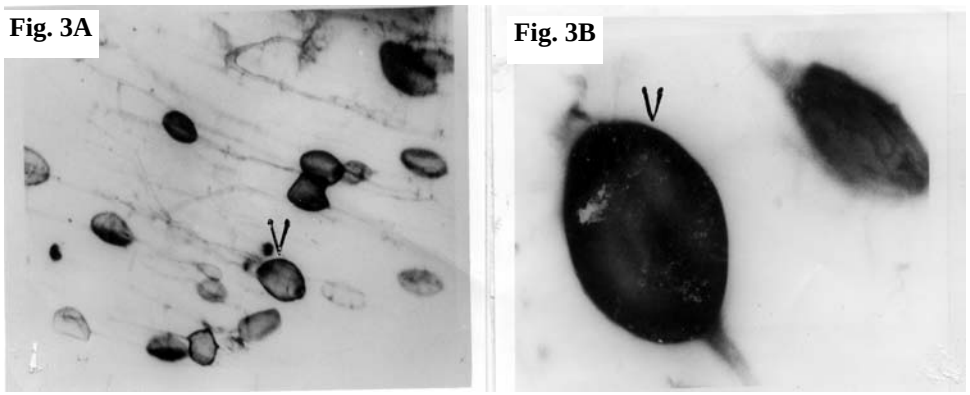

Fig. 3A. Longitudinal section of roots of *Leucaena leucocephala* showing elliptical vesicles (v) along with the hyphae X400.

Fig. 3B. A single enlarged elliptical vesicle (v) showing open ended connection with the hyphae X 2000.

whereas the elliptical vesicles varied between 10-17 µm. A single globular vesicle is shown at higher magnification (Fig. 2B) and an elliptical vesicle (Fig. 3B). Vesicles showed an open ended connection with the hypha (Figs. 2B & 3B). The occurrence of VAM in *L. leucocephala* support the reports of Saif (1975), Saif & Iffat (1976) and Saif *et al*., (1977) that VAM is the most common type of mycorrhizae found in flowering plants of Pakistan.

References

Anonymous. 1984. *Leucaena*: Promising forage and tree crop for the Tropics 2nd ed. National Academy Press Washington D.C. 1-100 pp.

Barea, J.M., C. Azcon-Aguilar and R. Azcon. 1988. The role of mycorrhiza in improving the establishment and function of *rhizobium*-legume system under field conditions. In: *Nitrogen fixation by legumes in Mediterranean Agriculture* (Eds.): D.P. Beak and L.A. Meterox. Martinus Nijhoff Publisher, Dordrecht, pp. 153-152.

Barea, J.M., C.P. Salamanca and M.A. Herrera. 1990. The role of VA mycorrhiza in improving Nitrogen fixation by woody legumes in arid zones. In: Fast growing trees and nitrogen fixating trees. (Eds.): D. Werner and P. Muller. Gustav Fisher Verlag, Stutgart, pp. 303-311.

Iqbal, R. and A. Mahmood. 1992b. Response of *Leucaena leucocephala* to inoculation with rhizobia from tropical legumes. *Pak. J. Bot*., 24(2): 153-156.

Johansen, D.A. 1940. *Plant Microtechnique*. McGraw Hill Book Company, Inc. New York, London, pp. 27-94.

Manjunath, A., D.J. Bagyaraj and H.S. Gopla. 1984. Dual inoculation with VA mycorrhiza and *Rhizobium* is beneficial to *Leucaena. Plant and Soil*, 78: 455-448.

Phillips, J.M. and D.S. Hayman. 1970. Improved procedure for clearing root parasitic and vesicular-arbuscular mycorrhizal fungi for rapid assessment of infection. *Trans. Br. Mycol. Soc*., 55: 158-160.

Qadri , R., S. Dawar and R. Iqbal. 2004. Presence of Endomycorrhiza and root nodules in *Samanea saman* (Jacq.) Merr. *Hamdard Medicus*, 57(1): 5-7.

Saif, S.R. 1975.The occurrence of mycorrhizae and endogone spores in rhizosphere of plants growing around University Campus, Islamabad *Pak. J. Bot*., 7(2): 175-182.

Saif, S.R. and N. Iffat. 1976. Vesicular-Arbuscular mycorrhizae in plants and endogonaceous spore in the soil of northern areas of Pakistan..*Pak. J. Bot*., 8(2): 163-179.

Saif, S.R., I. Ali and A.A. Zaidi. 1977. Arbuscular mycorrhizae in plants and endogonaceous spore in the soil of northern areas of Pakistan. III Dir and Chitral. *Pak. J. Bot*., 9(2): 129-148.

Wall, L.G. 2000. The Actinorrhizal Symbiosis. *J. Plant Growth Regul*., 19: 167-182.

(Received for publication 20 March 2004)