

ROLE OF ARBUSCULAR MYCORRHIZA STRAINS ON BIOMASS PRODUCTION AND P, Cu, Zn UPTAKE IN *PROSOPIS CINERARIA*

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Introduction

The significance of mycorrhiza in forest and agro-ecosystem is well established (Jefferies, 1987; Barea, 1991). Arbuscular Mycorrhizal (AM) fungi are known to have a worldwide distribution with little specificity in selection of their host plant (Mosse *et al.*, 1981; Daniel Hetrick, 1984). About 90 per cent of the plants surviving throughout the world form arbuscular mycorrhiza and a wide range of plants respond to this symbiotic association (Plenchette *et al.*, 1983). The host plant, growth enhances after AM inoculation by improving the supply of mineral nutrients of low mobility in soil, phosphorous in particular and also micronutrients like copper and zinc (Li *et al.*, 1991; Vijaya and Srivasuki, 1997). Researchers on mycorrhizal dependence of crops have clearly indicated that some important tropical crops and pastures are highly mycotrophic and will not grow or produce well in low P soils without an effective mycorrhizal association. Since mycotrophic plants depend on a AM association when grown under low external 'P' conditions, their yield can be enhanced by increasing the efficiency of AM fungi, either by inoculation, i.e. the introduction of more efficient fungal species or isolates to the rhizosphere, or by manipulation of the

population of native AM fungi through the use of agronomic practices that enhance the most efficient species in that population. Though some information is available on the mycorrhizal dependency of some tropical crops and forage species (Howeler *et al.*, 1987), no such work has been carried out in the area of forestry. Growth and vigour of *Prosopis cineraria* (L.) DRUCE seedlings are influenced by efficient strain of AM fungi and uptake of nutrients like P, Cu and Zn. Keeping this objective in view the present study was undertaken to select the efficient strains of AM fungi and uptake of immobile nutrients like P, Cu and Zn. The main purpose of this study was to select the most efficient strain of AM fungi for best growth and vigour of *P. cineraria* seedlings and to see the dependency and effect of AM infected trees on the uptake of P, Zn and Cu.

Material and Methods

An experiment was laid out under green house conditions to select the efficient strains of AM fungi for improvement in growth and vigour of *P. cineraria* seedlings and their effect on soil immobile nutrient uptake like P, Zn and Cu. Two months old seedlings of uniform size and growth were selected for the experiment. The plants were placed in polythene bags of

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size 25 x 15 cm holding 2.0 kg of sterilized soil: sand (2:1) mixture. One seedling was maintained per bag. The different AM fungal inoculums were obtained from Bangalore and Jodhpur (indigenous culture) and were maintained in a glasshouse in the pot using sterilized sand: soil mixture (1:1) as the substrate and *Pennisetum glaucum* (Pearl millet) as the host crop. The hyphae, spores and root segments present in this air dried substrate served as inoculum. The inoculum 10 g of 40 infective propagules per gram (400 infective propagules) applied of two months old seedlings by side banding method. The infective propagules were isolated by wet sieving and decanting technique (Gerdemann and Nicolsen, 1963). There were five treatments: T₀- Uninoculated; T₁- Mixed inoculum (indigenous): *Glomus* (70%), *Gigaspora* (14%), *Sclerocystis* (6%), *Scutellospora* (5%) and *Acaulospora* (5%); T₂- *Glomus microcarpum* (non indigenous); T₃- *Glomus aggregatum* (indigenous) and T₄- *Glomus fasciculatum* + *Glomus aggregatum* i.e., (50%: 50%). All treatments were replicated thrice and each replicate had sixteen plants. The experiment was laid out in Completely Randomised Design (CRD).

The plants were harvested 120 days after sowing and plant height (cm), root length (cm), shoot dry weight (g), root dry weight (g), plant biomass, root/shoot ratio, per cent root colonization, AM population 10 g⁻¹, uptake of P, Cu and Zn and Mycorrhizal Dependency (M.D.) was recorded. Mycorrhizal Dependency (MD) % was calculated by following formula (Menge *et al.*, 1978).

$$\% \text{ MD} = \frac{\text{Dry weight of inoculated seedling} - \text{Dry weight of un-inoculated seedling}}{\text{Dry weight of inoculated seedling}} \times 100$$

Height (cm) of seedlings was taken from the collar to tip with measuring scale. The shoots were cut near the base and roots of the harvested plants were excavated. Root length was recorded in (cm). For dry weight of shoot and root the samples were oven dried at 70°C for 72 hrs. For biomass estimation, dry weight of leaves + shoot + root was taken. Root:shoot ratio was also calculated.

Soil samples were processed for AM propagules using wet sieving and decanting techniques of Gerdemann and Nicolson (1963) and sucrose centrifugation technique as described by Jenkins (1964). Leica Kombistereio Microscope counted the AMF propagules. Roots were separated from collected soil samples and assayed for AM fungal association after staining in trypan blue as described by Phillips and Hayman (1970). A total of 100 root segments were examined for each replicate and percentage of segments with colonization was calculated. The AM fungal infection was examined by using Optiphot-2 "Nikon" compound microscope. The percentage of root infection was determined by Giovannetti and Mosse (1980).

Grinded and sieved (< 2 mm) plant samples were digested, using tri-acid mixture as described by Jackson (1973). Placed 0.5 g plant samples in 100 ml cooled conical flask and then added 5 ml of tri-acid mixture (HNO₃: H₂SO₄: HClO₄, (9:2:1). The digestion was carried out between 180 and 200°C until a clear solution remains after the acids were largely volatilised. After complete digestion the final volume was made up to 50 ml.

Phosphorus was estimated by ammonium metavanadate yellow colour

method (Jackson, 1973). The P uptake was calculated by multiplying dry matter of the plant with the P concentration of the plant. The micronutrients i.e. copper and zinc were determined after using Atomic Absorption Spectrophotometer (Perkin-Elmer-Model 3110 Double Beam Atomic) and were estimated in the absorption mode. Standard errors of means were calculated and when appropriate, analysis of variance was carried out and means were separated by the least significant difference (LSD) test (Sokal and Rohlf, 1981).

Results

In general, inoculation of *P. cineraria* with AM fungi resulted in better growth and nutrient uptake. It was observed in the present investigation that the nutrient concentration growth response and biomass accumulation had significantly ($P < 0.01$) different amongst the various AM fungi in six-month-old *P. cineraria* seedlings (Table

1). Infection levels ranged between 65 and 90% under different treatment. *G. aggregatum* performed the best registering the highest 90% infection followed by combined application (1:1) of *G. fasciculatum* + *G. aggregatum*. A similar trend was noticed with spore build up in the rhizosphere soil. In general, a maximum of two and a half-fold increase in the viable spores number (Table 1) was recorded. As the soil was sterilized no root colonization was found under control treatment.

The data pertaining to plant height and root length of *P. cineraria* seedlings as affected by different endophytes of AM fungi was presented in Table 1. The plant height and root length of AM inoculated seedlings were increased significantly as compared to control. The trend observed by different AM fungal isolates was *G. aggregatum* > *G. fasciculatum* + *G. aggregatum* > mixed inoculum > *G. microcarpum*.

Table 1

Plant height, Root length, Shoot and Root Dry wt, total Biomass, Root/Shoot ratio, MD (%), AM population and per cent root colonization in six months old Prosopis cineraria seedlings inoculated with different AM fungi

Treatments	Plant height (cms)	Root length (cms)	Shoot dry wt. (g)	Root dry wt. (g)	Total Biomass (g)	Root/Shoot Ratio	MD (%)	Root colonization (%)	AM population 10 g-1
T ₀ - Uninoculated	21.3	57.4	0.39	0.55	0.94	2.69	-	-	-
T ₁ - Mixed inoculum (indigenous)	26.8	66.2	0.6	0.62	1.23	2.47	23.57	84	80
T ₂ - <i>Glomus microcarpum</i> (Bangalore)	25.4	65.4	0.52	0.57	1.09	2.25	13.76	65	61
T ₃ - <i>G. aggregatum</i> (indigenous)	31.8	72	0.95	1.20	2.15	2.26	56.28	90	101
T ₄ - <i>G. fasciculatum</i> + <i>G. aggregatum</i>	28.7	67.6	0.80	0.87	1.67	2.35	43.71	86	94
LSD ($P=0.05$)	2.89	4.69	0.04	0.05	0.05	ns	3.14	4.72	4.92

Inoculum dose: 400 viable spores; ns - non significant

Shoot and root dry weight of *P. cineraria* seedlings as affected by different endophytes of AM fungi (Table 1) increased significantly as compared to control. Amongst the different AM fungi *G. aggregatum* gave significantly more shoot dry weight (0.95 g) of plants than other isolates tested. In general, there was 129% improvement in total biomass due to inoculation of *G. aggregatum* as compared to control. In the present study also root/shoot ratio (Table 1) recorded two times more in all the treatments. There was no significant difference in root: shoot ratio between the treatments. The data pertaining to total biomass of *P. cineraria* seedlings as affected by different AM endophytes (Table 1) showed AM inoculation affected plant dry weight significantly ($r = 0.917, n = 5, P < 0.01$) in *P. cineraria* seedlings as compared to control. Amongst the different inocula *G. aggregatum* gave significantly higher biomass as compared to other isolate tested.

It has been observed (Table 2) that the *G. aggregatum* resulted in highest phosphorus concentration (3.38 mg g^{-1}) in case of shoot as compared to control (uninoculated) (2.30 mg g^{-1}). Phosphorous concentration in *P. cineraria* root was

highest (2.80 mg g^{-1}) which was 27% higher than the uninoculated treatment. The other fungal isolates also performed better as compared to control.

The total uptake of P by shoot and root of *P. cineraria* plants (Table 2) was significantly affected by different AM fungal isolates. More P uptake ($3.21 \text{ mg plant}^{-1}$) was recorded in T_3 treatment, which registered more than three times increase as compared to control (uninoculated) in shoots. The trend of P uptake was observed as follows *G. aggregatum* > *G. fasciculatum* + *G. aggregatum* > indigenous inoculum (mixed) > *G. microcarpum* > control. Similarly highest P uptake by roots of *P. cineraria* plants was recorded in T_3 treatment and lowest ($1.21 \text{ mg plant}^{-1}$) phosphorus uptake in control plants (uninoculated). In general, *G. aggregatum* resulted in 2.5 fold more P uptake as compared to control plants. Total uptake of P was significantly ($r = 0.926, n = 5, P < 0.01$) affected by different AM fungal isolates. It was observed that *G. aggregatum* was superior in its ability to uptake P as compared to others. The trend observed in case of total phosphorus uptake was *G. aggregatum* ($6.57 \text{ mg plant}^{-1}$) > *G.*

Table 2

Concentration and total uptake of phosphorus in six months old *Prosopis cineraria* inoculated with different AM fungi

Treatment	Phosphorus concentration (mg g^{-1})		Phosphorus uptake (mg plant^{-1})		Total uptake (mg plant^{-1})
	Shoot	Root	Shoot	Root	
T_0 - Uninoculated	2.30	2.20	0.89	1.21	2.10
T_1 - Mixed inoculum (indigenous)	2.75	2.42	1.66	1.52	3.18
T_2 - <i>Glomus microcarpum</i> (Bangalore)	2.30	2.24	1.19	1.27	2.47
T_3 - <i>G. aggregatum</i> (indigenous)	3.38	2.80	3.21	3.36	6.57
T_4 - <i>G. fasciculatum</i> + <i>G. aggregatum</i>	2.84	2.74	2.28	2.39	4.67
LSD ($P=0.05$)	0.25	0.29	0.24	0.20	0.17

Table 3

Concentration and total uptake of copper in six months old
Prosopis cineraria inoculated with different AM fungi

Treatment	Phosphorus concentration (mg g ⁻¹)		Phosphorus uptake (mg plant ⁻¹)		Total uptake (mg plant ⁻¹)
	Shoot	Root	Shoot	Root	
T ₀ - Uninoculated	34	53	14	30	44
T ₁ - Mixed inoculum (indigenous)	51	68	31	43	74
T ₂ - <i>Glomus microcarpum</i> (Bangalore)	45	61	23	34	57
T ₃ - <i>G. aggregatum</i> (indigenous)	58	106	56	126	182
T ₄ - <i>G. fasciculatum</i> + <i>G. aggregatum</i>	53	72	43	63	106
LSD (P=0.05)	2.3	4.6	2.3	4.6	2.3

fasciculatum + *G. aggregatum* (4.67 mg plant⁻¹) > indigenous inoculum (mixed) (3.18 mg plant⁻¹) > *G. microcarpum* inoculum (2.47 mg plant⁻¹) > uninoculated control (2.10 mg plant⁻¹).

It has been observed (Table 3) that the *G. aggregatum* resulted in highest copper concentration (58 µg g⁻¹ in shoot and 106 µg g⁻¹ in root) as compared to control (uninoculated) (34 µg g⁻¹ in shoot and 53 µg g⁻¹ in root). The other fungal isolates also performed better as compared to control. Highest (56 µg plant⁻¹) copper uptake was recorded in T₃ treatment (inoculated with *G. aggregatum*), which registered more than three times increase as compared to control (uninoculated) in shoots. Total uptake of copper was significantly ($r = 0.869, n = 5, P < 0.05$) affected by different AM fungal isolates as compared to control (uninoculated). It was observed that *G. aggregatum* was superior in its ability to uptake copper as compared to others AM fungi tested. The trend observed in case of total copper uptake was *G. aggregatum* (182 µg plant⁻¹) > *G. fasciculatum* + *G. aggregatum* (106 µg plant⁻¹) > indigenous inoculum (mixed) (74 µg plant⁻¹) > *G. microcarpum* inoculum (57 µg plant⁻¹) > control (44 mg plant⁻¹).

The data (Table 4) reflected that the zinc concentration in *P. cineraria* shoot and root was positively affected by different AM fungal isolates. It was observed that the *G. fasciculatum* + *G. aggregatum* resulted in highest zinc concentration (75 µg g⁻¹) in case of shoot as compared to control (uninoculated) (49 µg g⁻¹) while zinc concentration in *P. cineraria* root was more (45 µg g⁻¹) under *G. aggregatum* treatment and lowest (25 µg g⁻¹) in control. The other fungal isolates also performed better as compared to control. Highest (65 µg g⁻¹) zinc uptake was recorded in T₃ treatment (inoculated with *G. aggregatum*), which registered nearly three times increase as compared to control (uninoculated) in shoots (Table 4). Total uptake of zinc was significantly ($r = 0.919, n = 5, P < 0.05$) correlated with AM infected plants. It was observed that *G. aggregatum* was superior in its ability to total uptake of zinc as compared to others. The trend of total zinc uptake was *G. aggregatum* (119 µg plant⁻¹) > *G. fasciculatum* + *G. aggregatum* (94 µg plant⁻¹) > indigenous inoculum (mixed) (57 µg plant⁻¹) > *G. microcarpum* (51 µg plant⁻¹) > control (32 g plant⁻¹).

Table 4

Concentration and total uptake of zinc in six months old
Prosopis cineraria inoculated with different AM fungi¹

Treatment	Phosphorus concentration (mg g ⁻¹)		Phosphorus uptake (mg plant ⁻¹)		Total uptake (mg plant ⁻¹)
	Shoot	Root	Shoot	Root	
T ₀ - Uninoculated	49	25	19	13	32
T ₁ - Mixed inoculum (indigenous)	54	41	32	25	57
T ₂ - <i>Glomus microcarpum</i> (Bangalore)	58	37	30	21	51
T ₃ - <i>G. aggregatum</i> (indigenous)	68	45	65	54	119
T ₄ - <i>G. fasciculatum</i> + <i>G. aggregatum</i>	75	39	60	34	94
LSD (P=0.05)	2.3	2.3	4.0	2.3	4.6

Discussion

The results of selection of efficient strains of AM fungal species (indigenous and non-indigenous) showed that the indigenous strains were superior to non-indigenous (Table 1). Amongst the four inocula *G. aggregatum* (indigenous) performed excellent with regards to all the parameters studied, which may be due to the variation in percentage of infection in *P. cineraria*. The AM fungal treated plants performed better in increasing the P, Zn and Cu concentration, biomass accumulation, percentage of root colonization and spore population than in un-inoculated plants. The result indicated that *P. cineraria* are highly dependent on mycorrhiza form by *G. aggregatum* (56.28%) as compared to the other mycorrhizal fungi. Although other AM fungi species were also exhibit mycorrhizal dependency towards *P. cineraria*, which varies between 13.76 to 43.71%. The data reflected that the copper concentration in *P. cineraria* shoot was significantly ($r = 0.973$, $n = 5$, $P < 0.01$) affected by different AM fungal isolates. The shoot and root copper uptake of *P. cineraria* plants (Table 3) showed that the uptake was significantly

and positively affected by inoculation of different AM fungi as compared to the control.

The results clearly showed that AM fungi help in accumulation of zinc content in *P. cineraria* plants. The uptake of zinc by shoot of *P. cineraria* plants was positively influenced by different AM fungal isolates. The efficiency of the native isolates is well known (Srivastava, 1997; Pande, 1999). The positive influence of AM fungi on other forest tree species like *Azadirachta indica* and *Acacia tortilis* (Kiran Bala *et al.*, 1989; Kalavathi *et al.*, 2000); *Dalbergia sissoo* (Khan *et al.*, 2001; Rahman *et al.*, 2003); *Acacia nilotica* (Mingqin *et al.*, 2000); *Prosopis juliflora* (Thapar *et al.*, 1991); *Populus deltoides* (Uniyal, 2003) was also available.

The mycorrhizal dependency depends on the ability to infect host plant to increase biomass production and root colonization. These differences could be attributed to the mechanism of mycorrhizal infection and development (Sanders *et al.*, 1977) or the physiological differences between AM endophytes in rate of nutrient uptake, translocation and release (Gianinazzi-

Pearson and Gianinazzi, 1983) or interaction between mycosymbionts and soil environments (Mosse, 1973). Our result suggested that *Glomus aggregatum* seemed to have a higher affinity for root colonization and greater positive response on growth and development of *P. cineraria* tree.

The contribution of AM fungi to plant nutrient uptake was mainly due to the acquisition of nutrients by the extrametrical mycorrhizal hyphae (George, 2000). Among the nutrients, P is often the key element for increased growth or fitness of mycorrhizal plants because phosphorus was transported in hyphae in large amounts compared to the plant Phosphorus demand. The results obtained from the present study (Table 2 to 4) revealed that AM fungi inoculated plants had high nutrient (P, Cu, Zn) status as compared to non-mycorrhizal plants. This may probably be due to the well established fact that AM fungal infection can markedly improve the efficiency of nutrient absorption in plants by increasing the surface area, mobilizing sparingly available nutrient sources (Tarafdar and Praveen Kumar, 1996). The results of this

investigation indicated that the increase in biomass of *P. cineraria* plants was probably due to higher nutrient uptake leading to better nutrition of *P. cineraria* plants (Tarafdar and Marcher, 1994). Likewise, accumulation of zinc and copper was also found to be high in AM inoculated fungi as compared to uninoculated plants. This could be due to the capability of AM fungi to enhance acquisition of relatively immobile nutrient as has been observed by Kothari *et al.* (1991) for zinc; Tarafdar and Praveen Kumar (1996) for copper; Clark and Zeto (2000) for zinc and copper. The mechanism leading to better acquisition of zinc and copper by the mycorrhizal roots was thought to be similar to that P (Barea *et al.*, 1987). The enhanced uptake of these nutrients as observed in the present study could also be attributed to alteration in root morphology or changes in root physiology (Macleod *et al.*, 1986). The other reason may be higher mobilization of these nutrients in soil through AM exudate (Römheld, 1987). The results of present study demonstrated that the mycorrhiza has the capacity to enhance the uptake of nutrients like P, Zn, Cu and transfer it to the host plants of *P. cineraria*.

SUMMARY

Prosopis cineraria was grown in a green house in a low phosphorus (4 mg kg⁻¹ Olsen's P) soil (Typic Camborthid) under arid environment inoculated with alone or mixed AM fungi either indigenous or from outside sources. Uptake of relatively immobile soil nutrients (P, Zn, Cu) as affected by different AM fungi was also studied. In general, significant and positive response of plant height, root and shoot biomass, root colonization and AM fungal population was observed in inoculated plants as compared to control. Inoculated plants had significantly higher P, Zn and Cu concentration as well as upto three times more total uptake. Mycorrhizal dependency to *P. cineraria* varied between 13.76 and 56.28%, which was more to *Glomus aggregatum*. In general, *G. aggregatum* was found to be the most efficient AM species for growth and nutrition of *P. cineraria* plant.

Keywords : Arbuscular Mycorrhizal fungi, Biomass Production, *Prosopis Cineraria*, *Glomus aggregatum*.

प्रोसोपिस सिनेरेरिया में पादप भार उत्पादन व फास्फोरस, कॉपर, जिंक अंतर्ग्रहण हेतु
ए.एम. स्ट्रेन की भूमिका

नीलम वर्मा, जे. सी. तरफदार व के. के. श्रीवास्तव

सारांश

शुष्क वातावरण में कम फास्फोरस (4 मिग्री/किग्रा आलसन की फास्फोरस विधि) वाली मृदा में खेजड़ी को हरित गृह में उगाकर स्थानीय या बाह्य स्रोत से एकत्रित अकेली या मिश्रित ए.एम. कवको से उपचारित किया गया। सामान्यतया कन्ट्रोल की तुलना में ए.एम. कवक से सर्वमित पौधों में प्ररोह की ऊँचाई, जड़ व प्ररोह जैव पुंज, जड़ उपनिवेशीकरण और ए.एम. कवकों की संख्या आदि के सकारात्मक और सार्थक परिणाम पाये गये।

उपचारित पौधों में फास्फोरस, जिंक और कॉपर की सांद्रता उल्लेखनीय अधिक पाई गई यहाँ तक कि सकल अंतर्ग्रहण की अपेक्षा यह तीन गुणा अधिक थी। पी. सिनेरेरिया में कवकमूल अश्रितता 13.76 से 56.28 प्रतिशत तक की पाई गई जो कि ग्लोमस एग्रीगेटम के प्रति सबसे अधिक थी। सामान्यतया ग्लोमस एग्रीगेटस को प्रोसोपिस सिनेरेरिया पौधे में वृद्धि व पोषण के लिए सबसे ज्यादा उपर्युक्त पाया गया।

References

- Barea, J. M., C. Azcon-Aguilar and R. Azcon (1987). Vesicular-arbuscular mycorrhiza improve both symbiotic N₂ fixation and N uptake from soil as assessed with a ¹⁵N technique under field conditions. *New Phytol.*, **106**: 717-725.
- Barea, J. M. (1991). Vesicular arbuscular Mycorrhizae as modifiers of soil fertility. *Adv. Soil Sci.*, **15**: 1-40.
- Clark, R. B. and S.K. Zeto (2000). Mineral acquisition by arbuscular mycorrhizal plants. *J. Plant Nutri.*, **23**: 867-902.
- Daniel Hetrick, B.A. (1984). Ecology of VA mycorrhizal fungi. In: *VA Mycorrhiza*, Ed.C.L. Powel and D.J. Bagyaraj, pp. 35-55. CRC Press, Boca Raton, FL.
- George, E. (2000). Nutrient uptake. In: *Arbuscular mycorrhizas: physiology and function*, Ed. Y. Kluwer. Academic Publishers, Dordtretch, The Netherlands, pp. 307-344.
- Gerdemann, J.W. and T.H. Nicolson (1963). Spores of mycorrhizal *Endogone* species extracted from soil by Wet Sieving and Decanting. *Trans. Brit. Myco. Soc.*, **73**: 261-270.
- Gianinazzi-Pearson and M. Gianinazzi (1983). The physiology of vesicular arbuscular mycorrhizal roots. *Plant Soil*, **71**: 197-209.
- Giovannetti, M. and B. Mosse (1980). An evaluation of techniques for measuring vesicular-arbuscular mycorrhizal infection in roots, *New Phytol.*, **84**: 489-500.
- Howeler, R.H., E. Sieverding and S. Saif (1987). Practical aspects of mycorrhizal technology in some tropical crops and pastures. *Plant Soil*, **100**: 249-283.
- Jackson, M.L. (1973). *Soil chemical analysis*. Prentice Hall of India Pvt. Ltd., New Delhi.
- Jefferies, P. (1987). Use of Mycorrhizae in agriculture. *CRC Ann. Rev. Biotech.*, **5**: 319-357.

- Jenkins, W.R. (1964). A rapid centrifugal-floatation technique for separating nematodes from soil. *Plant Dis. Rep.*, **73**: 288-300.
- Kalavathi, B.P., Santhanakrishnan and M.P. Divya (2000). Effect of VA-Mycorrhizal fungus and phosphorus solubilizing bacterium in Neem. *Indian Forester*, **126**: 67-70.
- Khan, S.N., K. Uniyal and R. Pandey (2001). Growth response of *Dalbergia sissoo* to AM and *Rhizobium* inoculations and fertilization in nursery. *Indian Forester*, **8**: 906-909.
- Kiran Bala, A.V. Rao, and J.C. Tarafdar (1989). Occurrence of VAM association in different plant species of the Indian desert. *Arid. Soil Res. Rehabil.*, **3**: 391-396.
- Kothari, S.K., H. Marschner, and V. Romheld (1991). Contribution of VA mycorrhizal hyphae in acquisition of phosphorus and zinc by maize grown in calcareous soil. *Plant Soil*, **131**: 177-185.
- Li, L., E. George and H. Marschner (1991). Extension of the Phosphorous depletion zone in a VA mycorrhizal white clover in a calcareous soil. *Plant Soil*, **136**: 441-448.
- Macleod, W.J., A.D. Robson and L. K. Abbott (1986). Effects of phosphates supply and inoculation with a vesicular arbuscular mycorrhizal fungus on the death of the root cortex of wheat, rape and subterranean clover. *New Phytol.*, **103**: 349-357.
- Menge, J.A., C.K. Labanauskus, E. L. V. Johnson and R.G. Platt (1978). Partial substitution of mycorrhizal fungi for phosphorus fertilization in the green house culture of citrus. *Soil Sci. Amer. J.*, **42**: 926-930.
- Mingqin, G., Wang-Feng Zhen, Chen-Yu, Chen-Ying Long (2000). Mycorrhizal fungal screening and inoculant effectiveness for two *Acacia* species. *Forest Res. Beijing*, **13**: 268-273.
- Mosse, B. (1973). Advances in the study of vesicular-arbuscular mycorrhizas. *Ann. Rev. Phytopathol.*, **11**: 171-196.
- Mosse, B., D.P. Stribley and F. LeTacon (1981) Ecology of mycorrhiza and mycorrhizal fungi. *Advances in Microbial Ecol.*, **5**: 137-210.
- Pande, M. (1999). Studies on VAM associations in neem based agroforestry systems in arid zone of Rajasthan. *Ph.D thesis*. FRI Deemed University, Dehradun.
- Phillips, J.M. and D.S. Hayman (1970). Improved procedure for clearing roots and staining parasitic and Vesicular Arbuscular Mycorrhizal fungi for rapid assessment of infection. *Trans. Brit. Myco. Soc.*, **55**: 158-161.
- Plenchette, C., J.A. Fortin and V. Furlan (1983) Growth response of several plants to mycorrhizae in soil of moderate P fertility. *Plant Soil*, **70**: 199-209.
- Rahman, M.S., M.A.U Mridha, S.M.N. Islam, S.M.S. Haque, P.P. Dhar and S.K. Shah (2003). Status of Arbuscular mycorrhizal colonization in certain tropical forest tree legume seedlings. *Indian Forester*, **3**: 371-376.
- Römheld, V. (1987). Existing of two different strategies for the acquisition of iron in higher plants; In; *Iron transport in microbes, plants and animals*. (Wimnken mann, G., Vander, D. hem, and J. B. Neilands, eds.) VCH Publishers, Weinhein. FRG. pp 353-374.

- Sanders, F. E., P.B.Tinker, R.I.B. Black and S. M. Palmerley (1977). The development of endomycorrhizal root system. I. Spread and infection and growth promoting effects with four species of vesicular-arbuscular endophyte. *New Phytol.*, **78**: 257-268.
- Sokal, R.R. and F J. Rohlf (1981). *Biometry The Principles and Practice of Statistics in Biological Research*. Freeman and Co., New York, p. 859.
- Srivastava, K.K. (1997). Dependency, Evaluation and Selection of Efficient Strains of VA-Mycorrhizal fungi for *Tecomella undulata* (Sm.) Seem. *Ph.D. Thesis*. JNV Univ. of Jodhpur, Jodhpur, Rajasthan.
- Tarafdar, J.C. and H. Marschner (1994). Phosphatase activity in the rhizosphere and hyphosphere of VA mycorrhizal wheat supplied with inorganic and organic phosphorous. *Soil Boil. Biochem.*, **26**: 387-395.
- Tarafdar, J.C. and Praveen Kumar (1996). The role of vesicular arbuscular mycorrhizal fungi on crop, tree and grasses grown in an arid environment. *J. Arid Environ.*, **34**: 197-203.
- Thapar, H.S., K. Uniyal and R.K. Verma (1991). Survey of native VAM fungi of sodic soils.
- Uniyal, K. (2003). Arbuscular Mycorrhizal association of *Populus deltoides*. *Indian Forester*, **4**: 527-530.
- Vijaya, T. and K.P. Srivasuki (1997). Response of forest legumes to *Glomus fasciculatum*. *J. Ind. Bot. Soc.*, **76**: 157 160.