OPEN ACCESS at journalijcir.com

Research Article ISSN: 2395 -5775

Significance of AM fungi on growth and yield of Hibiscus cannabinus L. good fiber yielding plant

Sandeepkumar.K[1*] ., K.P.Kolkar[2] ., Srinivas Naik. K[3] and Suma M.R[4]

Abstract

Present investigation to study the advancement of civilization, the use of plant fibers plants of Hibiscus cannabinus L. has gradually increased. The objective of this experiment is to compare the effectiveness of common AM fungal species like Slerocystis dussi, Acaulospora laevis, Gigaspora margarita and Glomus fasciculatum associated with fiber yielding plants and to check the efficacy of AM fungal species is best inoculation of growth and yield.

Key Words: AM Fungi, *Hibiscus cannabinus* L., *Slerocystis dussi*, *Acaulospora laevis*, *Gigaspora margarita* and *Glomus fasciculatum*

INTRODUCTION

Application of mycorrhizal biotechnology to crop production has the potential to reduce inputs such as pesticides or fertilizers and insure the sustainability of agro ecosystems (Hamel, 1996; Azcon Aguilar and Barea, 1997). As many reports have proved that AMF inoculation is effective to increase crop yield under experimental conditions (Nidchaporn, 2005 & Sato, et al., 1998). Therefore, it is necessary to select efficient AM fungi for the inoculation to the crop plants.

Arbuscular mycorrhizal association can be characterized as inducible mutualistic symbiosis involving bi-directional transfer of resources (Smith and Gianinazzi-Pearson, 1988). The plant receives minerals from fungi in return for carbon products from photosynthesis, lipids and protection (Strck *et al.,* 2003, Garg *et al.,* 2006). The AM fungi are obligate partners; while most plants are facultative (Smith and Giarl, 1988). Benefits of AM fungi to the host are numerous, growth and photosynthetic rates increases with mycorrhizal colonization in some species. Arbuscular mycorrhizal plants often have resistance to biotic and other abiotic challenges (Bayat *et al.,* 2009; Elsen *et al*., 2001; and Tonssaint *et al.,* 2007). The combined benefit to the plant leads to more vigorous productive, adaptable and competitive individuals.

The symbiosis can however, "cost" the host plant as much as 20% of its photosynthetically fixed carbon (Graham, 2000). Carbon is delivered in the form of hexose and sucrose, and the sugars are converted by the fungus, to the fungal carbohydrates trihalose and glycogen for use or strong (Strack *et al.,* 2003). Arbuscular mycorrhizal plants have increased minerals and nutrient uptake; external fungal hyphae extend beyond the root into the soil effectively scavenging soil resources which are enhanced directly to the plant root allowing access to a greater pool of resource (*Gaeg et al*., 2006).

Phosphorus nutrition is closely related to the rate of root exudation of compounds that encourage mycorrhizal colonization with phosphorus limited plants exuding greater amount of these necessary factors (Bueher *et al.,* 2009). As a result of these factors, many soils have ample amount of phosphorus, however, little is available for uptake by plants. To over come these plants have evolved multiple strategies to acquire and release inorganic phosphate from the soil (Hammad *et al.,* 2004 and Raghothama, 2005). Mycorrhizal symbiosis may be one of the most elegant and effective of these strategies. Increased growth of plants in phosphorus deficient soils can be as much as ten-fold when in symbiosis with AM fungi (Hayman and Mosse, 1971).

Arbuscular mycorrhizal (AM) fungi are one of the most interesting soil microorganisms which can solubilize fixed phosphate and retentive phosphate in soil. It was found that AM enhanced plant growth and P uptake particularly in the soil with low to moderate P level (Schenck, 1982; Bolan, 1991). In certain soils with high level of P, positive relations between AM and plant growth were also found (Kiernan *et al*., 1984.). However, many species of AM cannot enhance plant growth and P uptake of plant even in low fertile soils, mainly due to some limiting conditions

Department of Botany, Shri Annadaneshwar Arts, Science and Commerce college, Naregal
Correpondence and Reprint Requests: Sandeepkumar.K
Received: February 5 , 2016 | **Accepted:** February 25, 2016 | **Published Online:** March 28 , 2016

Conflict of interest: None declared | **Source of funding:** Nil

appearing in the soils. The quantity of available P is one of the important factors which limit the efficiency of AM. The mechanisms for acquisition of P and other mineral nutrients in their deficient soils, drought tolerance and protection of plant root from destruction by pathogen of mycorrhizal plants are described (Bolan, 1991). Therefore, the compatibility among certain soil environment, plants and AM fungal species must be realized in order to acquire appropriate benefit from AM-plant association. AM fungi are differed in their responses to phosphate application, produce different amounts of hyphae in the soil even at the same phosphate treatment (Marco, Cosme, 2011). Most of AMF species decrease their abilities on infecting plant roots with increasing phosphate application but some species can tolerate and show high activity even in high phosphate conditions (Marschner and Dell, 1994).

With the advancement of civilization, the use of plant fibers has gradually increased and their importance today is very great. The objective of this experiment is to compare the effectiveness of common AM fungal species associated with fiber yielding plants and to check the efficacy of AM fungal species.

MATERIALS AND METHODS

Soil and plant materials

The physical and chemical soil characteristics used for pot experiment were tested in soil testing laboratories at Jalavahini Management Private Limited, Dharwad district, India (Table 1). The soil was steam sterilized for one hour on two consecutive days. Fibre yielding plants seeds were collected from Seed Development Unit, University of Agricultural Sciences, Dharwad, India. Seeds were germinated in small plastic cups containing sterilized soil. Before sowing, the seeds were surface sterilized in 2% sodium hypochlorite and washed in distilled water for 2-3 times.

Table 1 The physico-chemical characteristics of soil used for experiments

PARAMETERS	RESULTS
Soil	Sandy loam
pH (1:2.5)	8.1
Conductivity (Fc) us/cm	320
Moisture (%)	4.86
Total organic carbon (%)	1.71
Nitrogen (%)	0.08
Potassium (%)	7.94
Phosphorus (%)	4.52
Magnesium (%)	0.121
Calcium (%)	0.472
Zinc (ppm)	3.86
Copper (ppm)	0.03
Manganese (ppm)	0.97
Iron (ppm)	8.24

AM fungal inoculum production

AM fungal spores were recovered from rhizosphere soil of four experimental plants by following the wet sieving and decanting method (Gerdmann and Nicolson, 1963). The spores were identified by using VAM fungal identification manual (Schenk and Perez, 1990).

All the recovered AM fungal spores belong in to six genera namely Glomus, *Sclerocystis*, *Acaulospora*, *Gigaspora*, *Scutellospora* and *Enterospora*. Among them a predominate AM fungal species such as *Glomus fasciculatum, Gigaspora margariata, Acaulospora leavis* and *Sclerocystis dussii* were selected. All the AM fungal species selected were mass multiplied by using Jawar (*Sorghum vulgare* L.) as a host plant in pots measuring 12 cm height, 24 cm diameter.

The pots contain autoclaved soil, sand mixture (3.1 v/v). Later the intensity of AM fungal root colonization was observed. After 60 days of sowing, the *Sorghum* roots showed maximum per cent colonization, from that day onwards watering has been stopped and shoot was cut at the ground level.

Then the pots were allowed to dry. The dry potting mixture containing AM fungi colonized roots AM fungal spores and mycelia was mixed thoroughly make it into homogenous mixture. This homogenous mixture having root bits AM fungal spores and mycelia propagules serve as AM fungal inoculum for further experimental studies.

Experimental design

The experimental pots were filled with growth media (soil and sand in 3:1 ratio). The soil based AM fungal inoculum (10 g) containing AMF infected root bits, mycelia and spore/sporacarps (250-300/10g inoculum) was placed as a thin layer just 2 cm below the soil surface. The seeds of all the experimental plants were surface sterilized by keeping them in 1% mercuric chloride solution for 2 to 3 min and then wash thrice with distilled water. Then these surface sterilized seeds were sown in the pre-prepared pots. The control treatment is maintained without any AM fungal inoculum the details of the treatments are as mentioned below.

Hibiscus cannabinus L.

- Uninoculated control (UIC)
- Mycorrhizal (*Slerocystis dussi*) inoculated
- Mycorrhizal (*Acaulospora laevis*) inoculated
- Mycorrhizal(*Gigaspora margarita*) inoculated
- Mycorrhizal (*Glomus fasciculatum*) inoculated

Table 2 Effect of AM fungi on growth parameters of *Hibiscus cannabinus* L. , phosphorus uptake in shoot and per cent mycorrhizal colonization, spore number at 60, 90 and 120 days after sowing.

Treatments	SL	FWS	DWS	RL	FWR	DWR	STD	PC	NFL	Fiber yield	SP	P-uptake
60 DAYS												
Uninoculated (UN)	18.50 ±0.057e	4.300 ±0.057e	1.300 ±0.057e	7.366 ±0.088e	1.633 ±0.033e	0.926 ±0.006e	1.233 ±0.033e	0.000 ±0.000e	0.000 ±0.000	0.000 ±0.000	0.000 ±0.000e	0.050 ±0.000e
Slerocystis dussii (SD)	23.66 ±0.088d	12.40 ±0.057d	3.833 ±0.033d	10.33 ±0.088d	2.366 ±0.120d	1.333 ±0.088d	1.433 ±0.033d	34.33 ±1.453d	0.000 ±0.000	0.000 ±0.000	71.33 ±1.763d	0.080 ±0.000d
Acaulospora laevis (AL)	26.53 ±0.145c	13.46 ±0.066c	4.600 ±0.650c	12.40 ±0.057c	2.600 ±0.115c	1.600 ±0.000c	1.766 ±0.033c	39.66 ±0.333c	0.000 ±0.000	0.000 ±0.000	82.33 ±2.185c	0.08 ±0.000c
Gigaspora margarita (GM)	29.46 ±0.145b	14.53 ±0.088b	4.533 ±0.176b	13.43 ±0.033b	2.766 ±0.033b	1.733 ±0.033b	1.633 ±0.066b	41.00 ±0.577b	0.000 ±0.000	0.000 ±0.000	97.33 ±1.763b	0.090 ±0.000b
Glomus fasciculatum (GF)	36.56 ±0.120a	17.70 ±0.152a	6.966 ±0.120a	15.70 ±0.057a	3.133 ±0.066a	1.933 ±0.033a	2.066 ±0.033a	51.66 ±1.453a	0.000 ±0.000	0.000 ±0.000	127.6 ±1.333a	0.110 ±0.000a
90 DAYS												
Uninoculated (UN)	28.76 ±0.088e	10.40 ±0.057e	3.366 ±0.088e	10.73 ±0.120e	4.233 ±0.133e	1.766 ±0.088e	1.533 ±0.066e	0.000 ±0.000e	0.000 ±0.000	0.000 ±0.000	0.000 ±0.000e	0.070 ±0.000e
Slerocystis dussii (SD)	37.96 ±0.233d	23.56 ±0.202d	8.266 ±0.409d	13.63 ±0.088d	6.266 ±0.120d	2.700 ±0.152d	1.833 ±0.066d	47.00 ±0.577d	0.000 ±0.000	0.000 ±0.000	102.0 ±1.527d	0.100 ±0.000d
Acaulospora laevis (AL)	40.73 ±0.120c	28.66 ±0.120c	18.43 ±0.120c	14.90 ±0.404c	7.566 ±0.120c	3.400 ±0.115c	1.866 ±0.088c	53.66 ±0.881c	0.000 ±0.000	0.000 ±0.000	115.3 ±1.763c	0.110 ±0.000c
Gigaspora margarita (GM)	43.76 ±0.088b	30.76 ±0.088b	12.96 ±0.233b	17.06 ±0.166b	8.466 ±0.176b	3.866 ±0.176b	1.900 ±0.057b	59.66 ±1.201b	0.000 ±0.000	0.000 ±0.000	125.3 ±2.027b	0.130 ±0.000b
Glomus fasciculatum (GF)	64.86 ±0.033a	40.30 ±0.602a	23.50 ±1.732a	23.43 ±1.097a	14.56 ±1.126a	7.833 ±0.876a	2.066 ±0.033a	74.33 ±1.201a	0.666 ±0.000a	0.000 ±0.000	150.6 ±3.844a	0.150 ±0.000a
120 DAYS												
Uninoculated (UN)	41.56 ± 0.202e	18.63 ±0.218e	5.600 ±0.300e	12.76 ±0.088e	5.066 ±0.033e	2.366 ±0.260e	1.700 ±0.152e	0.000 ±0.000e	0.666 ±0.333e	2.100±0.115e	0.000 ±0.000e	0.090 ±0.000e
Slerocystis dussii (SD)	53.36 ±0.622d	34.66 ±0.120d	17.76 ±0.088d	21.63 ±0.088d	9.700 ±0.404d	4.100 ±0.251d	2.066 ±0.088d	62.00 ±1.154d	0.666 ±0.333d	3.267±0.066d	133.3 ±4.255d	0.120 ±0.000d
Acaulospora laevis (AL)	61.83 ±0.033c	37.80 ±0.351c	17.33 ±0.120c	23.56 ±0.120c	10.60 ±0.321c	4.766 ±0.088c	2.100 ±0.057c	74.66 ±1.763c	1.333 ±0.333c	4.400±0.115c	144.3 ±2.728c	0.130 ±0.000c
Gigaspora margarita (GM)	64.80 ±0.057b	40.76 ±0.088b	21.10 ±0.472b	26.56 ±0.120b	12.96 ±0.393b	5.400 ±0.057b	2.166 ±0.066b	74.66 ±3.179b	2.000 ±0.577b	5.600±0.115b	158.6 ±4.910b	0.150 ±0.000b
Glomus fasciculatum (GF)	78.70 ±0.152a	46.50 ±0.173a	25.06 ±0.721a	30.96 ±0.185a	17.26 ±0.417a	7.866 ±0.185a	2.266 ±0.088a	89.66 ±1.201a	3.333 ±0.333a	6.733±0.371a	196.3 ±0.881a	0.180 ±0.000a

Note: SL: Shoot length, FWS: Fresh weight of Shoot, DWS: Dry weight of Shoot, RL: Root Length, FWR: Fresh weight of Root, DWR: Dry weight of Root, STD: Stem Diameter, PC: per cent of mycorrhizal colonization, NFR: Number of fruits, SP: Spore Number, P: Phosphorous. Values represent the mean ± SD. Means followed by the same letter within a column are not significantly P= 0.05 according to DMRT

All the experimental pots were arranged incompletely randomized block design with triplicate per treatment. The experimental pots were kept free of weeds, insects, pets, rodents etc. the pots were watered every alternate day and 10 ml of Hoagland solution without P was given to each seedling at the interval of 15 days.

Analysis of growth parameters

Plants were harvested after 60, 90 and 120 days after sowing. The plants parameters like shoot and root length, fresh weight of shoot and root, shoot and root dry weight, stem diameter and number of leaves, the per cent root colonization, spore number per 50 g soil, and phosphorus uptake in shoot were recorded. After the harvest, experimental plants shoot and root was oven dried at 70^0C until a constant weight was obtained to determine the dry weight.

Determination of Mycorrhizal Root colonization

The per cent root colonization was evaluated microscopically followed by clearing of roots in 10% KOH and staining with 0.05% trypan blue in lactophenol according to method described by Phillips and Hayman (1970). The following formula was used to calculate the root colonization (Giovannetti and Mosse, 1980).

$$\text{Percent mycorrhizal colonization} = \frac{\text{Number of root segments colonized}}{\text{Total number of root segments examined}} \times 100$$

Determination of AM fungal spores

Spores were separated from the soil by wet sieving and decanting technique (Gerdemann and Nicolson, 1963). 50 g of soil was mixed with water. The mixture was pour through different sieve size (250, 106, 45µm). After, several times of sieve washing the supernatant was collected in Petri dish and spores counted under binocular-microscope.

Determination of Fiber content

Smallholder plots are usually harvested by hand. The plants are cut at 2 to 3 cm above the soil and left on the ground to dry. The cut *Hibiscus cannabinus* L., is laid in swathes to dry for up to four days. This was

followed by water retting (the bundled hemp floats in water).

Phosphorus content

The phosphorus content in the shoots was determined by the vanado-molybdate phosphoric acid yellow color method outlined by Jackson (1973).

Statistical Analysis

Analysis of variance (ANOVA) was performed on all data and the means were separated using Duncans multiple Range Test (DMRT), with the help of SPSS student version-9 software.

RESULTS

The different AM fungi such as *Glomus fasciculatum, Gigaspora margarita, Acaulospora laevis* and *Sclerocystis dussii* were inoculated to test their efficacy on four fiber yielding plants. All the plant species inoculated with different AM fungi showed increased growth parameters over the control plants (Plate 1). The experimental results revealed that, not only the growth parameters of four experimental plants were increased but also the nutrient uptake and mycorrhizal status was significant compared to non-mycorrhizal plants. The growth parameters of all the experimental plants were determined at 60, 90 and 120 days after sowing. Initially slowly increased growths were observed but after 95 days significantly increased growth rate had been recorded. The greater values for growth parameters shoot length, fresh weight of shoot, dry weight of shoot, root length, fresh weight of root, dry weight of root, stem diameter, number of flowers, numbers of fruits and increased "P" uptake were recorded in experimental plants with inoculation AM fungus *Glomus fasciculatum* over the reaming treatments. Where as, plants inoculated with *Gigaspora margarita* and *Sclerocystis dussii* have shown to the less but they have significant values when compared to control plants. The intermediate growth rate had been recorded in plants inoculated with AM fungus *Acaulospora laevis*. It indicates that, the AM fungus *Acaulospora laevis* was the second best efficient indigenous AM fungus for the fiber yielding plants. (Table. 2.1-2.4).

Mycorrhizal parameters like per cent colonization, and spore number were determined at 60, 90 and 120 days. The mycorrhizal root colonization was found to be varied in each experimental plant it was less in beginning (at 45-60 days) but steadily increased after 90 days. It was observed that at 120 there was maximum colonization. Inoculated with different AM fungi the maximum amount of PMC was recorded in plants inoculated with *Glomus fasciculatum* and least was in plants roots inoculated with *Sclerocystis dussii*.

AM fungal spore number was recorded in all experimental plants. It was found to be highest at 120 days and least was noticed at 60 days, with increase in duration the spore number was increased. Maximum AM fungal spore number was observed in the rhizosphere soils of the experimental plants inoculated with *Glomus fasciculatum* and it was least in plants inoculated with *Slerocystis dussii*, whereas,, moderate spore number was noticed in plants inoculated with *Acaulospora laevis* and *Gigaspora margarita*. Among the plant species, the maximum number of AM fungal spores was found in the rhizospheric soils.

The plants were also analyzed for its nutrient continent in shoot, particularly phosphorus. All the AM fungal inoculated plants have shown increased nutrient content when compared to control plants. Maximum increased P uptake was observed in plants inoculated with *Glomus fasciculatum*. The moderately increased P Uptake in shoots was estimated in plants inoculated with *Gigaspora margarita* and it was least in plants inoculated with *Acaulospora laevis* and *Sclerocystis dussii*. Among all mycorrhizal inoculated plants, *Hibiscus cannabinus* L. had shown significantly increased P uptake over the reaming three fiber yielding plants. The least increased P uptake was reported in mycorrhizal *Gossypium hirsutum* L., whereas, the moderate P uptake was estimated in the reaming two plant species.

The fiber content in all the inoculated and control plants was measured. The fiber content in the mycorrhizal plant was greater when compared to control plants (Fig. 2.11). Among all the mycorrhizal plants, the plants inoculated with *Glomus fasciculatum* have shown maximum fiber yield. The least increased fiber content was recorded in plants inoculated with *Sclerocystis dussii*. It can be evident from the above results that, AM fungus *Glomus fasciculatum* was found more efficient and the next best species for the inoculation to the fiber yielding plants was *Acaulospora laevis*. Fiber yield was considerably more in inoculated plants. All the experimental plants have shown increased fiber yield, but with *Glomus fasciculatum* fiber yield was maximum.

CONCLUSION

The different AM fungi such as *Glomus fasciculatum, Gigaspora margarita, Acaulospora laevis* and *Sclerocystis dussii* were selected based on their abundance in the rhizosphere of the experimental plants. These AM fungal species inoculated to test their efficacy on four fiber yielding plants. The

greater values for growth parameters shoot length, fresh weight of shoot, dry weight of shoot, root length, fresh weight of root, dry weight of root, stem diameter, number of flowers, numbers of fruits and increased "P" uptake were recorded in experimental plants with inoculation AM fungus *Glomus fasciculatum* over the reaming treatments. The plants inoculated with *Glomus fasciculatum* have shown maximum fiber yield. The least increased fiber content was recorded in plants inoculated with *Sclerocystis dussii*. It can be concluded that, AM fungus *Glomus fasciculatum* was found to be more efficient for the growth and yield of the experimental plants and the next best species for the inoculation to the fiber yielding plants was *Acaulospora laevis*.

References

1. Azcon-Aguilar, C. and Barea, J. M. (1997). Interactions between mycorrhizal fungi and other rhizosphere microorganisms. In: M. J. Allen (ed.), *Mycorrhizal functioning: an integrative plant-fungal process*. Chapman and Hall, New York, N. Y. pp: 163-198.
2. Bayat, F., Mirlohi, A and Khodambashi, M (2009). Effects of endophytic fungi on some drought tolerance mechanisms of tall fescue in a hydroponics culture. *Russian journal of plant physiology*. 56:510-516.
3. Bolan, N. S. (1991). A critical review on the role of mycorrhizal fungi in the uptake of phosphorus by plants *Plant and Soil,* 13: 187–207
4. Bucher, M., Wegmuller, S and Drissner, D (2009).Chasing the structure of small molecules in arbuscular mycorrhizal signaling. Current opinion in plant Biology. 12:500-507.
5. Elsen, A., Declerk, S and Dewalle, D (2001). Effects of Glomus intradices on the reduction of the burrowing nematode (*Radopholus simitis*) in dixenic culture. *Mycorrhiza*. 11:49-51.
6. Gaeg, N ., Kaur, G and Kaur, A (2006). Arbuscular mycorrhiza nutritional aspects. *Archives of Agronomy and soil science*. 52:593-606.
7. Graham, J. H (2000). Assessing costs of arbuscular mycorrhizal symbiosis In:Podila G.K and Douds D.D (Eds). Current advances in mycorrhizae research St.Paul M.V.Ns press. Pp.127-140.
8. Hamel, C (1996). Prospect and problems pertaining to the management of Arbuscular mycorrhizae in agriculture. *Agric. Ecosys. Environ*. 60:197-210.
9. Hammod, J.P., Broadley, M.R and White, P.J (2004). Genetic responses to phosphorus deficiency. Annalea.Botonici.Fennici.94:323-332.
10. Hayman, D.S and Mosse, B (1971). Plant growth responses to vesicular arbuscular mycorrhiza; growth of endogone inoculated plants in phosphate deficient soils. *New phytologist*. 70:19-27.
11. Kiernan, J. M., Hendrix, J. W. and Maronek, D. M. (1984). Fertilizer-induced pathogenicity of mycorrhizal fungi to sweetgum seedlings. *Soil Biol. Biochem,* 15: 257-262.
12. Marco, Cosme., Michael, J., Stout. and Susanne. (2011). Wurst Effect of arbuscular mycorrhizal fungi (Glomus intraradices) on the oviposition of rice water weevil (Lissorhoptrus oryzophilus). *Mycorrhiza,* 21:651–658.
13. Marschner, H. and Dell, B. (1994). Nutrient uptake in mycorrhizal symbiosis. *Plant soil*, 159: 89-102.
14. Nidchaporn, N (2005). Effect of logarithm fertilization on diversity of Arbuscular mycorrhizal fungi under a cropping system in Thailand. Ph.D . thesis Kasetsart University. Thailand pp: 185 .
15. Raghothama, K.G (2005). Phosphorus. In :Broadly, M.R and White, P.J.(Eds). *Plant nutritional genomics. Blackwell,* Oxford.U.K. pp: 112-126.
16. Sato, T., Kume, T and Hirata, H (1998). Effect of *Glomus etunicatum* on soy bean (*Glycine max*. Merr).growth nitrogen and phosphorus nutrition and soybean nodule activities under different phosphorus application levels in an Andosol sub soil. *Jpn.J.Soil.Sci.Plant .Nut*. 69:62-72.
17. Schenk, N. C. and Perez, Y. (1990). Manual for the identification of VA-mycorrhizal fungi, 3rd edition. Gaines Ville, Florida, University of Florida. Pp:249.
18. Smith , S.E and Gianinazzi-Pearson, V (1988). Physiological interactions between symbionts in Vesicular- Arbuscular mycorrhizal plants. Aunual Review of plant physiology and plant molecular biology. 39:221-244.
19. Stract, D., Festa, T., Hause, B., Sc Wiemann W and Watter, M.H (2003). Arbuscular mycorrhiza biological, chemical and molecular aspects. *Journal and chemical Ecology,* 29:9-13.
20. Stract, D., Festa, T., Hause, B., Sc Wiemann W and Watter, M.H (2003). Arbuscular mycorrhiza biological, chemical and molecular aspects. *Journal and chemical Ecology,* 29:9-13.
21. Toussaint, J.P., Smith, F.A and Smith, S.E (2007). Arbuscular mycorrhizal fungi can induce the production of phytochemical in sweet basil irrespective of phosphorus nutrition. *Mycorrhiza*. 17:291-297.