



## Ability of arbuscular mycorrhiza to promote growth of maize plant and enzymatic activity of an alluvial soil

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**Abstract:** A pot experiment was conducted to evaluate the response of selected species of mycorrhizae for root colonization and phosphorus uptake by maize in an alluvial soil. Of all the species of mycorrhizae taken under consideration, *Glomus mosseae* was found to perform better in terms of root colonization, number of spores, grain yield and phosphorus uptake. The maximum plant height (28.5 cm), shoot dry weight (19.45 g plant<sup>-1</sup>) and root dry weight (4.77 g plant<sup>-1</sup>) was also found with the application of *G. mosseae*. Its application significantly increased the root dry weight by 99.58 and 72.82% over application of *G. intraradices* and control respectively, and was at par with the application of *G. coronatum* and *Gigaspora decipiens*. Application of *G. decipiens* reported the highest bacterial (39.11 cfu g<sup>-1</sup> soil) and fungal count (30.68 cfu g<sup>-1</sup> soil) that was found to be at par with application of *G. mosseae*. Application of *G. mosseae* significantly increased the actinomycetes population by 44.71 and 55.97% over application of a local mycorrhizal strain and control. Maximum dehydrogenase activity (56.00 g<sup>-1</sup> TPF g<sup>-1</sup> 24 h<sup>-1</sup>) and acid phosphatase activity (0.299 mg PNP g<sup>-1</sup> h<sup>-1</sup>) and was also observed with application of *G. mosseae*, which in turn resulted in higher yield which was 27.28%, 28.52%, 9.35 and 11.7% more than *G. intraradices*, *G. coronatum*, *G. decipiens* and the local species respectively. *G. mosseae* inoculation proved to be effective in modifying the soil microbe population and community structure and also in enhancing the soil enzymatic activities and phosphorus uptake of the crop.

**Keywords:** Alluvial soil, Grain yield, Maize, Mycorrhiza, Phosphorus

### INTRODUCTION

Mycorrhiza is a mutualistic symbiosis between certain groups of soil fungi and most plant root systems (Schubler *et al.*, 2001 and Hata *et al.*, 2010). The most publicized benefit of mycorrhiza is the improved growth rate which is mainly due to enhanced phosphorus (P) nutrition. Various mechanisms (e.g. exploration of large soil volume, faster movement of mycorrhizal hyphae and solubilization of soil phosphorus) are responsible for increasing the uptake of phosphorus by mycorrhizal plants. Non-nutritional benefits to plants, such as changes in water relations, phytohormone levels, carbon assimilation, secretion of enzymes, increased microbial count in soil, etc. have also been reported, but they are difficult to interpret. Other important roles of mycorrhiza in ecosystems include nutrient cycling (Andrade *et al.*, 1998).

Phosphorus is one of the major essential macronutrients which limit plant growth owing to its low bioavailability in soils (Feng *et al.*, 2004). Improving plant acquisition of P from soil is an obvious alternative for the management of those low P soils (Zhu *et al.*, 2003). It is commonly known that arbuscular mycorrhizae (AM), they act as a direct link between soil and roots, AM fungi help plants to capture water and nutrients (especially P) from the soil, and in return, the plant provides the fungus with relatively constant and

direct access to carbohydrates (Smith and Read, 2008), which are translocated from their source to root tissue and on to fungal partners.

Soil enzymatic activities regulate the various indices of soil fertility, soil productivity and soil quality (Busto and Perez-Mateos, 1997). AM fungi can increase soil enzyme activities, such as phosphatase (Mar Vázquez *et al.*, 2000), dehydrogenase, urease, protease and  $\beta$ -glucosidase (Caravaca *et al.*, 2004). Mar Vázquez *et al.* (2000) reported mycorrhizal colonization induced qualitative changes in the microbial population and enzyme activities in the rhizosphere of maize plants. On the other hand, soil phosphatase and urease are closely related to the P and N nutrition of plants. Thus, the enhancement of soil enzyme activities is one of the physiological and biochemical mechanisms involved in a mycorrhization effect on plant mineral nutrition. Rao and Tak (2001) found that mycorrhizal fungal inoculation resulted in enhanced plant growth, total uptake of N, P and many other nutrients, activities of dehydrogenase, phosphatases and nitrogenase in the rhizosphere in gypsum mine spoil. Owing to the energy and cost-intensive manufacture of chemical fertilizers, use of microbial inoculants to supplement a part of phosphorus requirement has attained immense importance. To get maximum agricultural benefit, inoculation of the soil with suitable type of AM fungi is nec-

essary. In view of the above-mentioned possibilities, a pot experiment was conducted for screening the VAM species with respect to their effect on the growth of maize plant and its nutrient acquisition.

## MATERIAL AND METHODS

The present study was undertaken to screen the AM fungi for maize crop during the *rabi* season of 2013-14 with a promising var. DHM-117, at Bihar Agricultural University, Sabour, Bhagalpur, India. Inoculums of the five AM species viz., *Glomus mosseae*, *Glomus coronatum*, *Glomus intraradices*, *Gigaspora decipiens* and *Gigaspora margarita* were commercial products of The Energy Resource Institute (TERI), New Delhi, India. The products consisted of fragments of colonized roots and spores of AM fungi in a vermiculite substrate. One uncharacterized local inoculum (control) was collected from the maize and litchi farm of Bihar Agricultural University, Sabour. The substrate used for the experiment consisted of soil from the Bihar Agricultural University, research farm and river bed sand of the Ganges (w/w, 3:1). The soil was collected from the surface (0–15 cm) and passed through a 2.00 mm aperture sieve to remove roots and debris. The river bed sand was thoroughly washed with tap water to remove salt. The substrate mixture was completely sterilized by autoclaving over 1 hr with step-wise increase in temperature till the centre reached 120°C (kept for 30 minutes). The substrate used for the pot experiment was loamy sand in texture, having a pH of 7.2 and EC of 0.22 dS m<sup>-1</sup>. The organic carbon content of the substrate was 0.56%, and the available nitrogen, phosphorus and potassium content was found 180.77, 25.89 and 220.66 kg ha<sup>-1</sup>, respectively.

Seeds were surface-sterilized by treatment with a 1:1 mixture of H<sub>2</sub>O<sub>2</sub> and absolute ethanol for 2 minutes followed by a treatment with 0.05% HgCl<sub>2</sub> for 1 minute. The sterilizing agents were drained aseptically, and the seeds were washed for 10-12 times in sterile distilled water to remove all traces of the chemicals. Earthen pots of 15 cm height and 30 cm diameter were filled with 10 kg of sterilized substrate. All AM species were maintained in the pots with five replications each. The following treatment structure was formulated for the study: T<sub>1</sub>- *G. mosseae*, T<sub>2</sub>- *G. coronatum*, T<sub>3</sub>- *G. intraradices*, T<sub>4</sub>- *G. decipiens*, T<sub>5</sub>- *G. margarita*, T<sub>6</sub>- Local (uncharacterized inoculum) and T<sub>7</sub>- control (without inoculum). About 5 g of the AM inoculum source (containing and 8-10 spores g<sup>-1</sup>) was mixed with the upper 4 cm of the substrate in each pot. In each pot, 4 sterile seeds of maize (var. DHM 117) were planted. Once in every 15 days, each pot was treated with 20ml of Hoagland solution minus phosphate (Hoagland and Arnon, 1938). One plant from each pot was uprooted after 75 days of sowing. After measuring the shoot height they were kept in a hot air oven to dry at 105°C for 72 hours until they attained a constant weight.

Samples of roots of the plants with adhering soil were collected at 75 days after sowing (DAS). They were washed repeatedly with sterilized distilled water and fragmented into small segments of 1 cm. The root segments were cleared in 10% KOH and stained with 0.5% Trypan blue by the method given by Phillips and Hayman (1970). The stained bits were examined and the Arbuscular mycorrhizal colonization in the roots was recorded in terms of per cent root segments showing mycorrhiza formation. The population of AM spores in the rhizospheric soil was estimated by extracting the spores from the root by the washing-sieving-decanting method of Gerdemann and Nicolson (1963). They were examined stereomicroscopically and population was computed in terms of number per 100 g of dry soil. Phosphorus concentration in straw and grain were determined by employing the vanadomolybdate yellow colour method given by Jackson (1973).

Rhizosphere samples were obtained by collecting the soil adhering to the roots. The 10 g of soil samples were placed in an Erlenmeyer flask containing 90 ml of sterilized distilled water, and shaken for 30 min. Ten-fold series dilutions were prepared, and appropriate dilutions were plated in specific media. For the isolation of bacteria, fungi and actinomycetes, the Plate Count Agar, Czapek-Dox Agar (Thom and Raper, 1945) and Kenknight and Munaier's Medium, respectively were used. The numbers of colony forming cells were determined in each plot by serial dilution pour plate method (Subba, 1986). The activities of three soil enzymes: dehydrogenase activity (Casida *et al.*, 1964), acid phosphatase and alkaline phosphatase (Tabatabai and Bremner, 1969) were determined. Analysis of variance (ANOVA) was performed as described by Gomez and Gomez (1984) to determine the effects of various treatments. Critical difference (CD) at 5% level of probability and P values was used to examine differences among treatment means.

## RESULTS

**Colonization and number of spores:** The occurrence and intensity of root colonization in maize are presented in table 1. All the five species including the local species of AM fungi colonized the roots of maize. The root colonization was significantly higher with inoculation of *G. mosseae* over all the given treatments. The treatment having application of *G. coronatum* significantly increased root colonization by 22.83, 50.02, 31.66 and 195.23% and the number of spores per 100 g of soil by 30.94, 24.66, 42.52 and 592.57% over *G. intraradices*, *G. margarita*, local and control, respectively. Similarly, *G. decipiens* significantly increased mycorrhizal infection in the root by 35.26, 18.71 and 175.00% more than *G. margarita*, local and control, respectively (Table 1). The maximum mycorrhiza infection (78%) and number of spores (311.66 per 100 g of soil) were reported with

**Table 1.** Effect of arbuscular mycorrhizal fungi on plant growth, colonization and spore formation under maize Rhizospheric soil at 75 DAS.

Treatment	AM infection (%)	Number of spore (100 g <sup>-1</sup> of soil)	Plant height (cm)	Shoot dry weight (g plant <sup>-1</sup> )	Root dry weight (g plant <sup>-1</sup> )
<i>G. mosseae</i>	78.00	311.66	28.50	19.45	4.77
<i>G. intraradices</i>	49.66	238.00	22.60	14.45	2.39
<i>G. coronatum</i>	61.00	301.66	24.80	16.98	4.66
<i>G. margarita</i>	40.66	250.00	20.16	13.98	3.07
<i>G. decipiens</i>	55.00	275.66	21.87	18.87	4.68
Local	46.33	221.66	22.67	13.44	3.99
Control	20.00	45.00	15.89	12.47	2.76
SEm (±)	3.50	24.99	1.48	1.98	1.20
C.D. (P=0.05)	7.48	47.61	3.34	4.83	2.00

**Table 2.** Effect of AM fungi on soil biological properties under maize rhizosphere at 75 DAS.

Treatment	Bacteria (cfu×10 <sup>6</sup> )	Actinomycetes (cfu×10 <sup>6</sup> )	Fungi (cfu×10 <sup>4</sup> )	DHA (g TPF g <sup>-1</sup> 24 h <sup>-1</sup> )	Acid phosphatase (mg PNP g <sup>-1</sup> h <sup>-1</sup> )	Alkaline phosphatase (mg PNP g <sup>-1</sup> h <sup>-1</sup> )
<i>G. mosseae</i>	38.97	19.45	27.77	56.00	0.299	0.555
<i>G. intraradices</i>	30.29	14.45	22.39	45.67	0.157	0.430
<i>G. coronatum</i>	36.33	16.98	25.66	49.98	0.217	0.565
<i>G. margarita</i>	33.66	13.98	16.07	39.75	0.248	0.433
<i>G. decipiens</i>	39.11	18.87	30.68	44.87	0.31	0.546
Local	30.29	13.44	18.99	39.76	0.205	0.457
Control	15.89	12.47	11.76	12.67	0.101	0.201
SEm (±)	1.48	1.98	3.39	2.88	0.035	0.05
C.D. (P=0.05)	4.34	5.83	9.95	5.16	0.089	0.09

**Table 3.** Response of maize to AM fungi for yield and yield attributes.

Treatment	Number of cob (plant <sup>-1</sup> )	Grain yield (g plant <sup>-1</sup> )	Biological yield (g plant <sup>-1</sup> )	100-seed weight (g)
<i>G. mosseae</i>	1.33	64.66	358.33	26.50
<i>G. intraradices</i>	1.33	50.8	315.33	23.00
<i>G. coronatum</i>	1.33	63.15	349.33	23.50
<i>G. margarita</i>	1.33	50.31	329.66	23.00
<i>G. decipiens</i>	1.00	59.13	330.66	25.07
Local	1.33	57.78	326.66	22.50
Control	1.00	46.86	281.33	22.00
SEm (±)	-	9.80	-	-
C.D. (P=0.05)	NS	5.04	NS	NS

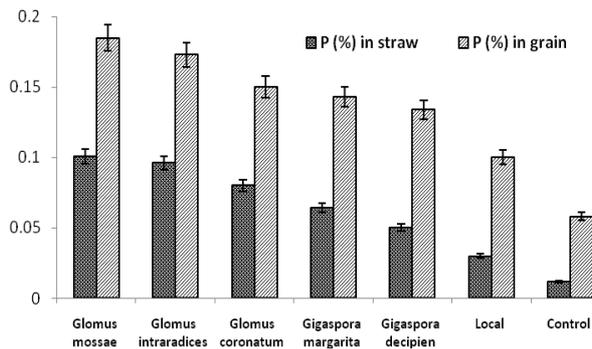
application of *G. mosseae*. Application of *G. decipiens* significantly increased the number of spores by 24.06 and 512.57% over application of local strain of mycorrhiza and control. *G. coronatum* gave 26.4%, 20.66% and 36.09% significantly higher number of spores than *G. intraradices*, *G. margarita* and local respectively. There is an absolute increase of 37 spores in case of *Gigaspora decipiens*, when we compare it with the number of *G. intraradices* (Table 1). Due to the favorable soil conditions for the development of *G. mosseae*, inoculation with *Glomus mosseae* gave significantly higher colonization as compared to *G. intraradices*, *G. coronatum*, *G. margarita*, *G. decipiens* and local.

**Plant growth:** Plant growth parameters like plant height (cm), shoot dry weight (g) and root dry weight increased with the inoculation of all the mycorrhiza fungi over non-inoculated control treatment. The maximum plant height (28.5 cm), shoot dry weight (19.45 g plant<sup>-1</sup>) and root dry weight (4.77 g plant<sup>-1</sup>) was found with the application of *G. mosseae*. The application of

*G. coronatum* also gave 23.01 and 56.07% significantly higher plant height over inoculation of *G. margarita* and control. Plant height increased numerically by 12.10 and 3.33% with the application of *G. intraradices* over *G. margarita* and *G. decipiens*. Similarly, application of *G. decipiens* significantly increased shoot dry weight by 34.97, 40.40 and 51.32% over application of *G. margarita*, local and control, respectively (Table 1). A numerical increment of 17.50, 21.45, 26.33 and 36.16% was found in shoot dry weight when compared with application of *G. intraradices*, *G. margarita*, local and control, respectively. The application *G. mosseae* also significantly increased the root dry weight by 99.58 and 72.82% over application of *G. intraradices* and control and at par with *G. coronatum* and *G. decipiens*.

#### Biological properties of rhizospheric soil

**Microbial population:** All the applied mycorrhizal species increased the microbial population over control (Table 2). Application of *G. decipiens* gave signifi-

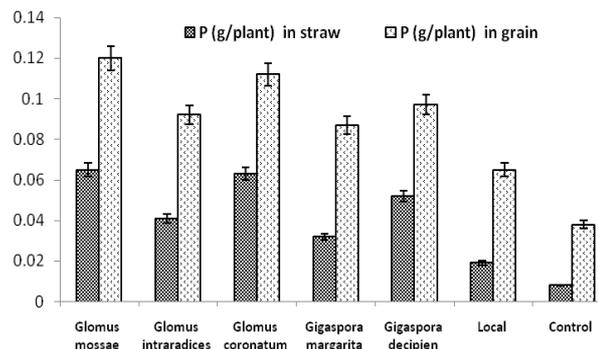


**Fig. 1. a** Effect of AM fungi on Phosphorus (P) concentration.

cantly 29.11, 16.19, 29.11 and 146.12% increased bacterial count over *G. intraradices*, *G. margarita*, local and control which is at par with the application of *G. mosseae*. Similar trend were observed in the population of fungi. Application of *G. mosseae* significantly increased the actinomycetes population by 44.71 and 55.97% over application of the local strain of mycorrhiza and control. Similarly application of *G. decipiens* significantly increased the actinomycetes population by 51.32% than control. The maximum fungi population (30.68 cfu g<sup>-1</sup> soil) was observed with application *G. decipiens*, which was 90.91, 61.55 and 160.88% over *G. margarita*, local and control, respectively. The fungi population was found significantly higher (118.19%) with the application of *G. coronatum* over control. Similarly, *G. mosseae* also significantly increased the fungi population by 44.71 and 136.13% over *G. margarita* and control.

**Soil enzymatic activity:** The soil enzymatic activity was analyzed after 75 days of sowing and the results (Table 2) showed that there is an increment in enzymatic activity with application of mycorrhiza as compared to the control treatment. Maximum enzymatic activity viz. dehydrogenase activity (56.00 g<sup>-1</sup> TPF/g/24 hrs) and acid phosphatase activity (0.299 mg PNP/g h) and was observed with application of *G. mosseae*. The maximum alkaline phosphatase activity (0.565 mg PNP/g h) was found with application of *G. coronatum*. Application of *G. coronatum* significantly increased the alkaline phosphatase activity by 9.43, 25.73 and 294.47% and Dehydrogenase activity by 31.39, 30.48 and 181.09% over application of *G. intraradices*, *G. margarita* and control respectively. Similarly, application of *G. mosseae* significantly increased the acid phosphatase activity by 90.44, 45.85 and 196.03% and alkaline phosphatase activity by 29.06, 21.44 and 176.11% over *G. intraradices*, local and control, respectively. Application of *G. decipiens* also gave significant increment by 26.97, 26.09 and 171.64% over *G. intraradices*, *G. margarita* and control. This treatment also significantly increased the acid phosphatase activity by 42.85, 51.21 and 206.93% over *G. coronatum*, local and control, respectively.

**Phosphorus content and uptake by grain and straw**  
**P content in grain and straw:** The maximum concen-



**Fig. 1. b** Effect of AM fungi on P uptake by straw and grain.

tration of P in straw (0.101%) and grain (0.120%) was found with treatment of *G. mosseae*. This treatment also significantly increased the P content in straw and grain over all the treatments. Application of *G. coronatum* also significantly increased P content in straw by 50, 92, 220 and 700% over *G. margarita*, *G. decipiens*, local and control, respectively (Fig. 1a). The similar trend was observed for P content in grain. Application of *G. intraradices* significantly increased P content in grain by 73 and 198.27% over local and control. Similarly, application of *G. coronatum* also numerically increased the P content in grain by 4.89, 11.94, 50 and 158.62% over *G. margarita*, *G. decipiens*, local and control, respectively.

**P uptake in grain and straw:** Higher P uptake was recorded with the application of *G. mosseae*, in both straw (0.065 g plant<sup>-1</sup>) and grain (0.12 g plant<sup>-1</sup>) (Fig. 1b). However, *G. coronatum* was quite competitive with *G. mosseae* with respect to the uptake of P in straw and grain. *G. mosseae* also gave significantly higher P uptake by straw in comparison with all treatments except *G. coronatum*. Application of *G. coronatum* significantly increased P uptake in straw by 53.65, 96.87, 21.14 and 231.57% more than *G. intraradices*, *G. margarita*, *G. decipiens* and local, respectively. A similar trend was observed in the uptake of P by grain. Application of *G. decipiens* significantly increased straw-P uptake by 62.5, 173.68 and 550% over *G. margarita*, local and control, respectively. Similarly, application of *G. decipiens* numerically increased the grain-P uptake by 11.49, 49.23 and 155.26% over application of *G. margarita*, local and control respectively.

#### **Yield and yield attributes**

**Number of cobs per plant:** The treatments did not differ significantly from each other for the number of cobs per plant (Table 3). However, higher cobs per plant were recorded by the application of *G. mosseae*, *G. intraradices*, *G. coronatum*, *G. margarita* and *G. decipiens*.

**Grain yield:** Grain yield was found to be higher in the plants or seeds inoculated with *G. mosseae* (64.66 g plant<sup>-1</sup>). Grain yield of maize plants followed the order: *G. mosseae* > *G. coronatum* > *G. decipiens* > local > *G. intraradices*. *G. mosseae* gave 27.28, 28.52, 9.35 and 11.7% more grain yield than application of *G. in-*

*traradices*, *G. coronatum*, *G. decipiens* and local, respectively (Table 3). *G. decipiens* produced significantly higher yield by 16.39 and 17.53% than *G. intraradices* and *G. margarita*, respectively. Yield of grains in plants inoculated with *G. coronatum* was found to be at par with the yield obtained with the application of *G. mosseae* as well as with *G. decipiens*.

**Biological yield:** Biological yield was not affected significantly by any of the applied treatments. However the highest biological yield (358.33 g plant<sup>-1</sup>) was recorded with the application of *G. mosseae*. Biological yield obtained with the application of local inoculum was 326.66 g plant<sup>-1</sup>, which is quite competitive with others. Application of *G. coronatum* numerically increased the biological yield by 10.79, 5.96, 4.23, 6.93 and 24.17% over treatments having *G. intraradices*, *G. margarita*, *G. decipiens* and local, respectively. **100-seed weight:** The treatments did not differ significantly from each other for the 100-seed weight (Table 4). However the highest 100-seed weight (26.50 g) was recorded with the application of *G. mosseae*. Application of *G. decipiens* increased the 100-seed weight by 9.00, 6.68, 9.00, 11.42 and 13.95 % over *G. intraradices*, *G. coronatum*, *G. margarita*, local and control respectively (Table 3).

## DISCUSSION

**Colonization and spore:** Higher root colonization was significantly observed in maize after inoculation with *G. species*. Mycorrhizal fungi differ in their ability to infect and colonize roots. *Glomus* species has ability to infect and colonize plant roots faster than *Gigaspora* species, making it highly competitive (Kurlle and Pflieger, 1994). The higher mycorrhizal colonization in maize could be due to strigolactones exuded by host plant roots and taken up by AMF since strigolactones stimulate fungal metabolism and branching (Parniske, 2008). Successful colonization and functional interaction between host plant and the mycobiont is based on the exchange of signaling molecules at different stages of symbiosis. The role of strigolactones as the key signaling compounds in the interaction between plants and soil-borne symbiotic AMF has been suggested recently (Soto *et al.*, 2010).

**Plant growth:** Significant differences ( $p \leq 0.05$ ) were detected for plant height, shoot dry weight and root dry weight among all treatments as well as when the six mycorrhizal inoculants were analyzed separately. It might be due to AM fungi which increased nutrient acquisition from an organic fertilizer source by enhancing root acid phosphatase (ACP) and alkaline phosphatase (ALP) activity thus facilitating P acquisition, increasing photosynthesis, and improving plant growth (Carpio *et al.*, 2009). AMF promotes plant growth by bringing morpho-physiological and biochemical changes in host plants. AMF causes modification in root morphology so as to mediate water and mineral uptake (Alqarawi *et al.*, 2014; Abeer *et al.*,

2015). The *G. mosseae* isolate provided the highest improvement with respect to the plant height, shoot dry weight and root dry weight of maize plant. *G. margarita* provided significantly less growth benefit than *G. coronatum*, *G. mosseae* and *G. decipiens* isolates. Results presented in Table 1 indicate that different isolates of the same genus may be distinct in their interaction with the same plant types. Isolates from the genus *Glomus* provided consistently greater growth benefit than the genus *Gigaspora* as well as the uncharacterized inoculum cultured from the litchi orchard soil used in the experiment. It may be justified by the fact that the inoculation of VAM enhanced the mitotic activity of stem cells resulting in taller plants and more availability of phosphorus for absorption by roots. The different genera of mycorrhiza isolates differ in the production of strigolactones which are a novel class of phytohormones involved in the regulation of shoot branching in plants and are secreted by plant roots for stimulating the presymbiotic growth of AMF. With the identification of strigolactones as the branching factor, not only its production, exudation into the rhizosphere and perception by the AMF but also its specific action in arbuscular mycorrhizal symbiosis (AMS) can now be explored in detail (Bucher *et al.*, 2009).

**Microbial soil properties:** Root structure and functions change due to mycorrhizal infection. Possible VAM induced changes in root exudates and the rhizosphere population, as well as possible physical barriers and chemical inhibitors from AM fungi may have practical implications in the biological control of some plant disease causing organisms. In addition to stimulating *Rhizobium*, VA mycorrhiza also influences rhizospheric bacteria beneficial to the plant. Some bacteria survive for a longer time under mycorrhizal infection than in non-mycorrhizal plants. Similarly, after colonization on plant roots with AM fungi, the quantity of rhizospheric microbes significantly increased (John, 2001). The number of both rhizospheric bacteria and actinomycetes enhanced when plant formed mycorrhizae, while the dominant species composition also changed (Secilia and Bagyaraj, 1987). There may be two pathways for AM fungi to change microbe community structure, the first one is that the AM fungal hypha secretion directly impacts microbe community structures; the another one is that both AM fungi in roots and on the roots alter plant physiological and biochemical processes, then directly or indirectly change the plant root secretion (Badri and Vivanco, 2009), thus alter those structures (Zhu *et al.*, 2005).

In this experiment, we further observed that activities of soil dehydrogenase, acid phosphatase and alkaline phosphatase in maize rhizospheric soils gradually increased when compared with control. It might be due to inoculation with AM fungi enrich soil microbe quantities, equilibrate proportion of various microbes, maintain a stabilization of proper proportion of the microbes, enhance soil carbon, nitrogen, and phosphorous cycling power, thus improve the soil enzyme ac-

tivity. Our results of increased activities of alkaline and acidic phosphatases in AMF inoculated plants. The similar type of findings was reported in *Fraxinus rotundifolia* (Kebradadi *et al.*, 2014), *Ipomoea carnea* (Amaya *et al.*, 2009), and *Chrysanthemum indicum* (Prasad *et al.*, 2012). Sunflower under cadmium (Cd) stress, Cd reduced the plant photosynthesis rate, growth, chlorophyll contents and cell membrane stability whereas, in AMF inoculated plants showed higher activities of acid and alkaline phosphatases which improved the plant growth, photosynthesis rate and cell membrane stability and reduced the Cd stress (Allah *et al.*, 2015). Gianinazzi *et al.* (1979) suggested that ALP production in mature intra-radical arbuscules led to polyphosphate breakdown. ACP can enhance release of inorganic phosphorus from organophosphates.

**P content in straw and grain:** Phosphorus content in straw and grain was found to be highest under *G. mosseae* inoculation (Fig.1a). It might be due to the solubilization of unavailable phosphorus and increase in phosphorus uptake through plants root since mycorrhiza is responsible for increase in the surface area of roots. About 95-99% of the total P in soil exists in insoluble form unavailable to plants. The remaining soluble P is mostly present on exchangeable sites in equilibrium with the small amount of P in the soil solution. Therefore any solubilization of insoluble P by AM fungi significantly adds to the available pool. These results corroborate with the results of Gui *et al.* (2011) who reported that inoculation of *G. mosseae* significantly increased plant P concentration over uninoculated maize plants.

**P uptake:** P uptake was found to be higher with the application of *G. mosseae*, than *G. coronatum* and *G. decipiens* (Fig.1b). AMF induced enhancement in phosphatase activity could possibly mediate the release of organically bound phosphorous and hence increasing transport and uptake of phosphorous in AMF inoculated plants. Increased activity of phosphatases has direct bearing with the phosphorus metabolism. Bhadrarajah *et al.* (1999) observed a high degree of correlation between phosphorous uptake and activity of phosphatases. Earlier studies by Bethlenfalvay (1993) also show that different species and strains of AMF differ in their effectiveness in increasing nutrient uptake and plant growth. The pattern of extra- and intra-radical forms of AMF hyphae can also justify the difference in phosphorus acquisition among the AMF isolates. Hence, the level of development of extra-radical mycelium in the soil is a major determinant of the efficiency of AMF for phosphorus uptake (Rakshit and Bhadoria, 2009). Similar results indicating that phosphorus uptake by mycorrhizal plants fluctuate with fungal isolates and genetic variability; have also been found on soybean cultivars (Diop *et al.*, 2003). The nitrogen and phosphorus transfer to the maize plants may also be a consequence of the competitive demand for the nutrients, with both host plant and fungus evolving transporters to take advantage of the localized increase in nutrients.

**Yield:** The yield and yield attributes increased significantly over control with the inoculation of AM species. Studies conducted by Sabia *et al.* (2015) also revealed a significant effect of AM inoculation on dry matter yield and quality of forage maize cultivated within a low input system. This might be due to enhanced nutrient uptake by the roots. Since immobile ions in soil like phosphate lead to formation of a zone of phosphate depletion around roots in phosphate deficient soils mycorrhizal growth helps the roots to absorb phosphate ions much faster which are replenished at the root surface by diffusion. The AM hyphae attached to the roots extend beyond this depletion zone and promote nutrient translocation from the soil to the plants through the root cortex.

## Conclusion

The inoculation with AM fungus enhanced the population of soil bacteria, fungi and actinomycetes and also improved soil dehydrogenase, acid phosphatase and alkaline phosphatase activities as compared to the control. It also contributed to relative better plant growth and higher uptake of phosphorus. As evident from the results, the AM fungal inoculation can effectively modify the soil microbe population and community structure by increasing the soil enzymatic activities and plant phosphorus uptake.

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## REFERENCES

- Abeer, H., Allah, E.F.A., Alqarawi, A.A., Al-Whibi, M., S. Alenazi, M.M., Egamberdieva, D. and Ahmad, P. Ahmad. (2015). Arbuscular mycorrhizal fungi mitigates NaCl adverse effects on *Solanum lycopersicum* L. *Pak. J. Bot.* 47(1):327-340.
- Allah, E.F.A., Abeer, H., Alqarawi, A.A. and Hend, A.A. (2015). Alleviation of adverse impact of cadmium stress in sunflower (*Helianthus annuus* L.) by arbuscular mycorrhizal fungi. *Pak. J. Bot.* 47(2): 785-795.
- Alqarawi, A.A., Allah, E.F.A. and Abeer, H. (2014). Alleviation of salt-induced adverse impact via mycorrhizal fungi in *Ephedra aphylla* Forssk. *J. Plant Interact.* 9 (1):802-810.
- Amaya C., L., Davies, F.T., Fox, T. and He, C. (2009). Arbuscular mycorrhizal fungi and organic fertilizer influence photosynthesis, root phosphatase activity, nutrition, and growth of *Ipomoea carnea* ssp. *Fistulosa*. *Photosynthetica* 47 (1): 1-10.
- Andrade, G., Linderman, R.G. and Bethlenfalvay, G.J. (1998). Bacterial associations with the mycorrhizosphere and hyphosphere of the arbuscular mycorrhizal fungus *Glomus mosseae*. *Plant Soil.* 202: 79-87.
- Badri, D.V. and Vivanco, J.M. (2009). Regulation and function of root exudates. *Plant Cell Environ.* 32(6):666-681.

- Bethlenfalvay, G.J. (1993). Mycorrhizae in the agricultural plant-soil system. *Symbiosis* 14(1-3): 413-414.
- Bhadraiah, B., Kankadurga, V.V. Kankadurga, Ramrao, P. Ramrao and Manoharachary C. (1999). Effect of VAM fungi and rock phosphate on phosphatase activities in *Terminalia arjuna*. National Conference on Mycorrhiza Section. *Physiol Biochem.* 5-7.
- Bucher, M., Wegmuller, S. and Drissner, D. (2009). Chasing the structures of small molecules in arbuscular mycorrhizal signaling. *Curr. Opin. Plant Biol.* 12: 500-507.
- Busto, M. D. and Perez-Mateos, M. (1997). Stabilisation of cellulases by cross-linking with glutaraldehyde and soil humates Extraction of humic-fl-glucosidase fractions from soil. *Biores Technol.* 60: 27-33.
- Caravaca, F., Alguacil, M.M., Azcon, R., Díaz, G., Roldan, A. (2004). Comparing the effectiveness of mycorrhizal inoculation and amendment with sugar beet, rock phosphate and *Aspergillus niger* to enhance field performance of the leguminous shrub *Dorycnium pentaphyllum* L. *Appl. Soil Ecol.* 25: 169-180.
- Carpio, A. L., Davies, F.T., Fox, T. and He, C. (2009). Arbuscular mycorrhizal fungi and organic fertilizer influence photosynthesis, root phosphatase activity, nutrition, and growth of *Ipomoea carnea* ssp. *Fistulosa*. *Photosynthetica* 47 (1): 1-10.
- Casida, L.E., Klein, D.A. and Santoro, T. (1964). Soil dehydrogenase activity. *Soil Sci.*, 98:371-376.
- Diop, T. A., Krasova-Wade, T., Diallo, A., Diouf, M. and Gueye, M. (2003). Solanum cultivar responses to arbuscular mycorrhizal fungi: growth and mineral status. *Afr. J. Biotech.*, 2(11): 429-433.
- Feng, K., Lu, H. M., Sheng, H. J., Wang, X. L. and Mao, J. (2004). Effect of organic ligands on biological availability of inorganic phosphorus in soils. *Pedosphere* 14: 85-92.
- Gerdemann, J.W. and Nicolson, T.H. (1963). Spores of mycorrhizal *Endogone* species extracted from soil by wet-sieving and decanting. *Trans. Br. Mycol. Soc.* 46: 235-244.
- Gianinazzi, S., Gianinazzi, V. and Pearson, J. (1979). Dexeheimer, Enzymatic studies on the metabolism of vesicular-arbuscular mycorrhiza. III. Ultrastructural localization of acid and alkaline phosphatase in onion roots infected by *Glomus mosseae* (Nicol. & Gerd.), *New Phytol.* 82: 127-132.
- Gomez, K.A. and Gomez, A.A. (1984) *Statistical Procedures in Agricultural Research*. Wiley Publishing House, New York
- Gui, Y. Z., Li, P. Z., Ming, F. W., Zhen, L., Qiao, L. F., Qi, R. S. and Guo, H. Xu. (2011). Effect of arbuscular mycorrhizal fungi, organic fertilizer and soil sterilization on maize growth. *Acta Ecologica Sinica* 31:192-196.
- Hata, S., Kobae, Y. and Banba, M. (2010). Interactions between plants and arbuscular mycorrhizal fungi. *Int. Rev. Cell Mol. Biol.* 281: 1-48.
- Hoagland, D.R. and Arnon, D.I. (1938). The water culture method of growing plants without soil. *California Agricultural Experiment Station, Circ.* 347.
- Jackson, M. L. (1973). *Soil Chemical Analysis*. Prentice Hall, New Delhi, 32, 34-36.
- John, M. (2001). Microbial interactions and biocontrol in the rhizosphere. *J. Experimental. Bot.* 552 (1):487-511.
- Kebrabadi, B.Z., Matinizadeh, M., Daryayi, M.G., and Salehi, A. (2014). Changes in acid and alkaline phosphatase enzyme activity in rhizosphere ash *Fraxinus rotundifolia* and its correlation with soil and plant phosphorus. *J. Biodiversity Environ Sci.* 4(5): 233-238.
- Kurle, J. E. and Pflieger, F.L. (1994). The effect of cultural practices and pesticides on VAM fungi. In: F.L. Pflieger and R.G. Linderman (Eds.) *Mycorrhizae and Plant Health*. APS Press, Minnesota, pp. 101-131.
- Mar Vazquez, M., Cesar S., Azcon R., Barea, J.M. (2000). Interactions between arbuscular mycorrhizal fungi and other microbial inoculants (*Azospirillum*, *Pseudomonas*, *Trichoderma*) and their effects on microbial population and enzyme activities in the rhizosphere of maize plants. *Appl. Soil Ecol.* 15:261-272.
- Parniske, M. (2008). Arbuscular mycorrhiza: the mother of plant root endosymbioses, *Nature Rev. Microb.* 6: 763-775.
- Philips, J.M. and Hayman, D.S. (1970). Improved procedures for clearing roots and staining parasitic and vesicular-arbuscular mycorrhizal fungi for rapid assessment of infection. *Trans Br Mycol Soc.* 55: 158-161.
- Prasad, K., Yadav A., Aggarwal K. and Tanwar, A. (2012). Impact of different levels of superphosphate using arbuscular mycorrhizal fungi and *Pseudomonas fluorescens* on *Chrysanthemum indicum* L. *J. Soil Sci. Plant Nut.* 12(3): 451-462.
- Rakshit, A. and Bhadoria, P. (2009). Influence of arbuscular mycorrhizal hyphal length on simulation of P influx with the mechanistic model, *Afr. J. Microb. Res.* 3 (1): 001-004.
- Rao, A.V., Tak, R. (2001). Influence of mycorrhizal fungi on the growth of different tree species and their nutrient uptake in gypsum mine spoil in India. *Appl. Soil Ecol.* 17: 279-284.
- Sabia, E., Claps, S., Morone, G., Bruno, A., Sepe, L. and Aleandri, R. (2015). Field inoculation of arbuscular mycorrhiza on maize (*Zea mays* L.) under low inputs: preliminary study on quantitative and qualitative aspects. *Italian J. Agron.* 10: 30-33.
- Schubler, A., Schwartzott, D. and Walker, C. (2001). A new fungal phylum, the Glomeromycota: phylogeny and evolution. *Mycological Res.* 105: 1413-1421.
- Secilia, J. and Bagyaraj, D.J. (1987). Bacteria and actinomycetes associated with pot cultures of vesicular arbuscular mycorrhizas. *Canadian J. Microbial.* 33(8):1069-1073.
- Smith, S.E. and Read, D.J. (2008). *Mycorrhizal symbiosis*. Academic Press, London, UK.
- Soto, M.J., Fernández-Aparicio, M., Castellanos-Morales, V., García-Garrido, J.M., Ocampo, J.A., Delgado, M.J. and Vierheilig, H. (2010). First indications for the involvement of trigolactones on nodule formation in alfalfa (*Medicago sativa*). *Soil Biol. Biochem.* 42: 383-385.
- Subba, R.N.S. (1986). *Rhizobium* and root nodulation. In: soil microorganisms and plant growth. Oxford IBH New Delhi.
- Tabatabai, M.A. and Bremner, J.M. (1969). Use of p-nitrophenyl phosphate for assay of phosphatase activity. *Soil Bio Biochem.* 1:301-307.
- Thom, C. and Raper, K.B. (1945). *A manual of the Aspergilli*. Williams and Wilkins Co., Baltimore, U. S. A.
- Zhu, H.H., Long, L.K. and Yang, S.Z. (2005). Influence of AM fungus on *Ralstonia Solanacearum* Population and bacterial community structure in rhizosphere. *Mycosystema* 24(11):137-142.
- Zhu, Y.G., Smith, F.A. and Smith, S.E. (2003). Phosphorus efficiencies and responses of barley (*Hordeum vulgare* L.) to arbuscular mycorrhizal fungi grown in highly calcareous soil. *Mycorrhiza.* 13:93-100.