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Effect of arbuscular mycorrhizal (AM) fungi inoculation on enzymatic activity and zinc uptake under direct seeded rice system

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Abstract: The application of treatment T_3 (*Glomus mosseae* + 100 % RDF NK) produced significantly more root volume by 72.60 %, 17.80 %, 12.25 %, 14.13 % over the application of treatment T_1 (Control), treatment T_5 (*Glomus coronatum*+ 100 % RDF NK), T_6 (*Gigasporadecipein* + 100 % RDF NK) and T_7 (BAU AM-1(*Glomus sp* + 100 % RDF NK), respectively. Similar trend shows at harvesting stage, here the maximum root volume (23c.c) was recorded by the application of T_3 (*Glomus mosseae* + 100 % RDF NK). Maximum AM colonization and spore count was observed at panicle initiation stage with the application of treatment T_3 (*Glomus mosseae* + 100 % RDF NK). This treatment also gave maximum dehydrogenase activity (55.86 µg TPF g^{-1} 24 hr^{-1}), acid phosphatase activity (0.299 mg PNP g^{-1} hr^{-1}) and alkaline phosphatase activity (0.54 mg PNP g^{-1} hr^{-1}) at panicle initiation stage. Application of treatment T_3 (*Glomus mosseae* + 100 % RDF NK) significantly increased DTPA extractable Zn in soil and Zn content in plant when compared with all the treatments except treatment T_6 (*Gigasporadecipien*+ 100 % RDF NK). The maximum zinc uptake (0.056 mg pot^{-1}) by grain was recorded under treatment T_3 (*Glomus mosseae* + 100 % RDF NK) followed by application of treatment T_6 (*Gigasporadecipien* + 100 % N and K). Highest grain yield (14.08 g pot-1) was found with the treatment T_3 (*Glomus mosseae* + 100 % RDF NK). 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 significantly increased the zinc uptake by grain in direct seeded rice (DSR).

Keywords: DSR, Mycorrhiza, Rice yield, Soil enzyme, Spore count, Zinc uptake

INTRODUCTION

In dry direct seeded rice (DSR), because of the aerobic condition and alternate wetting/drying cycles, the plant availability of several major nutrients including N and micronutrients like Zn and Fe is likely to be less. Due to poor native bioavailable Zn and low exogenous supply, depletion zones are formed around roots. In plants, Zn plays an significant role as integral co-factor of over 300 enzymes which are involved in biosyntheses and turnovers of proteins, nucleic acids, carbohydrates, and lipids. Zn deficiency causes a lot of health problems in humans, such as impairments of physical development, immune system, and brain function (Cakmak, 2008). In addition, Zn is critical for the synthesis of phytohormones such as auxin, absicic acid, gibberellins and cytokinins. Thus, its deficiency in plant tissues adversely affects various vital processes occurring within plant body. Although Zn is required by the plant in microconcentration, its bioavailable fraction in soil is very low. Its deficiency is a widely occurring constraint for rice production and for human nutrition. To improve zinc uptake by plants, it should be brought in close proximity of roots. This can be achieved either by external application of Zn or by improving the growth and surface area of roots thus enabling them to absorb nutrients beyond the depletion zone. Rhizosphere microflora especially mycorrhizal fungus is widely known for its impact on root architecture. Mycorrhizal plants take up Zn over longer distances crossing the depletion zone. Lehman *et al*. (2014) confirmed by a meta-analysis that AMF positively affects Zn concentrations in various crop plant tissues including rice. Hajiboland *et al*. (2009) studied the influence of AMF on the uptake of Zn and P by two contrasting rice genotypes in a pot experiment. After inoculation with mycorrhiza they observed up to two-fold higher Zn uptake by rice genotypes under Zn deficiency. An enhanced AMF inoculation under aerobic condition has been shown to increase rice Zn uptake as observed by Gao *et al*. (2007). They observed that AMF inoculated plants produced more biomass and took up more Zn than the non-mycorrhizal control. 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 Vazquez *et al.,* 2000), dehydrogenase, urease, protease and β-glucosidase (Caravaca *et al.,* 2004). Mar Vazquez *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. Considering with these facts the present investigation has been formulated to evaluate the role of vesicular arbuscular mycorrhizal fungi on the bioavailability of Zn in Direct Seeded rice grain. The present investigation was undertaken with an objective to determine the effect of AM inoculation on root volume, soil enzymatic activity and zinc uptake by grain under low soil phosphorous and zinc conditions typical of the direct seeded rice eco-system.

MATERIALS AND METHODS

The present study was undertaken to screen the AM fungi for direct seeded rice crop during the *kharif* season of 2015-16 with a promising variety ShushkSamrat, at Bihar Agricultural University, Sabour, Bhagalpur, India. Inoculums of the five AM species viz., *Glomus mosseae*, *Glomus coronatum,Glomus intraradices Gigasporadecipien* and 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 Local *(*BAU AM-1 (*Glomus sp.*) was taken from Department of Soil Science and Agricultural Chemistry, BAU, Sabour. The field soil (0-15 cm) used for the pot experiment was loamy sand in texture, having a pH of 7.8 and EC of 0.20 dS m^{-1}. The organic carbon content of the substrate was 0.47 %, and the available nitrogen, phosphorus and potassium content was found 125.44, 12.55 and 240.92 kg ha^{-1}, respectively. Among micronutrient zinc content was 0.48 ppm.

Seeds were surface-sterilized by treatment with a 1:1 mixture of H_2O_2 and absolute ethanol for 2 minutes followed by a treatment with 0.05 % $HgCl_2$ 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 15 kg of sterilized substrate. The following treatment structure was formulated for the study: T_1- control, T_2- RDF (100 %), T_{23} *G. mosseae* + 100 % RDFNK, T_4- *G. coronatum* + 100 % RDFNK, T_5- *G. intraradices* + 100 % RDFNK, T_6- *G. decipiens* + 100 % RDFNK and T_7- Local (BAU AM-1 (*Glomus sp.*) + 100 % RDFNK. About 5 g of the AM inoculum source (containing and 10 spores g^{-1}) was mixed with the upper 4 cm of the substrate in each pot. In each pot, 5 sterile seeds of rice were planted. The one third dose of nitrogen and full dose of phosphorous and potassium were applied at the time of sowing and remaining two third dose of nitrogen were applied at the tilering and panicle initiation stage. Irrigation was applied as per crop requirement. Five plants were up rooted from each pot by destructive sampling at harvesting stage and measure the root volume by volume displacement method provides (Harrington *et. al.,* 1994).

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, followed by acidification for a few minutes using HCl (2 %). Staining was done using trypan blue (0.05 % w/v) in lactoglycerol, which is a mixture of lactic acid, glycerol and distilled water 1:1:1. The root colonization observed by the method given by Philips and Hayman (1970). The percent root colonization was calculated by using following formula:

% of mycorrhizal colonization (%) =

$$\frac{\text{Number of root segment examine} \times 100}{\text{Total Number of root segment taken}}$$

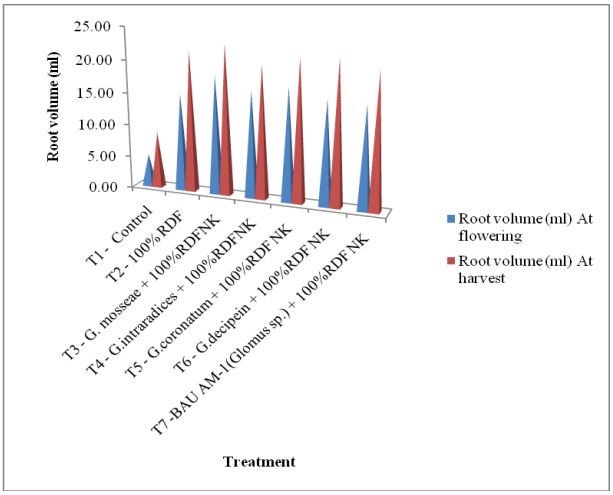

Fig. 1. *Effect of AM fungi species on root volume (cc) of DSR.*

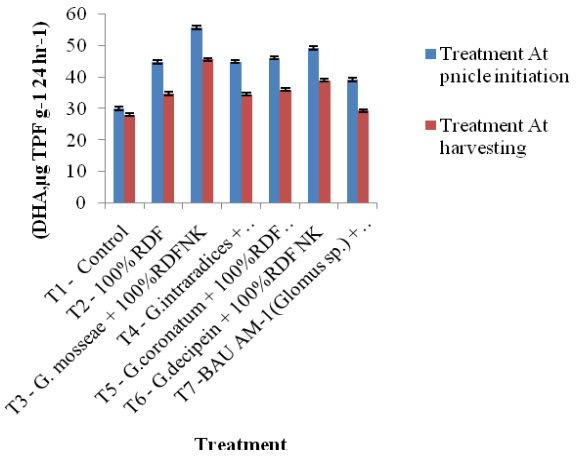

Fig. 2. *Effect of various AM fungi on dehydrogenase activity (µg TPF g^{-1} 24 hr^{-1}).*

The soil sample collected from the each pot along with plant roots. Soil sample were soaked for 30 minutes using about 400 mL of 2 % (w/v) "Calgon" (Na-hexametaphosphate) solution before being homogeneously mixed manually. Samples were mixed for 10 minutes, then the soil solution was allowed to settle for about 20 seconds before the supernatant was decanted carefully onto a series of sieves. The remaining soil was then mixed again with tap water, stirred for 5 minutes, settled for about 20 seconds, then decanted onto the sieve set. This process was repeated until the solution looked clear. There were different sieve sets used. However, the top one was a 750 µm mesh sieve, and the sieving fraction captured on this sieve was discarded (no spores were found). To capture spores for the counting, three sieves were attached below the top sieve. These sieves had opening sizes of 250, 105 and 53 µm from top to bottom (Wet Sieving and Decanting method, Gerdemann and Nicolson 1963). The spores captured on the gridded filter paper were counted under a dissecting microscope with 40x magnification.

Dehydrogenase activity was determined by using procedure as outlined by Casida *et al.,* 1964. Acid and alkaline phosphatase activity was determined by following procedure as outlined by Tabatabai and Bremer, 1969.

The five plant samples were dried in oven at 65±1 ^{o}C for 48 hours and dry biomass yield of the crop was recorded. The oven dried plant samples were ground thoroughly by a Wiley mill. A representative ground plant sample (1.0 g) was taken for digestion. For pre-digestion in conical flasks (100 mL capacity) the plant samples was soaked overnight with 5 mL of concentrated HNO_3 and finally digested in a di-acid mixture (8 mL) containing HNO_3 and $HClO_4$ acid (9:4) on an electric hot plate following the procedure described by Piper (1967). The digested material was cooled, diluted with distilled water and filtered through Whatman No. 1 filter paper. The volume was made up to 100 mL and stored in a polypropylene container (125 mL capacity) for further analysis. Total Zn content in the extract was determined by Atomic absorption Spectrophotomer (AAS). The same procedure has been used for the zinc content analysis in grain. Uptake of zinc by plant was calculated by following formula:

Zn uptake by grain (kg ha^{-1}) =

$$\frac{\text{Zn conc. in grain (\%)} \times \text{Grain yield (pot}^{-1}\text{)}}{100}$$

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 AND DISCUSSION

Root volume: The experimental results showed that the maximum root volume of rice was recorded at harvesting stage when compared with flowering stage (Fig. 1). The application of treatment T_3 (*G. mosseae* + 100 % RDF NK) produced significantly more root volume by 72.60 %, 17.80 %, 12.25 %, 14.13 % over the application of treatment T_1 (Control), treatment T_5 (*G.coronatum* + 100 % RDF NK), T_6 (*G. decipein* + 100 % RDF NK) and T_7 (BAU AM-1 (*G. sp* + 100 % RDF NK) respectively. Similar trend shows at harvesting stage, here the maximum root volume (23 cc) was recorded by the application of T_3 (*G. mosseae* + 100 % RDF NK). It might be due to the AM association besides facilitating nutrient uptake this also important in terms of root architecture as they are known to alter root topology (Schellenbaum *et al*., 1991). Positive effect on improvement of root volume by *G. intraradices* inoculation has been demonstrated in sesame (Boureima *et al*., 2008) and alteration in root length,

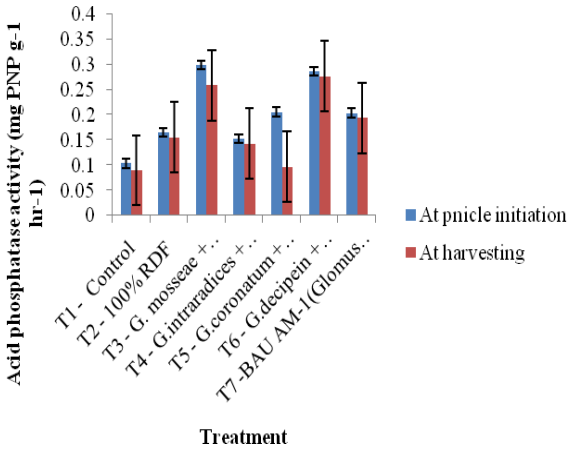

Fig. 3. *Effect of various AM fungi on acid phosphatase activity (mg PNP g^{-1} hr^{-1} .*

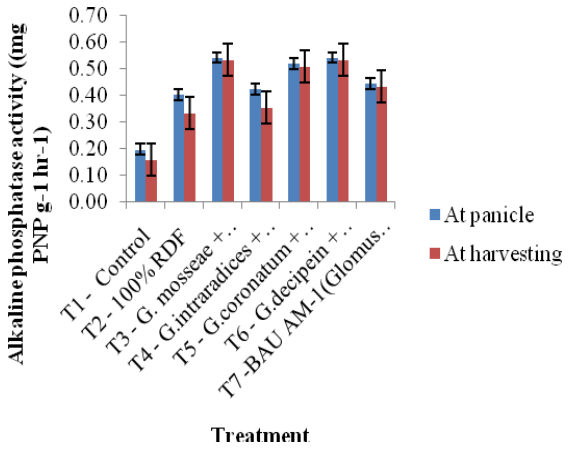

Fig. 4. *Effect of various AM fungi on alkaline phosphatase activity (mg PNP g^{-1} hr^{-1}).*

Table 1. Effect of AM fungi and N and K on AM colonization (%) at flowering and harvesting stage of DSR.

Treatment	AM colonization (%)		Spore count (100 g^{-1} soil)	
	Flowering stage	Harvesting stage	Flowering stage	Harvesting stage
T_1 - Control	24.67	15.00	42.33	35.00
T_2 - 100% RDF	46.67	36.67	216.00	202.00
T_3 - *Glomus mosseae*+ 100%RDFNK	66.67	56.67	314.33	299.33
T_4 – *Glomus intraradices*+ 100%RDFNK	53.33	43.33	241.00	228.00
T_5 – *Glomus coronatum*+ 100%RDF NK	56.67	46.67	301.67	286.33
T_6 – *Gigasporadecipein*+ 100%RDF NK	63.33	53.33	271.67	257.67
T_7 – BAU AM-1(*Glomus sp.*)+ 100%RDF NK	50.00	38.33	223.33	211.33
CD (0.05)	14.60	12.50	5.46	6.56
SE(m)	4.77	4.08	1.78	2.14
C.V.	15.99	17.07	1.34	1.71

surface area and volume as a result of mycorrhization has also been reported in *Citrus* and maize (Sheng *et al*., 2009; Wu *et al*., 2012).

AM colonization: The maximum AM root colonization was observed at flowering stage in comparison to the harvesting stage (Table 1). It might be due to the more infection of AM fungi with root of host plant and more release of plant photosynthate which is the carbon source to the AM fungi. The results are corroborating with the results of Singh and Kumar, 2007. At the harvesting stage application of treatment T_3 (*G. mosseae* + 100 % RDF NK) produced significantly more AM colonization except treatment T_4 (*G. intraradices* + 100 % RDF NK), T_5 (*G. coronatum* + 100 % RDF NK), and T_6 (*G. decipien* + 100 % RDF NK) by 62.99 %, 29.99 % and 25.03 % than application of T_1 (control) condition and T_2 (100 % RDF) and T_7 (BAU AM -1(*G. sp.*) + 100 % RDF NK), respectively. Similar trends was found at harvesting stage, hence it shows that the maximum AM colonization occur at flowering stage, which is inoculated with T_3 (*G. mosseae* + 100 % RDF NK). It might be due to the 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 (Kurle and Pfleger, 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). The role of strigolactones as the key signaling compounds in the interaction between plants and soilborne symbiotic AMF has been suggested recently (Soto *et al.,* 2010).

Spore count: The data regarding spore count showed that the maximum spore counts were recorded at flowering stage in comparison to harvesting stage in all the given treatments (Table 1). The application of treatment T_3 (*G. mosseae* + 100 % RDF NK) produced significantly higher spore count when compare with other applied treatments. It might be due to the more secretions of signal for root infection which are secreted by the host root, and more availability of plant photosynthate from plant to the associated fungi. The similar results were recorded by the Singh, *et al*., 2015 who found that the inoculation of *G. mosseae* produced maximum spore count when compared with other treatments at 75 DAS of maize in a pot experiment. The application of treatment T_3 (*G. mosseae* + 100 % RDF NK) produced significantly more spore count when compare with other applied treatments. This treatment also gave significantly more spore count than other inoculated application at harvesting stage. The maximum spore count (314.33 per 100 g, soil) found at flowering stage under the treatment T_3 (*G. mosseae* + 100 % RDF NK). It might be due to the 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 (Singh *et al.,* 2015).

Soil enzymatic activity: The data on dehydrogenase activity is affected by various treatments at the panicle initiation and harvesting stage of rice which are presented in the results (Fig. 2). Microbial respiration is supposed to be highest in panicle initiation stage over harvesting stage of rice plant. This finding might be explained in the light of the report made by (Zeng Lusheng *et al.,* 2005). In the present result at flowering stage, inoculation with T_3 (*G. mosseae* + 100 % RDF NK) having given significantly higher dehydrogenase activity by 46.16 %, 19.72 %, 19.39 %, 17.33 %, 11.70 %, 29.58 % over treatments T_1 (control) condition, T_2 (100 % RDF), T_4 (*G. intraradices* + 100 % RDF NK), T_5 (*G. coronatum* + 100 % RDF NK), T_6 (*G.decipein* + 100 % RDF NK) and T_7 (BAU AM -1 (*Glomus sp.*) + 100 % RDF NK) respectively. It might be due to the mycorrhizal fungi can increases soil enzymatic activities, such as phosphatase (Mar Vazquez *et al.,* 2000), dehydrogenase, urease, protease and β-glucosidase (Caravaca *et al.*, 2003 and 2004).

The similar trend was observed for the acid phosphatase enzyme during the study. The inoculation with T_3 (*G. mosseae* + 100 % RDF NK) having more significantly acid phosphatase activity with respect to other inoculated species of VAM fungi (Fig. 3). Similar trends follow at harvesting stage, but in this stage inoculation with T_3 (*G. mosseae* + 100 % RDF NK) having significantly more acid phosphatase activity except

Table 2. Effect of *Glomus mosseae, Glomus coronatum, Glomus intraradices, Gigasporadecipien* on available P (kg ha^{-1}), DTPA, Zn (mg kg^{-1} soil) Zn content and uptake by grain of rice.

Treatment	Available P (mg kg^{-1} soil)	DTPA, extractable Zn (mg kg^{-1} soil)	Zn content in grain (%)	Zn uptake (g pot^{-1}) by grain	Grain yield
T_1 - Control	5.24	0.49	0.0024	0.019	7.77
T_2 - 100% RDF	6.27	0.50	0.0028	0.040	14.09
T_3 - *Glomus mosseae* + 100 % RDFNK	7.40	0.52	0.0040	0.056	14.08
T_4 – *Glomus intraradices* + 100 % RDFNK	6.77	0.48	0.0033	0.040	12.00
T_5 – *Glomus coronatum* + 100 % RDF NK	6.91	0.51	0.0033	0.042	12.87
T_6 – *Gigasporadecipein* + 100 % RDF NK	7.07	0.50	0.0034	0.044	12.92
T_7 – BAU AM-1(*Glomus sp.*) + 100 % RDF NK	5.59	0.51	0.0033	0.036	10.99
CD (0.05)	0.22	0.01	0.001	0.009	0.97
SE(m)	0.10	0.07	0.0002	0.004	0.45
C.V.	0.16	16.52	1.65	1.870	4.58

T_6 (*G. decipein* +100 % RDF NK), while the numerical value with the inoculation of *G. mosseae* is 0.28 mg $PNPgm^{-1}hr^{-1}$ and *G.decipein* having 0.26 mg $PNPgm^{-1}hr^{-1}$. This finding has been well supported by the experimentation, undertaken by Mar Vazquez *et al*. (2000) who reported that the mycorrhizal colonization induced qualitative changes in the microbial population and enzyme activities in the rhizosphere of maize plants.

The application with the treatment T_3 (*G. mosseae* + 100 % RDF NK) and treatment T_6 (*G.decipein* + 100 % RDF NK) given maximum Alkaline phosphatase activity at both the growth stages (Fig.4). The results for alkaline phosphatase activity by the application of *G. mosseae*are the confirmation of Singh *et al.,* 2015. The major contribution of soil enzyme pool is mainly mediated by soil microorganisms. In addition to that other soil microflora, plant residues undergoing varying degree of decay also contributed to this pool (Krishna *et al.,* 2005). Alkaline phosphatase activity of soil increased up to panicle initiation stage afterwards it declined. Rice crop produced higher amount of root exudation in initial stages of growth which enhanced microbial activity in crop and modify nutrient concentration in soil (Dotaniya *et al.,* 2014 and Singh *et al*., 2015).

Available phosphorous (mg kg^{-1}) in soil after harvest: The presented data (Table 2) showed that addition of AM inoculation increased soil P concentration which was significantly higher over control. The application with the treatment T_3 (*G. mosseae* + 100 % RDF NK) given significantly more availability of phosphorus by 10.42 %, 4.37 %, 2.43 %, 1.06 % and 8.52 % than treatment T_1 (control) condition, T_2 (100 % RDF), T_4 (*G. intraradices* + 100 % RDF NK), T_5 (*G. coronatum* + 100 % RDF NK) and T_7 (BAU AM -1 (*Glomus sp.*) + 100 % RDF NK), respectively. It may be due to the *Glomus mosseae* had pronounced effect for phosphorus acquisition in soil. A parallel trend was also seen in case of available N, K and organic carbon status of alluvial soil (Olsson *et al*., 2010; Beura *et al*., 2016).

DTPA extractable zinc (mg kg^{-1}) in soil after harvest: The perusal data (Table 2) stated that application with treatment T_3 (*Glomus mosseae* +100 % RDF NK) increased significant DTPA extractable zinc by 5.76 %, 3.84 %, 7.69 % and 3.84 % than T_1 (control) condition, T_2 (100 % RDF), inoculation with T_4 (*G. intraradices* + 100 % RDF NK) and T_6 (*G. decipein* 100 % RDF NK), respectively. It might be due to the application of arbuscular mycorrhizal fungi increase the solubilization of unavailable zinc through their mechanism of acidulation. Higher root biomass mediated exudation and this was perhaps the most important reason for the increase. This result was in consonance with the findings of Habasy *et al*. (2005); Archana *et al*. (2012); Balakrishnan and Subramanian (2012); and Beura *et al*. (2016).

Effect of AMF species on zinc uptake by grain: The data from the result revealed that the application of different arbuscular mycorrhizal fungi increased the zinc content and uptake by the plant and grain (Table 2). The application of treatment T_3 (*G. mosseae* + 100 % RDF NK) given significantly more uptake of Zinc content in grain by 66.07 %, 28.57 %, 28.57 %, 25 %, 21.42 % and 35.71 % than T_1 (control) condition, T_2 (100 % RDF) and inoculation with T_4 (*G. intraradices* + 100 % RDF NK), T_5 (*G. coronatum* + 100 % RDF NK), T_6 (*G. decipein* + 100 % RDF NK) and T_7 (BAU AM -1 (*G. sp.*) + 100 % RDF NK), respectively. It might be due to the more availability of zinc in the rhizosphere and absorption of more zinc from the beyond of depletion zone. AM fungi aid in going beyond this depletion zone to reach a new pool of soluble phosphate and some micronutrients (Marschner, 1995; Smith, 1997) via its extra-radical mycelial network extending the absorbing area (Johanson *et al.,* 1993, Li *et al.,* 1991) for increased P uptake and zinc According to Burkert and Robson (1994), arbuscular mycorrhizae can acquire Zn from a distance of 40 mm from the root surface. Jansa*et al.* (2003) noted that *G. intraradices* can take up Zn from a distance of 50 mm from the roots of maize.

Effect of AMF species on yield: The yield of direct

seeded rice increased significantly over control with the inoculation of AM species (Table 2). 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 inoculation with treatment T_3 (*G. mosseae* + 100 % RDF NK) significantly increased grain yield by 44.81 %, 14.77 %, 8.59 %, 8.23 % and 21.94 % than T_1 (control) condition and inoculation with T_4 (*G. intraradices* + 100 % RDF NK), T_5 (*G. coronatum* + 100 % RDF NK), T_6 (*G. decipien*+ 100 % RDF NK) and T_7 (BAU AM -1 (*Glomus sp.*) + 100 % RDF NK), respectively. It might be due to 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. 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. Singh *et al.,* 2015 also conducted a pot experiment for screening the AM fungi species for maize crop and found that with the application of *G. mosseae the* grain yield was found maximum (64.66 g plant^{-1}).

Conclusion

The inoculation with AM fungus enhanced the soil dehydrogenase, acid phosphatase and alkaline phosphatase activities as compared to the control and application of 100 % RDF. It also contributed to relative better plant growth and higher uptake of zinc by grain and shoot. 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 enhanced the zinc uptake by grain (0.036-0.056 g pot^{-1}) of DSR.

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