A Holistic Approach to Utilize Mycorrhizae for Improving Soil Health and Enhancing Yield of Wheat in Bangladesh

M. T. Iqbal

Department of Agronomy and Agricultural Extension, University of Rajshahi, Rajshahi 6205, Bangladesh

Abstract

Arbuscular mycorrhizal fungi (AMF) species may have positive impacts on wheat productivity through translocation of nutrients within wheat plant tissue. To quantify effect of mycorrhizae species on wheat plant productivity, two experiments were conducted in this study. One preliminary seedlings growth experiment was conducted in sand culture to reckon exact mycorrhizae effect on wheat seedlings growth response and changes in sand properties due to mycorrhizae inoculation. Another pot experiment was conducted to quantify wheat productivity, nutrient content within wheat plant tissue and changes in soil physiochemical characteristics due to four mycorrhizae species amendment. Result showed that wheat seedling responded better due to mycorrhizae inoculation. Mycorrhiza inoculation also improves nutrient content within wheat seedlings tissue and store carbon in sand. Finding also showed that mycorrhizae species inoculation increased growth and yield of wheat as well as phosphorus accumulation within wheat grain. Both experiments showed that mycorrhizae species inoculums store carbon in soil and within wheat plant tissue.

Keywords: Chlorophyll; AMF spores; mycorrhizae species; mineral nutrients

Introduction

Arbuscular mycorrhizal fungi are widespread and obligate plant symbionts known to play a key role in the functioning of crop productivity. These fungi establish a symbiosis with many land plants and provide mineral nutrients to the host plant in exchange for plant assimilated carbohydrates. They also form a large network of hyphae and have a great impact on soil formation and soil aggregation (Wilson et. al., 2009). The AMF are especially important for sustainable farming systems because AMF are efficient when nutrient availability is low and are bound to organic matter and soil particles. Mycorrhizal association aids in the uptake of nutrients from the soil (Harrison, 2005). Likewise, mycorrhizae species enhances growth of the plants by increased absorption of water and nutrients from soil (Singh 2003). Wheat inoculated by AMF strengthened root systems in comparison to non-inoculated plants. This results longer root length, greater root surface area, smaller root average diameter, heavier fresh and dry root weights of wheat plant.

*Corresponding author e-mail: toufiq_iqbal@yahoo.com
Wheat root surface area can also be enhanced through mycorrhizal association. Generally the stronger root systems are, the smaller the root average diameter should be, which is partly due to fibrous root system of mycorrhizae inoculated wheat plant. To get maximum wheat plant productivity, inoculation of the soil with suitable type of mycorrhizae species is necessary. Therefore, this research work was undertaken to utilize mycorrhizae species for minimizing chemical fertilizer application, enhancement soil health and nutrient uptake in improving wheat yield.

**Materials and methods**

**Experimental location and soil**

The experimental location was the Agronomy Field Laboratory, Department of Agronomy and Agricultural Extension, University of Rajshahi. Geographically, the Agronomy field laboratory is located at 24°22′36″N latitude and 88°38′27″ E longitude at an elevation of 20 m above the sea level belonging to the Agro-Ecological Zone-11 (AEZ-11) of Bangladesh, named as High Barind Tract. The soil was collected from cultivable plot of the Agronomy Farm in which paddy rice was grown before soil collection. The texture of the soil was clay. The organic matter status and soil fertility was low and the pH of the soil was in between 6.8-8.6 (Bhuiya et al., 2008). The soil was passed through a 4-mm sieve to eliminate coarse rock and plant material, thoroughly mixed to ensure uniformity and stored at 4°C before use (not more than 2 weeks). The soil was mixed thoroughly before incubation experiment. A subsample of about 0.5 kg was taken, air dried, passed through a 2-mm sieve and used for the determination of physical and chemical properties. The physio-chemical properties of the soil are shown in Table 1.

| Soil pH | AMF spores (nos/10g soil) | OM (%) | TN (%) | P (me/100g) | K (me/100g) | S (me/100g) | Ca (me/100g) | Mg (me/100g) | Fe (ppm) | Mn (ppm) | Cu (ppm) | Zn (ppm) | B (ppm) |
|---------|---------------------------|--------|--------|-------------|-------------|-------------|-------------|-------------|---------|---------|---------|---------|-------|
| 8.3     | 51                        | 1.39   | 0.08   | 12.5        | 0.16        | 14.4        | 15.63       | 1.89        | 27.3    | 13.7    | 1.26    | 0.66    | 0.50  |

Data were means of three replicates.

**Wheat variety and mycorrhizae species under this study**

The recently released Bangladesh Agricultural Research Institute (BARI) wheat variety BARI Gom 28 (Raj et al., 2012) was used as a testing plant. Four mycorrhizae species viz. *Funneliformis mosseae* (formerly *Glomus mosseae*), *Rhizophagus irregularis* (formerly *Glomus intraradices*), *Glomous Eaticatium* and *Glomus claroideum* (Rodriguez et al., 2005).

**Measurements of soil/sand physical and chemical properties**

The pH, organic carbon, the nitrogen content, available P, K were analyzed according to Page et al. (1982). Bulk soil/sand available S was determined by calcium phosphate extraction method with a
spectrophotometer at 535 nm (Petersen et. al., 1996). The Zn in the bulk soil/sand sample was measured by an atomic absorption spectrophotometer (AAS) after extracting with DTPA (Soltanpur and Schwab, 1977).

**Experimental procedure/conditions for sand culture experiment**

After AMF inoculation, sand was incubated at room temperature for 4 weeks to allow stabilization of sand chemical properties before the experiment was initiated. During incubation period, water was maintained to field capacity by weighing pot. Sand was not sterilized because it was expected that sand did not have any microbial activities. The experiment consisted of three treatments, the control treatment, the mycorrhizal (+ AMF) treatment and the non-mycorrhizal (-AMF) treatment, each being replicated 3 times and set up in a plant growth chamber. The relative humidity varied 50 to 70% during wheat seedlings sand culture experiment period. Wheat seedlings were grown for 13 days in a plant growth chamber, Department of Agronomy and Agricultural Extension, University of Rajshahi. Seedlings were regularly phoionologically observed for nutrient deficiency and any other issues for the growth period.

**Root and shoot dry weight and nutrient concentration analysis**

Roots and shoots were dried at 70 °C for 48 h and weighed for dry weight (DW) determination. Dried shoot tissue was ground, homogenized and digested with nitric and perchloric acid. For the determination of nutrient concentration analysis, ICPOES was used by dry ashing.

**Statistical analysis**

Results were analysed by a one-way or two-way analysis of variance (ANOVA) using Genstat 12th edn for Windows (Lawes Agricultural Trust, UK). One way ANOVA were conducted for treatment effects on growth and yield of wheat. Two ways ANOVA were conducted for treatment effects on nutrient distribution in root, shoot and grain of wheat. In order to investigate the effect of mycorrhizae species on bulk soil physiochemical properties data were analyzed using the Statistical Analysis System (SAS 9.1.3). All the statistical testing was performed based on $P \leq 0.05$ for least significance difference (LSD) as the critical level for the significance of Tukey test.

**Results and discussion**

**Experiment 1: Sand culture along with Swiss AMF inoculums utilization**

**Wheat seedlings growth response**

Dry wheat seedlings biomass after harvest was highest due to AMF inoculation. Interestingly, plant biomass was significantly ($P \geq 0.05$) highest in mycorrhizae amended soil than non-mycorrhizae and control pot. Dry wheat seedling biomass increased about 18% for the AMF inoculation. Dry wheat seedling biomass was 0.025, 0.028 and 0.033 g/plant for the control, without AMF and with AMF, respectively (Figure 1). Regardless of those mycorrhizae responsive was found 17.86% from the sand experiment. Similarly, mycorrhization percentage was 17.86% in this sand culture seedlings growth experiment.
Fig. 1. Dry wheat seedlings biomass after harvest for the several treatments. In the control treatments nothing was added. The-AMF means same nutrients were applied but no AMF was added. The +AMF means same nutrients with Glomus intraraduis addition. Data were mean of three replicates. Vertical bar represents LSD (P ≥ 0.05) dry wheat seedlings biomass and treatment interaction.

**Wheat seedlings tissue nutrient content**

Mycorrhizae amendment improved wheat seedlings tissue nutrient content. All measured wheat seedlings tissue nutrient was highest in AMF inoculated sand pot except Mn (Table 2). The Ca content for control, non-AMF and AMF were 2.74, 6.42 and 8.08 ppm, respectively. Interestingly, mycorrhizae amendment increase P uptake. The P content in wheat plant tissue was 1.35, 3.95 and 6.29 ppm for control, non-AMF and AMF treatment, respectively. Similarly, the K content within wheat plant tissue were 2.83, 12.73 and 22.66 ppm for control, -AMF and +AMF treatments, respectively. Likewise, the K content within wheat seedlings tissue was 1.87, 4.79 and 5.25 ppm for control, non-AMF and AMF treatment, respectively. The Mg content for control, -AMF and +AMF treatment was 2.33, 2.67 and 3.39 ppm, respectively.

**Experiment 2: Role of mycorrhizae species on wheat productivity and nutrient concentration within wheat plant tissue**

**Wheat plant growth response**

Wheat plant height differs among several mycorrhizae species amendment. Mycorrhizae species effect was found week after wheat seed sowing. The plant height at 16 DAS (Days after sowing) for *Funneliformis mosseae*, *Glomous Eaticatum*, *Rhizophagus irregularis*, *Glomus claroideum* and control were 24.19, 26.26, 25.85, 25.5, 25.29 and 24.19 cm, respectively (Fig. 2a). Similarly, the plant height at harvest for *Funneliformis mosseae*, *Glomous Eaticatum*, *Rhizophagus irregularis*, *Glomus claroideum* and control were 72.98, 72.21, 72.15, 70.96 and 67.57 cm, respectively (Fig. 2b).
Table 2. Wheat seedlings tissue nutrient content for the sand culture experiment

| Elements (ppm) | Control pot sand | Mycorrhizae added pot (+AMF) | Non-mycorrhizae added pot (-AMF) |
|---------------|------------------|-----------------------------|---------------------------------|
| Ca            | 2.744            | 8.077                       | 6.422                           |
| Cu            | 0.008            | 0.013                       | 0.009                           |
| Fe            | 1.868            | 5.246                       | 4.788                           |
| K             | 2.830            | 22.66                       | 12.73                           |
| Mg            | 2.327            | 3.392                       | 2.668                           |
| Mn            | 0.123            | 0.248                       | 0.266                           |
| P             | 1.346            | 6.290                       | 3.951                           |
| Zn            | 0.011            | 0.094                       | 0.055                           |

Fig. 2(a) and 2(b). Changes of wheat plant height due to mycorrhizae species amendment in soil. Vertical bar represents LSD (P ≥ 0.05) for treatment interaction. Data were means of three replicates.

Root proliferation of wheat plant occurs due to mycorrhizae amendment in soil. The root dry weight for *Funneliformis mosseae, Glomous Eaticatium, Rhizophagus irregularis, Glomus claroideum* and control were 3.73, 3.60, 2.47, 2.49 and 2.20 g/plant, respectively (Fig. 3a). The shoot dry weight for *Funneliformis mosseae, Glomous Eaticatium, Rhizophagus irregularis, Glomus claroideum* and control were 11.12, 10.82, 10.77, 10.41 and 9.53 g/plant, respectively (Fig. 3b).

Wheat plant yield response and chlorophyll content

Mycorrhizae species inoculation significantly (P ≥ 0.05) differ wheat grain yield. The wheat grain yield for *Funneliformis mosseae, Glomous Eaticatium, Rhizophagus irregularis, Glomus claroideum* and control were 6.16, 5.08, 4.96, 4.16 and 4.16 g/plant, respectively (Fig. 4a).
The role of chlorophyll in photosynthesis is vital during photosynthesis, chlorophyll captures the sun light and synthesize carbohydrates, which allows the plant to grow. The chlorophyll content in wheat plant for *Funneliformis mosseae*, *Glomous Eaticatium*, *Rhizophagus irregularis*, *Glomus claroideum* and control were 41.21, 40.52, 40.20, 40.18 and 40.08, respectively (Fig. 4b).
Effect of mycorrhizae species inoculation within wheat plant tissue

The potassium content in shoot tended to be increased due to mycorrhizae species inoculation. However, no changes were found in root and grain due to mycorrhizae species inoculation. Most of the K was accumulated in shoot and grain (Fig. 5a). The magnesium accumulate maximum in the wheat grain for the *Rhizophagus irregularis* mycorrhizae species inoculation followed by *Funneliformis mosseae*, *Glomus Eaticatium*, *Glomus claroideum* and control. The Mg did not change remarkably in wheat root and shoot due to mycorrhizae species inoculation (Fig. 5b).

![Graph showing potassium and magnesium content](image)

Fig. 5. (a) Potassium and (b) Magnesium content in root, shoot and grain of wheat due to mycorrhizae amendment. Vertical bar represents LSD ($P \geq 0.05$) for plant biomass x treatment interaction. Data were mean of three replicates.

![Graph showing phosphorus and iron concentration](image)

Fig. 6. (a) Phosphorus and (b) Iron concentration in root, shoot and grain due to mycorrhizae amendment. Vertical bar represents LSD ($P \geq 0.05$) for plant biomass x treatment interaction. Data were mean of three replicates.
Mycorrhizae species amendment significantly \((P \geq 0.05)\) increased P accumulation in wheat grain. The *Funnelliformis mosseae* mycorrhizae species accumulate highest P in wheat grain. Likewise, *Glomus Eaticatium* mycorrhizae species accumulates second highest P in wheat grain. Most of the P accumulated in grain than root and then shoot. Root and shoot P content did not change remarkably due to mycorrhizae species inoculation (Fig. 6a). Almost all iron accumulated within root tissue of wheat plant. The *Funnelliformis mosseae* mycorrhizae species tended to be accumulated highest Fe in wheat grain. Less amount of Fe accumulate in shoot tissue. Very small amount of Fe translocated in wheat grain. However, no significant difference was found in shoot and grain Fe content in wheat among treatments (Fig. 6b).

The mycorrhizal plants had higher shoot P and Fe concentrations than non-mycorrhizal plants at all samplings regardless of soil moisture conditions (Fig. 6a and 6b). VAM fungi assist the plants to absorb mineral nutrients from the soil, particularly low available elements like P, and Mo (Mosse, 1973). These fungi are also reported to consistently stimulate plant absorption of Zn and Cu and also increase plant resistance to various stresses like water, drought, salt and heavy metal toxicity (Jackson and Mason, 1984). Effective nutrient acquisition by VAM-fungi is generally attributed to the extensive hyphal growth beyond the nutrient depletion zone surrounding the root (Tisdle *et al.*, 1995).

**Effect of mycorrhizae on soil properties**

Bulk soil physiochemical properties affected by mycorrhizae inoculation. Bulk soil pH increased 0.30 units due to mycorrhizae inoculation. Similarly, bulk soil organic matter increased 0.30% due to mycorrhizae inoculation. Likewise, bulk soil N improved 0.02% for the several mycorrhizae inoculation (Table 3). The ability of mycorrhizal hyphae to extend and explore a greater area of soil than the host plant’s roots can reach has been demonstrated. The fungus seems to be a great help in the acquisition of soil nutrients, reaching micro and macro elements which the plant alone could not reach (Table 3). In this way, the depletion zone created via a plant’s rapid nutrient uptake in the proximity of its root system (Bucher, 2007) can be bridged, and an adequate supply of nutrient elements is translocated to the plant (Javaid, 2009).

**Table 3. Bulk soil physiochemical properties of the wheat experiment that grown with mycorrhizae species inoculated soil.**

| Treatment                  | Soil pH (in H₂O) | SOM (%) | N (%) | P (ppm) | K (me/100g) | S (ppm) | Ca (ppm) | Mg (me/100g) | Fe (ppm) | Mn (ppm) | Cu (ppm) | Zn (ppm) | B (ppm) |
|----------------------------|------------------|---------|-------|---------|-------------|---------|---------|-------------|---------|---------|----------|---------|--------|
| Control                    | 8.10b            | 1.36c   | 0.08b | 12.30a  | 0.18a       | 0.89b   | 23.60b  | 3.22a       | 16.10a  | 13.17a  | 0.77a    | 0.87c   | 0.50a  |
| *Funnelliformis mosseae*   | 8.40a            | 1.53a   | 0.09ab| 10.87a  | 0.19a       | 1.74b   | 24.89a  | 3.52a       | 15.97a  | 13.27a  | 0.79a    | 1.52ab  | 0.66a  |
| *Rhizophagus irregularis*  | 8.37b            | 1.50a   | 0.09ab| 23.47a  | 0.19a       | 2.54b   | 25.48a  | 3.30a       | 14.80ab | 13.80a  | 0.72a    | 0.90bc  | 0.54a  |
| *Glomus Eaticatium*        | 8.33b            | 1.64ab  | 0.10a | 11.90a  | 0.19a       | 3.99ab  | 24.97a  | 3.43a       | 15.07ab | 11.77a  | 0.73a    | 1.16a   | 0.56a  |
| *Glomus claroideum*        | 8.20b            | 1.66bc  | 0.10a | 13.83a  | 0.19a       | 8.45a   | 24.65ab | 3.37a       | 13.87b  | 13.03a  | 0.72a    | 1.15a   | 0.54a  |
Conclusions

The results showed that mycorrhizal fungi can contribute to wheat plant productivity because they help to uptake nutrients from soil solution. Moreover, mycorrhizal fungi can directly and indirectly contribute to carbon sequestration within plant tissue and soil. Mycorrhizal effects include enhanced nutrient uptake, enhanced seedling establishment and stimulation of soil structure. Findings also showed that mycorrhizae accumulate phosphorus within wheat plant tissue. Additional research is needed to develop farming systems that optimize the use of natural resources such as mycorrhizal fungi for sustainable agricultural production.

Acknowledgements

The author is thankful to Ministry of Science and Technology, Bangladesh to provide grants for this research.

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