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Response of Mung Bean Plants to Arbuscular Mycorrhiza and Phosphorus in Drought Stress

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ABSTRACT: In order to evaluate drought stress and arbuscular mycorrhiza with phosphorus on related root and shoot traits and grain yield of mungbean, a factorial experiment was carried out based on a randomized completely design in pot culture. Irrigation regimes 25, 50, 75 and 100mm of evaporation from a pan possessed irrigation's levels as the first factor. At the second factor arranged 5, 15mgPKg^{-1} soil and 5, 15 with *Glomus mosseae* with three replications were conducted in Urmia University in 2010. Results showed that grain yield of inoculated mungbean with 15 and 5mgPKg^{-1} soil 834.17 and 699.317mg/plant had the highest values, respectively. Both 15 and 5mgPKg^{-1} soil with *G. mosseae* had more leaf phosphorus, plant height, leaf number, leaf dry weight, pod number, seed/pod, root dry weight, root length, root volume, and chlorophyll index than 15 and 5mgPKg^{-1} soil non-inoculated plants. Leaf phosphorus, plant height, leaf number, leaf dry weight, pod number, seed/pod, root dry weight, root length, root volume, and chlorophyll index had positive correlation coefficients with grain yield. The highest (743.33mg/plant) and lowest (423.33mg/plant) grain yield achieved in irrigation after 25 and 100mm evaporation, respectively. With increasing water deficit stress decreased leaf phosphorus, leaf number, leaf dry weight, seed/pod, root dry weight and chlorophyll index. Although drought stress reduced grain yield, but inoculated it reduced the severity of stresses. Inoculated plants increased 69% of the potential yield than control.

KEYWORDS: *Glomus mosseae*, Grain yield, Phosphorus, *Vigna radiata*, Water stress.

1 INTRODUCTION

Mycorrhizal plants can be affected water balance under irrigated and drought stress conditions [23]. Symbiotic plants with *G. intraradices* increase biotic and a-biotic stresses. There were several mechanisms for expression increasing root hydraulic conductivity and improvement root contact with soil particles through connecting mycorrhizal hyphae [7]. At mycorrhizal faba-bean enhanced nodule number, nodule dry weight, days to flowering, number of pods and grain yield compared to non-mycorrhizal plants under different irrigation regimes [15]. AM fungi by increasing mineral nutrients, especially phosphorus tolerate to biotic and a-biotic stresses [28], [29], [2].

1.1 PHOSPHORUS

Phosphorus is one of the essential mineral macronutrients, which is required for maximum yield of agriculturally important crops. Most of the essential plant nutrients, including phosphorus, remain in insoluble form in soil [1], [33].

Phosphorus is critical for plant growth, and is a component of the nucleic acid structure of plants and bio-membranes. Therefore, it is important in cell division and tissue development. Phosphorus is also involved in the energy metabolism of cells and is required for the biosynthesis of primary and secondary metabolites in plants. Consequently, plants have evolved a range of strategies to increase phosphorus uptake and mobility [18].

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1.2 MYCORRHIZAE AND DROUGHT STRESS

Inoculated plants with arbuscular mycorrhizal (AM) fungi can improve crop production under drought stress conditions [3], [4]. Several studies demonstrated that the symbiotic relationship between AM fungi and the roots of plants increased drought resistance in host plants [6], [24], [29], [26]. Mycorrhiza is a symbiotic fungus that caused beneficial relationship between soil and plant. Increasing of water absorption and nutrient (especially phosphorus) uptake by mycorrhizal hyphae can be more due to growth hyphae to the 20mm root surface than 1.5mm hairy roots [30]. Also, low root penetration compared to high hyphae penetration into cracks and pores of soil. Hyphae of fungus infiltrate into the soil where the roots are unable to penetrate. Speed of inorganic phosphate into the hyphae was 2cm/h which is several times higher than diffusion in the soil [21], [23], [24].

The rate of photosynthesis improved due to more phosphorous absorption and chlorophyll content in mycorrhizal pepper plants [10]. In mycorrhizal mung bean plants, grain yield, leaf phosphorus and ecosystem water use efficiency were improved compared with the non-mycorrhizal plants. Two species of mycorrhiza, *G. mosseae* and *G. intraradices* significantly improved the grain yield and reduced the water-deficit stress in the field [13]. AM symbiosis of corn plants by improving water absorbing, changing water relations, expanding root system, improving plant nutrition and increasing plant metabolism tolerated to water deficit [8]. At mycorrhizal sesame enhanced roots through increasing volume and dry weight of root [9]. In another experiment, lavender plants inoculated with *G. mosseae* expanded roots 35% [19].

The aim of this study was to evaluate the effect of *G. mosseae* with phosphorus, on grain yield, root and shoot traits of mung bean plants under different levels of irrigation.

2 MATERIALS AND METHODS

2.1 EXPERIMENTAL LOCATION

A trial was conducted in the Agricultural Faculty of Urmia University in Iran. The experiment located in longitude 37°, 39' north, latitude 44°, 58′ east and 1365m altitude. Environmental conditions of the experimental site, including the highest and lowest temperatures and humidity, sum of sunny hours, daily and monthly solar radiation and potential evapo-transpiration of the study are shown in Table 1. Some physicochemical properties of soil, which is used in 240 pots were determined (Table 2).

2.2 EXPERIMENTAL DESIGHN

A factorial experiment based on a randomized completely design carried out with three replications. Irrigation regimes with four levels of 25, 50, 75 and 100 mm of evaporation from a Class A pan and inoculation mycorrizal mungbean (*Vigna radiata* L.) cultivar NM92 with four levels, including 5, 15 mg P Kg^{-1} soil and 5, 15 with *Glomus mosseae* of mycorrhiza species arranged as the first and second factors, respectively. Each plot consists 5 pots which all was 240 pots. Depth and diameter of pots were 22 cm which filled with 7kg of soil. Seeds of the mung bean cultivar NM92 were provided by the Agricultural Research Station of Dezfol. A species of AM fungi used in this study was *G. mosseae*, which were produced on maize (Zea mays L.) host plants by Dr. E.M. Goltapeh at Tarbiat Modarres University, Tehran, Iran. The mycorrhizal inoculum was placed in the holes (30 g per hole) below the mungbean seeds and lightly covering with soil from the hole on the day of planting. For non-mycorrhizal control plants were sown with no inoculation. Seeds were sown on 29 June 2010 into a Sandy loam soil with pH 7.65 and 2.4 mg kg^{-1} of P. At three primary leaf stage were applied irrigation regimes. Total water consumption during the growing season was 84, 51, 39 and 27 liters at per pot for irrigations of 25, 50, 75 and 100 mm of evaporation from a Class A pan, respectively. Root and shoot dry weights were determined after drying samples in oven 72°C. Length and volume of roots measured from 10 randomly selected plants at the end of the growing season. Chlorophyll index was determined *in vivo* using a Minolta SPAD CCM-200 Chlorophyll-meter featuring integrated data logger. Grain yield recorded from all pots of each treatment. At the maturity time, the percentage of colonization of mung bean roots by AM fungi was determined on 15 plants per experimental unit. Root colonization was measured in fresh roots cleared in 10% KOH for 10 min at 90°C and stained in 0.05% lactic acid–glycerol–Trypan Blue [22]. The percentage of root colonization by arbuscular mycorrhizal fungi was microscopically determined using the grid line intersection method [11]. To measure leaf phosphorus, dried leaves were milled, digested, and analyzed as described by Watanabe and Olsen [31] and Ohnishi et al. [20]. The method described for phosphorus involves drying, homogenization, and combustion (4 h at 500°C) of the leaf sample. The plant ashes (5 mg) are digested in 1 ml of concentrated HCl. The samples are then filtered, and total phosphorus is quantified as PO_4^- using the ascorbic acid method [31] . The amount of PO_4^- in solution was determined colorimetrically at 882 nm [12].

2.3 STATISTICAL ANALYSIS

Analysis of variance of the data was performed using MSTATC software. The effects of irrigations, application of mycorrhizae and phosphorus were analyzed by ANOVA and the means compared with the Duncan's Multiple Range test ($P \leq 0.05$).

3 RESULTS AND DISCUSSION

Different levels of irrigations and mycorrhizae with phosphorus for traits of leaf phosphorus, leaf number, leaf dry weight, seed/pod, root dry weight, chlorophyll index and grain yield were significant differences (Table 3).

3.1 RELATED ROOT AND SHOOT TRAITS

Colonization percentage of *G. mosseae* reduced with increasing water stress and phosphorus. Variations of this trait were from %29.24 to %48.79. Treatment of *G. mosseae* with 15 mg P Kg^{-1} soil had the most plant height (21.63cm), leaf number (9.61), leaf dry weight (0.32g), pod number (2.69), root dry weight (0.19g), root length (23.61cm), root volume (0.45cm^3), and chlorophyll index (75.75) (Table 4). Chlorophyll index, dry weight of roots and leaves decreased with severe stress. Irrigation levels of 25 and 100 mm of evaporation from a Class A pan were 70.64, 0.19g, 0.33g and 53.56, 0.11g, 5.80g values of them, respectively. Leaf number and seed/pod in 25, 50 and 75 mm of evaporation from a Class A pan were the same group, but at 100 mm of evaporation were reduced (Table 5). Expanded roots of mychorrhizal plants enhanced root area. Therefore, water uptake in mycorrhizal plants was due to more root expansion than control [14]. Mycorrhizal corn plants through the expanding root system, improving hydraulic conductivity and water uptake increase drought tolerance [8]. In lavender inoculated plants with *G. mosseae* improved root growth (35%) than control [19].

3.2 OSMOTIC COMPONENTS

Phosphorus accumulation in leaves of inoculated plants at both *G. mosseae* with 15 and 5 mg P Kg^{-1} soil was higher (236.28 and 228.62mg/100g dry leaf) than control. The highest and lowest accumulations were allocated to 25 and 100 mm of evaporation from a Class A pan with 227.23 and 194.60mg/100g dry leaf, respectively (Tables 5 and 6). Nutrient uptake under drought stress decreased through reducing transpiration, disruption of active transport systems and membrane permeability and reducing root absorption. mycorrhizal hyphae uptake fixed phosphorous where plant roots couldn't absorb [17]. Influence of AM plants on leaf phosphorus in this experiment coordinated by other researchers [5], [25], [28], [29], [2], [32].

3.3 GRAIN YIELD

Grain yield in 25, 50 and 75 mm of evaporation from a Class A pan were the same group, but at 100 mm of evaporation with severe stress was reduced grain yield. *G. mosseae* with 15 mg P Kg^{-1} soil was higher grain yield (834.17 mg/plant) than control. Grain yield of mycorrhizal plants with 5 mg P Kg^{-1} soil was equal treatment 15 mg P Kg^{-1} soil without inoculation with mycorrhizal plants (Tables 5 and 6). Grain yield differences in mycorrhizal treatments are related to increasing water absorption and mineral nutrients [5], [10], [15], [16], [21], [23], [27].

3.4 RELATIONSHIPS OF TRAITS

Leaf phosphorus (r = 0.60**), plant height (r = 0.70**), leaf number (r = 0.70**), leaf dry weight (r = 0.73**), pod number (r = 0.68**), seed/pod (r = 0.80**), root dry weight (r = 0.69**), root length (r = 0.51**), root volume (r = 0.68**), and chlorophyll index (r = 0.65**) had positive correlation coefficients with grain yield. These observations indicate that plants having a higher leaf phosphorus, leaf dry weight, root dry weight and seed/pod produce higher grain yield.

4 CONCLUSION

Inoculated plants with *G. mosseae* showed more leaf phosphorus, plant height, leaf number, leaf dry weight, pod number, seed/pod, root dry weight, root length, root volume, and chlorophyll index than control. With decreasing water deficit stress, increased leaf phosphorus, leaf number, leaf dry weight, seed/pod, root dry weight and chlorophyll index and consequently will lead to increase grain yield. Relationships between traits showed that with increasing leaf phosphorus, root dry weight, leaf dry weight and seed/pod in inoculated mycorrhizal mung bean plants enhanced grain yield. The overall results of this study showed that in low amounts of soil phosphorus (5 mg P Kg^{-1} soil), mycorrhiza can be functionally equivalent with 15 mg P Kg^{-1} soil non-mycorrhizal plants. Therefore recommended in areas with low P soil content, *G. mosseae* of mycorrhizal species be used as a biological fertilizer in mung bean plants.

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ANNEXE

Table 1. Environmental condition at the experimental site during summer 2010

Parameter	June	July	August	September
Highest temperature (°C)	32.4	37.9	38.0	32.6
Lowest temperature (°C)	5.2	9.7	10.1	7.9
Highest relative humidity (%)	76	65	65	81
Lowest relative humidity (%)	29	21	18	28
Sum of sunny hours (no.)	360	389	362	325
Solar radiation (MJ m^{-2} d^{-1})	27.6	28.1	25.3	21.2
Solar radiation (MJ m^{-2} mo^{-1})	828.0	871.0	784.3	636.0
Potential evapo-transpiration (mm mo^{-1})	215	275	236	175

Table 2. Some soil physico-chemical characteristics

Saturation (%)	Electrical conductivity (Ds m^{-1})	PH	Organic carbon (%)	Phosphorus (Mg kg^{-1})	Potassium (Mg kg^{-1})	Soil texture
29	1.3	7.65	0.20	2.4	85	Sandy loam

Table 3. Mean squares traits of mung bean affected by mycorrhizal infection with phosphorus under different irrigation regimes

S.O.V	df	Mean squares										
		Grain yield	Leaf phosphorus	Plant height	Leaf number	Leaf dry weight	Pod number	Seed/pod	Root dry weight	Root length	Root volume	Chlorophyll index
Irrigation (I)	3	0.19**	3486.07**	8.14	21.93**	0.044**	0.17	15.97**	0.015**	28.10	0.018	613.68**
Mycorrhizae (M) + Phosphorus(P)	2	0.58**	6456.79**	74.63**	30.58**	0.043**	3.56**	16.46**	0.015**	104.41**	0.127**	1858.24**
M P× I	6	0.02	622.94	0.36	0.32	0.001	0.12	0.42	0.001	2.24	0.004	32.38
Error	24	0.02	753.20	4.91	1.59	0.002	0.17	0.92	0.001	7.45	0.004	126.234
CV (%)	-	5.87	12.80	11.79	16.41	16.41	21.00	19.56	16.07	13.53	17.26	18.32

** Significant at the 5% probability level.*

*** Significant at the 1% probability level.*

Table 5. Means comparison of mung bean traits by mycorrhizae with phosphorus

Mycorrhizal Symbiosis + phosphorus (mg P kg^{-1} soil)	Grain yield (mg/plant)	Leaf phosphorus (mg/100g dry leaf)	Plant height (cm)	Leaf number	Leaf dry weight (g)	Pod number	Seed/pod	Root dry weight (g)	Root length (cm)	Root volume (cm^3)	Chlorophyll index
5	337.50c	184.52b	15.75c	5.90c	0.18c	1.42c	3.57c	0.11c	16.68c	0.22c	47.36c
15	567.50b	208.02ab	18.08bc	7.01bc	0.24b	1.74bc	4.53bc	0.15b	19.18bc	0.33b	55.58bc
Glomus mosseae +5	699.17b	228.62a	19.71ab	8.21ab	0.29ab	2.10b	6.37a	0.17ab	21.22b	0.41a	66.67ab
G. mosseae + 15	834.17a	236.28a	21.63a	9.61a	0.32a	2.69a	5.16b	0.19a	23.61a	0.45a	75.75a

Means followed by the same letter(s) in each column are not significant differences

Table 6. Means comparison of mung bean traits by irrigation regimes

Irrigation regimes	Grain yield (mg/plant)	Leaf phosphorus (mg/100g dry leaf)	Plant height (cm)	Leaf number	Leaf dry weight (g/plant)	Pod number	Seed/pod	Root dry weight (g/plant)	Root length (cm)	Root volume (cm^3)	Chlorophyll index
25	743.33a	227.23ab	-	8.85a	0.33a	-	36.05a	0.19a	-	-	70.64a
50	667.50a	229.88a	-	8.44a	0.28a	-	5.62a	0.16ab	-	-	62.19ab
75	604.17a	205.28ab	-	7.64a	0.22b	-	4.46a	0.14c	-	-	58.97ab
100	423.33b	194.60b	-	5.80b	0.19b	-	3.50b	0.11c	-	-	53.56b

Means followed by the same letter(s) in each column are not significant differences

Table 7. Correlation coefficients between mung bean traits

Treatment	Grain yield	Leaf phosphorus	Plant height	Leaf number	Leaf dry weight	Pod number	Seed/pod	Root dry weight	Root length	Root volume
Leaf phosphorus	0.60**									
Plant height	0.70**	0.58**								
Leaf number	0.70**	0.51**	0.62**							
Leaf dry weight	0.73**	0.56**	0.63**	0.76**						
Pod number	0.68**	0.41**	0.61**	0.65**	0.52**					
Seed/pod	0.80**	0.65**	0.58**	0.75**	0.85**	0.54**				
Root dry weigh	0.69**	0.65**	0.58**	0.73**	0.79**	0.61**	0.58**			
Root length	0.51**	0.28	0.41**	0.27	0.32*	0.47**	0.72**	0.16		
Root volume	0.68**	0.57**	0.70**	0.60**	0.69**	0.62**	0.61**	0.73**	0.46**	
Chlorophyll index	0.65**	0.47**	0.65**	0.70**	0.78**	0.59**	0.73**	0.72**	0.35*	0.76**

** and ** Significant at P≤0.05 and P≤0.01, respectively*