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Mycorrhizal fungi influence on quantitative and morphological characteristics in Basil induced by phosphorus fertilizer under water deficit conditions

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In order to investigate the effects of mycorrhiza (*Glomus Fasciculatum*) and phosphorus fertilizer on grain yield, biological yield, harvest index, plant height and number of branch in basil under water deficit condition, an experiment was conducted as split plot in randomized complete block design with four replications at Iran in 2011. The main-plot factor included: Two levels of irrigation regime (I_1 and I_2 irrigation after 40 and 80 mm evaporation respectively) while the sub-plot factors included phosphorus fertilizer application (P_1,P_2 and P_3 application 0, 35, 70 kg/ha respectively) and mycorrhiza inoculation at two levels (M_1 = non- application and M_2 = application of mycorrhiza). The results showed that the traits were all highly significantly different under water deficit condition. Phophorus and mycorrhiza affected grain yield and biological yield significantly .The results also showed that the highest grain yield, biological yield, harvest index, plant height and number of branch were obtained from application of phosphorus fertilizer (70 kg/ha) under normal irrigation.

Key words: Drought stress, Glomus *Fasiculatum*, Basil, phosphorus.

INTRODUCTION

Application of biofertilizer, especially plant growth promoting rhizobacteria (PGPR) and mycorrhiza fungus is one of the most important strategies for plant nutrition compared to chemical fertilizers, especially in sustainable management of agroecosystems (Babai, 1995). Mycorrhizal fungi live in a symbiotic relationship with plants. They grow in close association with the roots and play an important role in the connections and transfer of soil nutrients to the plant (Tilak, 1992; Turan et al., 2006). Mycorrhiza are important for the establishment growth and survival of seedling, particularly in marginal habitats, where the symbiosis improves stress tolerance as well as conserves soil structures (Sharma, 2002). Vivas (2003) shows that the increased metabolically active fungal biomass in inoculated plants was independent of phosphorus levels and was not related to phosphorus uptake from the poor nutrients soil. One third of the world lands are classified an arid and semi-arid region and the remains are faced with water seasonal or local fluctuations (Beweley and Krochko, 1982). Availability of water rather than land is the main constraint on agricultural production in arid and semi-arid environment (Bannayan et al., 2008; Elikaei et al., 2008). The potential of medicinal and aromatic plants for growing under limited water conditions make them suitable alternative crops in such agro-ecosystems (Koocheki and Nadjafi, 2003; Haj, 2011). Various studies have shown that water stress and biofertilizers affected yield and yield components of medicinal plants such as *Nigella sativa* (Karimi-Yeganeh et al., 2010; Valadabadi et al., 2010);

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Table 1. The results of soil analysis.

Soil texture	Sand (%)	Silt (%)	Clay (%)	EC (Ds/m)	pH	Total [N(%)]	P (Mg/kg)	K (Mg/kg)	Organic matter (%)	Depth of sampling (cm)
Sa.Si	58.72	22.09	19.9	0.75	7.90	0.09	38	21	0.88	0-30

Table 2. Mean temperature and rainfall during May until September 2011.

Parameter	May	Jun	Jul	Aug	Sep
Rainfall (mm)	28	12	5	0	5
Temperature (℃)	18	21	26	24	22

and *Mentha piperita* (Khorasaninejad et al., 2011). Basil *(Ocimum basilicum* L.) is originally native to india and other tropical regions of Asia, having been cultivated there for more than 5000 years (Copetta et al., 2006). Basil is an annual plant and it grows between 30 and 130 cm tall, with opposite, light, green and silky leaves. The flowers are small, white in colour and arranged in a terminal spike. Recently, there has been much research into the health benefits conferred by the essential oils found in basil. Scientific studies *in vitro* have established that compounds in basil oil have potent antioxidant, antiviral and antimicrobial properties (Chiang et al., 2005; Almeida et al., 2007). So, the main objective of this study was to investigate the effects of mycorrhizae and phosphorus fertilizer under irrigation deficit condition on quantitative and morphological characteristics in basil.

MATERIALS AND METHODS

This study was conducted during the growing season of 2011 at the experimental field of Islamic Azad University, Shahr-e-Qods branch at Iran (27 38 N, 40 21 E ;1417 m), with sandy loam (Table 1). Meteorological data on rainfall and mean temperature from May until September 2011 showed that Table 2 and rainfall in the study area is distributed with an annual mean of 215 mm. The experimental design was split-plot based on Randomized Complete Block Design (RCBD) with four replications. The main-plot factor included: Two levels of irrigation régime (I_1 and I_2 irrigation after 40 and 80 mm evaporation from class A pan respectively) while the sub-plot factors included phosphorus fertilizer application as super phosphate (P_1,P_2 and P_3 application 0, 35 and 70 kg/ha respectively) and mycorrhiza inoculation at two levels (M_1 = non-application and M_2= application of mycorrhiza). Phosphorus fertilizer was applied pre-planting and it was added as super phosphate at different levels of 0 (unfertilized), 35 and 70 kg/ha. Mycorrhizal inoculation was done at sowing by applying 3 g of mycorrhizal inoculum and it was moistened with 2% concentration water sugar and soaked 2 kg inoculants/100 kg seed. Weeds were controlled manually. All necessary cultural practices and plant protection measures were followed uniformly for all the plots during the entire period of experimentation. The traits consisted of number of branch, harvest index, plant height, grain yield and biological yield. At the beginning of the flowering period, plant height and number of branch were measured. Dry weight of whole plants was calculated in each plot after drying the plants in the oven (75 for 48 h) (Darzi et al., 2012). Final seed yield was measured from 1 m^2 of each plot. Harvest index was determined by the following formula:

Harvest index= Economical yield /Biological yield

All data were subjected to statistical analysis (ANOVA) using SAS software. Mean of comparisons were performed by Duncan multiple range test (DMRT) at 5%.

RESULTS

The results indicated that the most of traits were significantly affected by irrigation, phosphorus and mycorrhiza treatments (Table 3).

Plant height

Plant height did not respond to mycorrhiza and phosphorus treatments, but water deficit condition had significant effects on plant height (Table 3) and we obtained highest plant height with normal irrigation (61.3 cm) (Table 4). The results showed that non-significant difference between interaction effects of mycorrhiza, phosphorus and irrigation. Freitas et al. (2004) indicated that inoculation of *Mentha arvensis* with mycorrhiza resulted in increased plant height.

Number of branches

Results of variance analyzing showed that only water deficit effect was significant on number of branches at 5% probability level (Table 3). Mean comparison indicated that highest of number of branches was obtained under normal irrigation (Table 4). Copetta et al (2006) have shown that inoculation of plants with mycorrhiza can result in a significant change in number of branches and number of follicles per plant.

Grain yield

The results indicated that water deficit, mycorrhiza and

Table 3. Analysis of variance.

S.O.V	Mean square					
	df	Biological yield	Plant height	Number of branch	Grain yield	Harvest Index
Replication	3	2784427.439**	346.65	4.347	193579.886**	0.0001
Irrigation(I)	1	200069341.98**	7874.563**	92.13**	8811994.332**	0.0001
E(a)	3	4426.025	37.723	2.701	3882.029	0.0001
Phosphorus (P)	2	1037099.492**	31.72	1.97	122925.354**	0.0001
(M).(P)	2	89182.982	17.671	2.183	9158.4	0.0001
Mycorrhiza(M)	1	1525025.622**	209.167	0.047	203525.065**	0.0001*
(M).(I)	1	459808.1*	5.88	3.05	76493.086**	0.0001*
(P).(I)	2	334679.076**	16.459	1.258	56418.305**	0.0001*
(M).(P).(I)	2	2609.41	4.107	4.421	1696.312	0.0001
E(b)	30	61695.11	33.233	2.73	7164.741	0.0001
CV(%)		4.52	11.89	17.37	7.16	3.09

*,** significant at 5 and 1% levels, respectively.

Table 4. Mean comparison of traits at various levels of treatments (simple effect).

Treatments	Biological yield (kg/ha)	Plant height (cm)	Number of branch	Grain yield (kg/ha)	Harvest index (%)
(M_2)	5705.5[a]	50.6[a]	9.5[a]	1247.5[a]	20.1[a]
(M_1)	5319[b]	46.4[a]	9.5[a]	1107.3[b]	20.25[a]
(P_1)	5035.8[c]	46.9[a]	9.1[a]	988.2[c]	19.6[a]
(P_2)	5495.5[b]	49.2[a]	9.8[a]	1188.7[b]	21.6[a]
(P_3)	5702.3[a]	49.4[a]	9.6[a]	1249[a]	21.9[a]
(I_1)	7538.8[a]	61.3[a]	11[b]	1610.48[a]	21.36[a]
(I_2)	3455.6[b]	35.7[b]	8.1[b]	600.3[b]	17.37[b]

Means within the same column and row factors, followed by same letter are not significantly difference ($p<0.05$).

phosphorus fertilizer influence significantly grain yield (1%) (Table 3). Mean comparison showed that grain yield varied between 600.0 and 1610 kg/ha, which were obtained from irrigation deficit condition and normal irrigation, respectively. Phosphorus was affected grain yield significantly (1%) (Table 3); this is due to effect of phosphorus on grain filling stage (Wu et al., 2005). Grain yield increased with the use of mycorrhiza. Interaction of irrigation-phosphorus and irrigation-mycorrhiza affected grain yield significantly (1%) (Table 3, 5 and 6) and with the use of phosphorus (70 kg/ha) grain yield increased up to1249 kg/ha and the other hand application of mycorrhiza increased it to 1247.5 kg/ha (Table 4); but the interaction of mycorrhiza-phosphorus and mycorrhiza - irrigation- phosphorus did not differ significantly.

Biological yield

Variance analyzing of biological yield data showed that biological yield was significantly influenced by mycorrhiza, phosphorus and irrigation (Table 3). Mean comparison of the application of phosphorus showed that the use of it (70 kg/ha) yielded high biological yield (5702.3 kg/ha), (Table 4). Mycorrhiza affected biological yield significantly (1%) (Table 3), and we obtained high biological yield with the use of mycorrhiza (5705.5 kg/ha) (Table 4). Irrigation deficit condition affected biological yield significantly (1%) and under water deficit it decreased from 7538 to 3455.6 kg/ha. Interaction of irrigation-phosphorus and irrigation-mycorrhiza affected biological yield significantly (1 and 5% respectively) (Table 3, 5 and 6). The interaction of mycorrhiza-phosphorus and mycorrhiza-irrigation-phosphorus did not differ significantly (Table 3). On the basis of average performance, the highest values of biological yield (7709 kg/ha) was produced with application of phosphorus (70 kg/ha) under normal irrigation (irrigation after 40 mm evaporation) (Table 6).

Harvest index

The results of variance analyzing indicated that mycorrhiza

Table 5. Mean comparisons of interaction effects of irrigation and mycorrhiza on traits.

Treatments	Biological yield (kg/ha)	Grain yield (kg/ha)	Harvest index (%)
(M1) (I1)	6731^{a}	1521^{a}	19.75^{a}
(M1) (I2)	3271^{c}	586^{c}	16.25^{c}
(M2) (I1)	6821^{a}	1486^{a}	19.26^{a}
(M2) (I2)	3921^{b}	921^{b}	17.26^{b}

Means within the same columns and row factors, followed by same letter are not significantly difference ($p<0.05$).

Table 6. Mean comparisons of interaction effects of irrigation and phosphorus on traits.

Treatments	Biological yield (kg/ha)	Grain yield (kg/ha)	Harvest index (%)
(P1) (I1)	6129^{b}	1106^{c}	19.25^{b}
(P2) (I1)	6572^{a}	1349^{b}	19.75^{b}
(P3) (I1)	7709^{a}	1679^{a}	22.45^{a}
(P1) (I2)	2725^{e}	508^{f}	16.31^{d}
(P2) (I2)	3141^{d}	671^{e}	17.85^{c}
(P3) (I2)	3497^{c}	705^{d}	17.95^{c}

Means within the same column and row factors, followed by same letter are not significantly difference ($p<0.05$).

influenced, while phosphorus fertilizer had not significant effect on harvest index, but irrigation effected on it significantly (5%) (Table 3) and under less irrigation harvest index 19% decreased (Table 4). The interaction of mycorrhiza-irrigation and phosphorus-irrigation affected harvest index significantly (5%) (Table 3,5and6) and we obtained the highest values of harvest index (22.45) with application of phosphorus (70 kg/ha) under normal irrigation (Table 6).

DISCUSSION

As it was shown in the results of this study, significantly different in agro-morphological traits were observed following application of mycorrhiza and phosphorus fertilizer under irrigation deficit conditions. We obtained at least of biological yield, grain yield, number of branch, harvest index and plant height in basil under irrigation deficit biomass production in the plants (Junqueira and Oliveira, 1997). Similar results were obtained by Haj Seyed Hadi et al. (2012), Merrien (1996) and Xia (1994). Khalid (2006) reported that irrigation treatments have significant effects on fresh and dry weights in *Ocimum basilicum* L. and *Ocimum americanum* L. The results showed that quantitative traits (biological yield and grain yield) were affected by water deficit than morphological traits (plant height and number of branch) and with use of phosphorus fertilizer and mycorrhiza limited effects of water stress. Mycorrhiza enhanced growth production of basil. It seems that mycorrhiza increased plant water and nutrient supply by extending the volume of soil accessible to plants. Mycorrhiza improved the uptake of phosphorus, similarly mycorrhiza application with phosphorus fertilizer (35 kg/ha) under normal irrigation with non-application mycrrhiza and application of phosphorus (70 kg/ha). Gupta et al. (2002) found that *G. fasciculatum* inoculated mint plants depleted the available N, P and K in the rhizosphere soil as compared to non inoculated control plants, however the extent of nutrient depletion was greater for P than N and K. The results indicated that water deficit during the vegetative period can result in shorter plants and smaller leaf areas of mint (Abbaszadeh et al., 2008), and chicory (Taheri et al., 2008), and it reduced water use due to the reduction in plant size of calendula (Rahmani et al., 2008). Therefore, application of mycorrhiza and phosphorus fertilizer under irrigation deficit condition effected on biological yield, grain yield, harvest index and plant height in basil, and we obtained the highest of values of traits by application of mycorrhiza and phosphorus (70 kg/ha) under normal irrigation.

Conclusion

In general it appears that application of mycorrhiza with phosphorus fertilizer improved yield and other plant criteria under water deficit. Therefore, it appears that application of mycorrhiza could be promising in the production of basil by the reduction of chemical fertilizer application; it seems using this mycorrhiza in agroecosystems could increase water uptake by its positive effects on root parameters and of course help farmers to save water in arid and semi-arid regions.

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