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## Aims and Scope

Plant Ecophysiology Journal publishes original scientific and technical research articles on ecological, physiological and ecophysiological aspects of plants growth and development. Studied plants can include crops, herbs, horticultural plants, etc. with an emphasis on all aspects of environmental factors and their effects on plants.



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# Effects of arbuscular mycorrhizal fungi on yield and yield components of maize under water stress

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**Abstract**

Maize (*Zea mays* L.) is an effective host of mycorrhiza in infertile and drought conditions. In order to study the effects of arbuscularmycorrhizal fungi (AMF) on yield and yield components of maize under well-watered ($I_1$), moderate drought stress ($I_2$) and severe drought stress ($I_3$) conditions, two field experiments were conducted at Agricultural Research Station in Khorramabad,Iran in 2011 and 2012. Two experiments were carried out as split-plot factorial based on randomized complete block design with three replications. Irrigation was imposed at three levels based on 70% ($I_1$), 50% ($I_2$) and 30% field capacity ($I_3$), as the main plot. Mycorrhizal biofertilizer (species *Glomusintraradices*) was applied at two levels; control or without mycorrhizal fungi application ($M_1$) and 100 kg $ha^{-1}$mycorrhizalbiofertilizer application ($M_2$), as the sub plot. Phosphorus fertilizer was applied at three levels; control or without application of phosphorus fertilizer ($P_1$), application of 75 kg $ha^{-1}$ triple superphosphate ($P_2$) and application of 150 kg $ha^{-1}$ triple superphosphate ($P_3$), as the sub plot. The results of combined variance analysis showed that, the year in the two-year experiments significantly affected row number per ear, grain yield and biological yield, but did not have significant effect on plant height, seed number per row and 400-seed weight. Different irrigation treatments significantly affected plant height, seed number per row, row number per ear, 400-seed weight, grain yield and biological yield. Different P fertilizer levels significantly affected plant height, 400-seed weight, grain yield and biological yield. Although different P fertilizer levels did not significantly affect seed number per row and row number per ear, maximum seed number per row was observed in $P_3$ application. Mycorrhizal biofertilizer significantly affected plant height, grain yield and biological yield, but did not havesignificant effect on seed number per row, row number per ear and 400-seed weight. In spite of no significant difference in mycorrhizal biofertilizer application, seed number per row and 400-seed weight have been increased by application of mycorrhizal biofertilizer.

*Keywords:* Mycorrhiza, Maize, Phosphorus, Water stress.

## Introduction

The term mycorrhiza literally means fungal-root, derived from the Greek *mykes* and *rhiza*, which mean fungus and root, respectively (Bardgett, 2005). It was first used by German researcher Albert Bernhard Frank in 1885. The symbiotic relationship between AM and the roots of higher plants contributes significantly to plant nutrition and growth (Auge, 2001), and has been shown to increase the productivity of a variety of field crops including maize (Sylvia *et al.,* 1993). These positive responses of productivity to AM colonization have mainly been attributed to the enhanced uptake by AM of relatively immobile soil ions such as phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg), sulfur (S), iron (Fe), zinc (Zn), copper (Cu) and manganese (Mn) (Marschner and Dell, 1994; Marschner, 1995; Liu *et al.,* 2000a,b; Liu *et al.,* 2007), but also in-

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volve the enhanced uptake and transport of far more mobile nitrogen (N) ions, particularly under drought conditions (Tobar *et al.,* 1994; Azcon *et al.,* 1996; Liu *et al.,* 2007). In maize and other species, the most widely recognized contribution of AM fungi to host-plant nutrition involves their ability to extract P from outside the P depletion zone around plant roots (Marschner, 1995; Miller, 2000; Liu *et al.,* 2003; Smith *et al.,* 2003; Liu *et al.,* 2007).

Drought is considered a natural disaster that originates from a deficiency of precipitation over an extended period of time being harmful to the different activities of the population. The damages caused by effect of droughts are much stronger in areas of extreme poverty. Drought causes negative impacts such as: reduced water availability for irrigation purposes; delay in the sowing dates and reduced crop yield; loss of productivity in natural prairies and dry-land crops; increased erosion on plains and high areas; environmental stress in hydromorphic areas; soils salinization due to the reduction in the volume of water for irrigation and drying up of wet areas; effect of frosts due to the delay of the sowing in dry lands; intensification of freatic water table and drying up of wells; and expansion of desertification in the arid, semi-arid and sub-humid ecosystems (Sivakumar and Motha, 2007).

Ortas (2010) showed that under field conditions, mycorrhiza inoculation significantly increased cucumber seedling survival, yield, P and Zn shoot concentrations. Also, it has indicated that indigenous mycorrhiza inoculum was also successful in colonized cucumber roots and resulted in better plant growth and yield (Ortas, 2010). Liu *et al.,* (2000a,b) conducted an experiment on maize (*Zea mays* L.) and concluded that mycorrhizal plants had significantly higher shoot dry weights than non-mycorrhizal plants (Liu *et al.,* 2000a,b). Sharif and Claassen (2011) concluded that the application of P increased shoot dry matter yield of *Capsicum annuum* L. The treatment of AM inoculation increased the shoot yield and shoot P content. They reported that at low P supply an infection with AM fungi significantly increased the yield of *Capsicum annuum* L. and it was related to an increased P uptake (Sharif and Claassen, 2011). Cozzolino *et al.*, (2010) reported that the leaf and root dry weight significantly increased with AMF inoculation and P application. When both factors were combined, the yield was 57.6% higher compared to non-inoculated plants. No significant differences were observed between inoculated and non-inoculated plants in biomass production, when P was not added (Cozzolino *et al.,* 2010). Kohler *et al.,* (2009) concluded that the shoot fresh biomass of inoculated plants was about 34% higher than that of non-inoculated plants. There was a significant interaction between microbial inoculation and water-stress for shoot fresh biomass; it was enhanced by *Pseudomonas mendocina* and *Glomusintraradices* in non-stressed plants but was not affected by the microbial inoculations in stressed plants. Water deficit caused a significant decrease in the shoot fresh and dry biomass and shoot water content of all plants. However, the microbial inoculation factor had no significant effect on the shoot water content. In non-stressed plants, inoculation with the AM fungus led to its active colonization of the root system of the lettuce (*Lactuca sativa* L.) seedlings. The percentage of roots colonized by AM fungi was not affected by bacterial inoculation, but decreased significantly with water stress (Kohler *et al.,* 2009). Also, they reported that the root biomass of plants grown under well-watered conditions was significantly higher than that of the stressed plants, particularly the *G. intraradices* - inoculated plants. Shoot dry biomass and mycorrhizal colonization were decreased significantly under water-stress conditions (Kohler *et al.,* 2009). Ruiz-Sanchez *et al.,* (2010) reported that AM colonization increased rice shoot biomass by 50%, and this effect was also attributed to enhancement of rice photosynthetic efficiency (Ruiz-Sanchez *et al.,* 2010). Ruiz-Sanchez *et al.,* (2011) reported that AM and non-AM plants were remarkably different in plant size (Ruiz-Sanchez *et al.,* 2011). Erman *et al.,* (2011) in an experiment on chickpea observed that AMF inoculation resulted in increased plant growth and nutritional parameters (Erman *et al.,* 2011). Efeoglu *et al.,* (2009) conducted an experiment on maize under drought conditions and observed that maize cultivars exposed to drought had a lower fresh and dry biomass than their controls due to a significant drought-induced reduction in growth. Fresh biomass of cultivars was significantly reduced under drought stress conditions. In addition, dry biomass was significantly decreased under drought stress (Efeoglu *et al.,* 2009). Celebi *et al.* (2010) conducted an experiment on maize and reported that the effect of different irrigation levels and AMF applications on the plant height was found to be significant in two years. While the highest plant height was measured in different irrigation levels, higher plant heights were measured in mycorrhizal plots compared to non-mycorrhizal plots. Moreover, it was realized that the difference of plant height between mycorrhizal and non-mycorrhizal plots was increased in different levels where the water was limited. They also reported that the effect of different irrigation levels and AMF applications on fresh and dry matter yield was found to be significant in two years (Celebi *et al.,* 2010). Zhang *et al.,* (2011) reported that maize plants inoculated with *G.mosseae* grew significantly higher than non-inoculated plants. However, at the high organic fertilization rate, there were not significant differences between +AM and -AM treatments. They also reported that the values of treatments with AM were significantly increased by the increasing of organic fertilizer but decreased at the highest fertilization rate compared to those without AM. The root-shoot dry weight ratio was significantly lower in +AM than in -AM treatments at low fertilization rates but there were no significant difference between +AM and -AM treatments at the high fertilization rates (Zhang *et al.,* 2011).

## Materials and methods

### *Site of experiment*

Two experiments were conducted at the Agricultural Research Station in Khorramabad, Iran in 2011 and 2012 (June $7^{th}$), with Lat. 33˚, 29′ N; Long. 48˚, 21′ E; Alt. 1171 m above sea level; mean temperature during the growth season in the first and second year were 24.90°c and 25.92°c, respectively.

### *Experimental design and agronomic applications*

Two experiments were carried out as split-plot factorial based on randomized complete block design with three replications. Irrigation was imposed at three levels; (a) well-watered conditions ($I_1$), based on 70% field capacity; (b) moderate drought stress conditions ($I_2$), based on 50% field capacity; (c) severe drought stress conditions ($I_3$), based on 30% field capacity, as the main plot. Mycorrhizal biofertilizer (species *Glomus intraradices*) was applied at two levels; (a) control or without application of mycorrhizal biofertilizer ($M_1$); (b) application of mycorrhizal biofertilizer ($M_2$) 100 kg ha $^{-1}$, as the sub plot. Phosphorus fertilizer was applied at three levels; (a) control ($P_1$); without application of phosphorus fertilizer; (b) application of 75 kg ha$^{-1}$ triple superphosphate ($P_2$); (c) application of 150 kg ha$^{-1}$ triple superphosphate ($P_3$), as the sub plot (values were used according to the soil testing).

The experimental field was ploughed in fall and disked twice in spring. Each plot was 8 m in length and consisted of 4 rows separated by 0.75 m, with a 0.20 m on-row spacing (Winterhalter *et al.,* 2011). The studied hybrid was NS-640. According to the soil testing, nitrogen and potassium fertilizers were determined, including 250 kg ha $^{-1}$ urea and 100 kg ha $^{-1}$ potassium sulfate. One third of nitrogen (N), all of mycorrhizal biofertilizer, phosphorous (P) and potassium (K) fertilizers were applied at planting and the remaining N was applied during the vegetative growth (Sajedi *et al.,* 2010). Farm operations for two years were the same.

### *Soil water content measurement*

Soil water content was measured by weighing the soil before and after drying at 105˚C for 24 h. Moisture weight percentage was calculated by using the following equation proposed by Kirkham, (2005).

$$\theta m = \frac{W_1 - W_2}{W_2} \times 100$$

where $\Theta_m$, $W_1$ and $W_2$ are water content (moisture content) percentage, soil wet weight and soil dry weight, respectively. Samples were collected from the 0 – 30 and 30 – 60 cm depths. The soil texture was clay loam. Bulk density was 1.35 g cm$^{-3}$. Moisture weight percentage in field capacity was 26.5 and 24.2 in 2011 and 2012, respec-

Table 1. Chemical characteristics of the soil in the experimental site

| Year | Depth | EC × $10^3$ | pH | T. N. V | O. C | P (av.) ppm | K (av.) ppm |
|---|---|---|---|---|---|---|---|
| First | 0-30 | 0.55 | 7.48 | 32.2 | 1.13 | 3.5 | 455 |
| | 30-60 | 0.67 | 7.70 | 35.0 | 0.95 | 2.2 | 340 |
| Second | 0-30 | 0.50 | 7.40 | 33.6 | 1.20 | 3.2 | 500 |
| | 30-60 | 0.62 | 7.40 | 35.2 | 0.85 | 2.5 | 370 |

Table 2. Mean square values in the combined analysis of variance of PH, SNR, RNE, 400SW, GY and BY

| S. O. V | df | PH | SNR | RNE | 400SW | GY | BY |
|---|---|---|---|---|---|---|---|
| Year (Y) | 1 | 303.108$^{ns}$ | 8.490$^{ns}$ | 20.367$^{*}$ | 105.475$^{ns}$ | 33376648.177$^{**}$ | 49178808.773$^{*}$ |
| R(Y) | 4 | 3145.535 | 156.459 | 6.539 | 2013.443 | 12202869.367 | 80164393.451 |
| Irrigation (I) | 2 | 27108.670$^{**}$ | 2965.014$^{**}$ | 77.496$^{**}$ | 4776.267$^{**}$ | 179635573.389$^{**}$ | 928315541.802$^{**}$ |
| Y × I | 2 | 169.143$^{ns}$ | 84.673$^{ns}$ | 13.066$^{ns}$ | 177.568$^{ns}$ | 379786.146$^{ns}$ | 5908735.852$^{ns}$ |
| Error (a) | 8 | 1030.039 | 45.376 | 3.951 | 202.509 | 1491887.544 | 18074206.651 |
| Triple superphosphate (P) | 2 | 1842.435$^{**}$ | 27.214$^{ns}$ | 0.138$^{ns}$ | 335.964$^{**}$ | 2021292.147$^{**}$ | 38292904.866$^{**}$ |
| Y × P | 2 | 138.326$^{ns}$ | 10.100$^{ns}$ | 1.184$^{ns}$ | 14.348 | 83295.217$^{ns}$ | 8422681.826$^{ns}$ |
| I × P | 4 | 116.378$^{ns}$ | 10.131$^{ns}$ | 0.328$^{ns}$ | 28.039$^{ns}$ | 281279.310$^{ns}$ | 6400944.162$^{ns}$ |
| Y × I × P | 4 | 30.564$^{ns}$ | 5.856$^{ns}$ | 0.549$^{ns}$ | 14.591$^{ns}$ | 276532.888$^{ns}$ | 2139847.839$^{ns}$ |
| Mycorrhiza (M) | 1 | 944.300$^{**}$ | 10.342$^{ns}$ | 0.239$^{ns}$ | 143.959$^{ns}$ | 2718369.245$^{**}$ | 72700386.663$^{**}$ |
| Y × M | 1 | 0.725$^{ns}$ | 8.245$^{ns}$ | 0.472$^{ns}$ | 56.492$^{ns}$ | 79576.235$^{ns}$ | 9242420.862$^{ns}$ |
| I × M | 2 | 48.383$^{ns}$ | 2.562$^{ns}$ | 0.494$^{ns}$ | 18.153$^{ns}$ | 190712.212$^{ns}$ | 13451760.116$^{ns}$ |
| Y × I × M | 2 | 75.220$^{ns}$ | 1.339$^{ns}$ | 0.687$^{ns}$ | 18.402$^{ns}$ | 171239.381$^{ns}$ | 8392598.264$^{ns}$ |
| P × M | 2 | 179.981$^{ns}$ | 14.679$^{ns}$ | 0.847$^{ns}$ | 9.389$^{ns}$ | 21828.525$^{ns}$ | 151669.764$^{ns}$ |
| Y × P × M | 2 | 127.237$^{ns}$ | 20.174$^{ns}$ | 0.090$^{ns}$ | 10.723$^{ns}$ | 121920.970$^{ns}$ | 189360.451$^{ns}$ |
| I × P × M | 4 | 33.558$^{ns}$ | 21.442$^{ns}$ | 0.527$^{ns}$ | 42.685$^{ns}$ | 83433.233$^{ns}$ | 1738845.546$^{ns}$ |
| Y × I × P × M | 4 | 27.437$^{ns}$ | 19.252$^{ns}$ | 0.854$^{ns}$ | 28.422$^{ns}$ | 91854.062$^{ns}$ | 1000346.322$^{ns}$ |
| Error (b) | 60 | 72.141 | 14.647 | 0.737 | 38.080 | 242366.509 | 4658592.114 |
| C. V % | - | 3.87 | 11.75 | 6.01 | 5.43 | 6.75 | 11.07 |

**: Significant at $P \leq 0.01$, *: Significant at $P \leq 0.05$ and ns: Non- significant; plant height (PH), seed number per row (SNR), row number per ear (RNE), 400 seeds weight (400SW), grain yield (GY) and biological yield.

tively. The soil pH was 7.5.

Irrigation time was determined by weighting soil samples (taken by Auger from the root extension depth) to obtain moisture weight percentage. Then by using the following equation proposed by Doorenbos and Pruitt (1975), irrigation water volume was calculated.

$$V = \frac{(FC - \beta m) \times \rho b \times Dr \times A}{100}$$

where $V$ is the irrigation water volume (m$^3$), $FC$ is the gravimetric soil water content at field capacity (%), $\beta m$ is the soil water content before irrigation by weight (%), $\rho b$ is the bulk density of the soil (g cm$^{-3}$), $Dr$ is the root extension depth (m) and $A$ is the irrigated area (m$^2$).

*Grain yield measurement*

Final harvest was accomplished on October 10$^{th}$ at the physiological maturity stage when black layer formation at the bottom of seed (Daynard and Duncan, 1969). Grain yield (GY) was measured by gleaning ear from each treatment and replication at the mid-canopy position (10 ears) with calculating 14% moisture (Naghashzadeh *et al.,* 2009).

*Dry weight measurement*

The leaves and stems were cut from 10 plants selected. Dry weight (DW) was determined by weighing the segments after 48 h at 70$^{\circ}$C in oven.

*Biological yield measurement*

Biological yield was calculated by using the following equation:

$$BY = GY + DW$$

Where $BY$, $GY$ and $DW$ are biological yield, grain yield and dry weight respectively (Uddin *et al.*, 2010)

*Statistical analysis*

The recorded data were statistically analyzed (ANOVA analysis) using the software MSTATC and SAS. Means comparisons were calculated using Duncan's Multiple Range Test at $P \leq 0.05$ .

## Results

*Plant height*

The results of combined variance analysis showed that different irrigation treatments, different P fertilizer levels and mycorrhizal biofertilizer application significantly affected plant height (Table 2). The results of mean comparisons showed that plant height was decreased by increasing drought stress (Table 3). Also, plant height was increased by increasing P application (Table 3). It was observed that plant height has increased in AM plants (Table 3).

Table 3. Means comparison of PH, SNR, RNE, 400SW, GY and BY

| Factor | PH (cm) | SNR | RNE | 400SW (g) | GY (kg $ha^{-1}$) | BY (kg $ha^{-1}$) |
|---|---|---|---|---|---|---|
| *Year* | | | | | | |
| $Y_1$ | 221.168 a | 32.296 a | 14.714 a | 114.593 a | 7852.205 a | 20165.141 a |
| $Y_2$ | 217.818 a | 32.856 a | 13.845 b | 112.616 a | 6740.372 b | 18815.534 b |
| *Irrigation* | | | | | | |
| $I_1$ | 246.8 a | 40.34 a | 15.74 a | 125.7 a | 9262 a | 24480 a |
| $I_2$ | 219.8 b | 34.80 b | 14.30 b | 112.4 b | 7759 b | 19660 b |
| $I_3$ | 191.9 c | 22.60 c | 12.80 c | 102.7 c | 4867 c | 14330 c |
| *Triple superphosphate* | | | | | | |
| $P_1$ | 211.5 b | 31.91 a | 14.24 a | 111.1 b | 7063 b | 18630 b |
| $P_2$ | 221.6 a | 32.26 a | 14.25 a | 112.7 b | 7289 b | 19210 b |
| $P_3$ | 225.3 a | 33.56 a | 14.35 a | 117.0 a | 7537 a | 20630 a |
| *Mycorrhiza* | | | | | | |
| −M | 216.536 b | 32.267 a | 14.232 a | 112.450 a | 7137.638 b | 18669.879 b |
| +M | 222.450 a | 32.886 a | 14.326 a | 114.759 a | 7454.939 a | 20310.796 a |

The same letters within each column indicate no significant difference among treatments ($P \leq 0.05$); plant height (PH), seed number perrow (SNR), row number per ear (RNE), 400-seed weight (400 SW), grain yield (GY) and biological yield (BY).

*Seed number per row (SNR)*

The results of combined variance analysis showed that different irrigation treatments significantly affected SNR (Table 2). The results of mean comparisons showed that SNR was decreased by increasing drought stress (Table 3). Different P fertilizer levels and mycorrhizal biofertilizer application did not have significant effect on SNR (Table 2). In spite of no significant difference in different P fertilizer levels and mycorrhizal biofertilizer application, SNR was increased by increasing P and mycorrhizal biofertilizer application (Table 3).

*Row number per ear (RNE)*

The results of combined variance analysis showed that year and different irrigation treatments significantly affected RNE (Table 2). Row number per ear of the first year was more than that of the second year (Table 3). Row number per ear was decreased by increasing drought stress (Table 3). Different P fertilizer levels and mycorrhizal biofertilizer application did not have significant effect on RNE (Table 2). Although different P fertilizer levels and mycorrhizal biofertilizer application did not have significant effect on RNE, maximum row number per ear value was observed in $P_3$ (150 kg $ha^{-1}$ triple superphosphate) and AM plants (Table 3).

*Four hundred-seed weight (400SW)*

The results of combined variance analysis showed that different irrigation treatments significantly affected 400SW at $P \leq 0.01$ (Table 2). Four hundred-seed weight was decreased by increasing drought stress (Table 3). Different P fertilizer levels significantly affected 400SW at $P \leq 0.05$ (Table 2). The results showed that 400SW was increased by increasing P application (Table 3). Mycorrhizal biofertilizer did not significantly affect 400SW (Table 2). In spite of no significant difference in mycorrhizal biofertilizer application, 400SW was increased by application of mycorrhizal biofertilizer (Table 3).

*Grain yield (GY)*

The results of combined variance analysis showed that year, different irrigation treatments, different P fertilizer levels and mycorrhizal biofertilizer significantly affected GY at $P \leq 0.01$ (Table 2). Grain yield of the first year was more than that of the second year (Table 3). Grain yield was decreased by increasing drought stress (Table 3). The results of mean comparisons showed that GY was increased by increasing P application (Table 3). In spite of significant difference in different P fertilizer levels, there were no significant difference in GY between $P_1$ and $P_2$ (Table 3). However, maximum GY value was observed in $P_3$ application (Table 3). In this study, grain yield of AM plants was more than that of non-AM plants (Table 3).

*Biological yield (BY)*

The results of combined variance analysis

Table 4. Means comparison of PH, SNR, RNE, 400SW, GY and BY as affected by two-way interaction effects

| Factor | | PH (cm) | SNR | RNE | 400SW (g) | GY (kg ha$^{-1}$) | BY (kg ha$^{-1}$) |
|---|---|---|---|---|---|---|---|
| *Irrigation* | *P* | | | | | | |
| $I_1$ | $P_1$ | 236.3 c | 39.20 a | 15.60 a | 122.6 b | 8970 b | 22940 b |
| | $P_2$ | 248.3 b | 39.91 a | 15.85 a | 124.8 ab | 9371 ab | 24050 b |
| | $P_3$ | 255.8 a | 41.90 a | 15.76 a | 129.6 a | 9446 a | 26460 a |
| $I_2$ | $P_1$ | 215.0 e | 34.53 b | 14.10 b | 110.0 d | 7504 d | 18790 c |
| | $P_2$ | 220.5 de | 34.13 b | 14.27 b | 110.1 d | 7602 d | 19590 c |
| | $P_3$ | 223.8 d | 35.73 b | 14.53 b | 117.1 c | 8172 c | 20590 c |
| $I_3$ | $P_1$ | 183.3 g | 21.29 c | 12.76 c | 100.7 e | 4716 e | 13990 d |
| | $P_2$ | 196.1 f | 23.44 c | 12.81 c | 103.2 e | 4894 e | 14150 d |
| | $P_3$ | 196.4 f | 23.06 c | 12.84 c | 104.3 e | 4992 e | 14850 d |
| *Irrigation* | *Mycorrhiza* | | | | | | |
| $I_1$ | −M | 243.1 b | 39.84 a | 15.68 a | 124.0 a | 9030 b | 22960 b |
| | +M | 250.5 a | 40.83 a | 15.79 a | 127.4 a | 9494 a | 26000 a |
| $I_2$ | −M | 218.1 c | 34.79 b | 14.27 b | 112.0 b | 7673 c | 19110 c |
| | +M | 221.4 c | 34.80 b | 14.33 b | 112.7 b | 7846 c | 20200 c |
| $I_3$ | −M | 188.3 e | 22.17 c | 12.63 c | 101.3 c | 4710 d | 13940 d |
| | +M | 195.5 d | 23.02 c | 12.98 c | 104.2 c | 5025 d | 14720 d |
| *P* | *Mycorrhiza* | | | | | | |
| $P_1$ | −M | 206.0 d | 30.87 a | 14.11 a | 109.5 c | 6898 d | 17880 d |
| | +M | 217.1 c | 32.95 a | 14.37 a | 112.6 bc | 7229 bcd | 19380 bcd |
| $P_2$ | −M | 219.9 bc | 32.11 a | 14.12 a | 112.1 bc | 7110 cd | 18380 cd |
| | +M | 223.4 ab | 32.41 a | 14.38 a | 113.3 bc | 7468 ab | 20040 b |
| $P_3$ | −M | 223.8 ab | 33.52 a | 14.21 a | 115.7 ab | 7406 abc | 19750 bc |
| | +M | 226.9 a | 33.60 a | 14.49 a | 118.3 a | 7668 a | 21510 a |

The same letters within each column indicate no significant difference among treatments ($P \leq 0.05$); plant height (PH), seed number per row (SNR), row number per ear (RNE), 400-seed weight (400 SW), grain yield (GY) and biological yield (BY).

showed that year has significantly affected BY at $P \leq 0.05$ (Table 2). Different irrigation treatments, different P fertilizer levels and mycorrhizal biofertilizer significantly affected BY at $P \leq 0.01$ (Table 2). Biological yield of the first year was more than that of the second year (Table 3). Biological yield was decreased by increasing drought stress (Table 3). Biological yield was increased by increasing P application (Table 3). Although different P fertilizer levels significantly affected biological yield, there was no significant difference in BY between $P_1$ and $P_2$ (Table 3). However, maximum BY value was observed in $P_3$ application (Table 3). Biological yield of AM plants was more than that of non-AM plants (Table 3).

## Discussion

The AM symbiosis generally increases host plant growth due to improved plant nutrition (Smith and Read, 1997). Kohler *et al.,* (2009) concluded that the shoot fresh biomass of inoculated plants was about 34% higher than that of non-inoculated control plants. There was a significant interaction between microbial inoculation and water-stress for shoot fresh biomass; it was enhanced by *Pseudomonas mendocina* and *Glomus intraradices* in non-stressed plants but was not affected by the microbial inoculations in stressed plants. Water deficit caused a significant decrease in the shoot fresh and dry biomass and shoot water content of all plants. However, the microbial inoculation factor had no significant effect on the shoot water content. In non-stressed plants, inoculation with the AM fungus led to its active colonization of the root system of the lettuce (*Lactuca sativa* L.) seedlings. The percentage of roots colonized by AM fungi was not affected by bacterial inoculation, but decreased significantly with water stress (Kohler *et al.,* 2009). Also, they reported that the root biomass of plants grown under well-watered conditions was significantly higher than that of the stressed plants, particularly the *G. intraradices* - inoculated plants. Shoot dry biomass and mycorrhizal colonization were decreased significantly under water-stress conditions (Kohler *et al.,* 2009). Efeoglu *et al.,* (2009) conducted an experiment on maize under drought conditions and observed that maize cultivars exposed to drought had a lower fresh and dry biomass than their controls due to a significant drought-induced reduction in growth. Fresh biomass of cultivars was signifi-

Table 5. Means comparison of PH, SNR, RNE, 400SW, GY and BY as affected by three-way interaction effects

| Factor | | | PH (cm) | SNR | RNE | 400SW (g) | GY (kg ha$^{-1}$) | BY (kg ha$^{-1}$) |
|---|---|---|---|---|---|---|---|---|
| *Irrigation* | *P* | *Mycorrhiza* | | | | | | |
| $I_1$ | $P_1$ | −M | 230.6 d | 39.82 abc | 15.45 abc | 121.1 bcd | 8708 cd | 21520 cd |
| | $P_2$ | −M | 242.0 c | 40.00 ab | 15.75 a | 124.2 abc | 9232 abc | 24360 b |
| | $P_3$ | −M | 245.1 bc | 40.11 ab | 16.04 a | 121.5 bcd | 9049 bc | 22870 bc |
| | $P_1$ | +M | 251.4 abc | 38.29 abcd | 15.65 ab | 128.1 ab | 9334 ab | 24500 b |
| | $P_2$ | +M | 253.7 ab | 41.39 ab | 15.59 ab | 129.4 a | 9558 ab | 25230 b |
| | $P_3$ | +M | 257.9 a | 42.40 a | 15.93 a | 129.8 a | 9693 a | 28420 a |
| $I_2$ | $P_1$ | −M | 212.4 f | 32.24 e | 13.91 de | 109.0 fghi | 7385 g | 18340 e |
| | $P_2$ | −M | 217.6 ef | 33.27 e | 14.63 bcd | 111.0 fgh | 7623 fg | 19030 de |
| | $P_3$ | −M | 219.7 def | 36.81 bcde | 14.27 d | 112.2 efg | 7575 fg | 19240 de |
| | $P_1$ | +M | 221.3 def | 34.31 de | 13.92 de | 108.1 fghi | 7630 fg | 19950 de |
| | $P_2$ | +M | 222.4 def | 34.99 cde | 14.43 cd | 114.9 def | 8059 ef | 20150 de |
| | $P_3$ | +M | 225.2 de | 37.14 bcde | 14.64 bcd | 119.2 cde | 8285 de | 21220 cd |
| $I_3$ | $P_1$ | −M | 175.0 h | 20.55 f | 12.65 f | 98.65 j | 4600 h | 13230 f |
| | $P_2$ | −M | 191.5 g | 22.03 f | 13.03 ef | 102.8 ij | 4831 h | 13770 f |
| | $P_3$ | −M | 194.8 g | 22.96 f | 12.82 ef | 102.7 ij | 4706 h | 14540 f |
| | $P_1$ | +M | 197.3 g | 22.03 f | 12.79 f | 102.6 ij | 4824 h | 14750 f |
| | $P_2$ | +M | 195.2 g | 24.08 f | 12.41 f | 103.7 hij | 5082 h | 14820 f |
| | $P_3$ | +M | 197.7 g | 23.93 f | 13.11 ef | 106.1 ghij | 5161 h | 14890 f |

The same letters within each column indicate no significant difference among treatments ($P \leq 0.05$); plant height (PH), seed number per row (SNR), number per ear (RNE), 400-seed weight (400 SW), grain yield (GY) and biological yield (BY).

cantly reduced under drought stress conditions. In addition, dry biomass was significantly decreased under drought stress (Efeoglu *et al.,* 2009). Ruiz-Sanchez *et al.,* (2011) reported that AM and non-AM plants were remarkably different in plant size (Ruiz-Sanchez *et al.,* 2011). Celebi *et al.,* (2010) reported that the effect of different irrigation levels and AMF applications on the plant height of maize was found to be significant in two years. While the highest plant height was measured in different irrigation levels, higher plant heights were measured in mycorrhizal plots compared to non-mycorrhizal plots. Moreover, it was realized that the difference of plant height between mycorrhizal and non-mycorrhizal plots was increased in different levels where the water was limited. They also reported that the effect of different irrigation levels and AMF applications on fresh and dry matter yield was found to be significant in two years (Celebi *et al.,* 2010).

Cozzolino *et al.,* (2010) reported that the leaf and root dry weight significantly increased with AMF inoculation and P application. When both factors were combined, the yield was 57.6% higher compared to non-inoculated plants. No significant differences were observed between inoculated and non-inoculated plants in biomass production, when P was not added (Cozzolino *et al.,* 2010). Sharif and Claassen (2011) concluded that the application of P increased shoot dry matter yield of *Capsicum annuum* L. The treatment of AM inoculation increased the shoot yield and shoot P content. They reported that at low P supply, an infection with AM fungi significantly increased the yield of *Capsicum annuum* L. and it was related to an increased P uptake (Sharif and Claassen, 2011).

Liu *et al.,* (2000a,b) in an experiment on maize (*Zea mays* L.) concluded that mycorrhizal plants had significantly higher shoot dry weights than non-mycorrhizal plants (Liu *et al.,* 2000a,b). Ruiz-Sanchez *et al.,* (2010) reported that AM colonization increased rice shoot biomass by 50%, and this effect was also attributed to enhancement of rice photosynthetic efficiency (Ruiz-Sanchez *et al.,* 2010). Erman *et al.,* (2011) conducted an experiment on chickpea and observed that AMF inoculation resulted in increases in plant growth and nutritional parameters (Erman *et al.,* 2011). Zhang *et al.,* (2011) reported that maize plants inoculated with *G.mosseae* grew significantly higher than non-inoculated control plants. However, at the high organic fertilization rate, there were not significant differences between +AM and -AM treatments. They also reported that the values of treatments with AM significantly increased by the increasing of organic fertilizer but decreased at the highest fertilization rate compared to those without AM. The root-shoot dry weight ratio was significantly lower in +AM treatments than in -AM treatments at low fertilization rates but there were no significant difference between +AM and -AM treatments at the high fertilization rates (Zhang *et al.,* 2011). Sajedi *et al.,* (2010) conducted an experiment on maize under drought conditions and reported that AM fungi significantly increased corn grain yield at the well-watered than drought stress conditions. Although different irrigation treatments significantly affected seed number per row and row number per ear, AM fungi did not have significant effect on seed number per row and row number per ear (Sajedi *et al.,* 2010), which is somehow in accordance with the results of this experiment.

## Conclusion

We observed that plant height (PH), seed number per row (SNR), row number per ear (RNE), 400-seed weight (400SW), grain yield (GY) and biological yield (BY) in maize plants were affected greatly by water stress conditions. The data showed that the mycorrhizal biofertilizer application increased PH, GY and BY in maize plants as a consequence of enhancing nutrient uptake and water status of the plants. Generally, AM plants had a greater effect than non-AM plants. Different P fertilizer levels significantly affected PH, 400SW, GY and BY, but did not have significant effect on SNR and RNE. Two-year experiment significantly affected RNE, GY and BY. This indicated that maize plants encountered different environmental conditions, so that measured traits showed different reactions in the first and second years. We concluded that the difference between weather in the first and second experimental years led to different reaction in the first than the second year. With respect to environmental problems associated with fertilizer and water limitation in future, it is essential that we apply water resources appropriately and decrease fertilizers application in order to improve soil fertility, productivity and water quality.

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