

## Effects of Water Deficit Stress and Two Mycorrhiza Species on Some Physiological Attributes and Tuber Fresh Weight of Potato cv. Agria Under Field Condition

Saman Khosravifar<sup>1</sup>, Farhad Farahvash<sup>1\*</sup>, Nasser Aliasgharzad<sup>2</sup>, Mehrdad Yarnia<sup>1</sup> and Farrokh Rahimzadeh Khoie<sup>1</sup>

Received: August 17, 2015 Accepted: September 18, 2015

<sup>1</sup>Department of Agronomy and Plant Breeding, Tabriz Branch, Islamic Azad University, Tabriz, Iran

<sup>2</sup>Department of Soil Science, Faculty of Agriculture, University of Tabriz, Tabriz, Iran

\*Corresponding author; Email: [farahvash@iaut.ac.ir](mailto:farahvash@iaut.ac.ir)

### Abstract

In order to evaluate the tuber fresh weight and some physiological attributes of Agria potato cultivar in response to water deficit stress and mycorrhiza inoculation, a field trial was conducted as a split plot scheme based on randomized complete block design with three replications at the research site of Islamic Azad University, Tabriz Branch in 2012. Four water levels including: irrigation after 60 (S<sub>0</sub>) mm evaporation from class A pan as the control and irrigation after 90 (S<sub>1</sub>), 120 (S<sub>2</sub>) and 150 (S<sub>3</sub>) mm evaporation from the pan as water deficit stress conditions were arranged in the main plots and four levels of arbuscular mycorrhizal fungi (AMF) including: no AMF (M<sub>0</sub>), inoculation of *Glomus etunicatum* (G<sub>e</sub>) (Becker and Gerdemann), *Glomus intraradices* (G<sub>i</sub>) (Schenck and Smith) and dual inoculation of the two species (G<sub>ei</sub>) were arranged in the sub-plots. Water application and AMF inoculation had significant effect on all of studied attributes. Increasing of water deficit stress intensity increased stomatal resistance and decreased leaf relative water content and tuber fresh weight. On other hand, positive effect of AMF inoculation was observed on stomatal resistance, leaf relative water content, leaf chlorophyll content and tuber fresh weight. Also, significant interaction between water application and AMF inoculation was observed for stomatal resistance, leaf relative water content, leaf chlorophyll content and tuber fresh weight. In general, water deficit stress decreased tuber fresh weight in non-mycorrhizal and mycorrhizal potatoes. In all of water levels, maximum tuber fresh weight was observed by inoculation with *G. intraradices*.

**Keywords:** Arbuscular mycorrhiza; Potato; Stomatal resistance; Tuber fresh weight; Water deficit

### Introduction

Drought stress has an adverse effect on crop plants (Xoconostle-Cazares *et al.* 2010) and limits productivity of agricultural systems and food production (Boyer 1982). Half of the human death by natural disasters is caused by drought (Hoerling and Kumar 2003).

Arbuscular mycorrhiza (AM) is a widespread mutualistic symbiosis (non-pathogenic association) between soil-borne fungi and plant roots (Marschner and Dell 1994; Quilambo 2003) in majority of crop and horticultural plants (Smith and Read 2008). Studying the arbuscular

mycorrhizal fungi (AMF) is difficult because of their multiple ecological functions in the soil system (Hamel and Strullu 2006). Mycorrhiza with important role in plant production can improve nutrient uptake in crops and increase their yield. Also, it plays a key role in sustainable agriculture (Moawad 1979; Sharif and Moawad 2006). AMF can protect host plant (Ellis *et al.* 1985; Huang *et al.* 1985; Puppi and Bras 1990; Sanchez-Diaz and Honrubia 1994; Ruiz-Lozano *et al.* 1999; Quilambo 2003), increase its drought tolerance by changing soil-plant water relations and enhancement of water absorption capacity

(Allen 1982; Fitter 1985; Hardie 1985; Koide 1985; Kothari *et al.* 1990; Faber *et al.* 1991; Sylvia *et al.* 1993; Vonreichenbach and Schonbeck 1995; Dell'Amico *et al.* 2002; Echave *et al.* 2005). Also, AM plants recover faster than non-mycorrhizal (NM) plants in the water stress conditions (Hardie and Leyton 1981; Al-Karaki and Clark 1998).

Potato (*Solanum* spp.) with extraordinary adaptation range, easy cultivation and high nutritional content has helped sustain humanity for centuries (DeFauw *et al.* 2012). Potato can't be matched with other crops in view of its production of food energy and food value per unit area (Guenthner 2010). As a staple food, potato is the fourth most important crop after rice, wheat and maize (Gaurav *et al.* 2010) and is very

drought sensitive because it consumes great amount of water during growth period (Lahlou *et al.* 2003).

The aim of this study was to evaluate the effect of water deficit stress on some physiological attributes and tuber fresh weight of potato in the presence of two mycorrhiza species under field condition.

### Materials and Methods

The field experiment was conducted at the Agricultural Research Station of Islamic Azad University, Tabriz Branch (38°5' N, 46°17' E, 1360 m) in 2012. Meteorological data of the station during the growing season are shown in Table 1.

**Table 1. Average monthly temperature and precipitation of the research site for 2012**

	May	June	July	August	September	October
Temperature	19.05	23.88	26.30	31.99	36.30	16.40
Percipitation	0.87	0.59	0.53	0.53	0.17	0.19

The experiment was conducted as the split plot scheme based on randomized complete block design with three replications. Four water levels, irrigation after 60 ( $S_0$ ) mm evaporation from class A pan (located in the experimental field) as the control and irrigation after 90 ( $S_1$ ), 120 ( $S_2$ ) and 150 ( $S_3$ ) mm evaporation from the pan as water deficit stress conditions were arranged in the main plots and four types of arbuscular mycorrhizal fungi (AMF) including no application of AMF ( $M_0$ ), inoculation with *Glomus etunicatum* ( $G_e$ ) (Becker and Gerdemann), *Glomus intraradices* ( $G_i$ ) (Schenck and Smith) and dual inoculation of the two species ( $G_{ei}$ ) were arranged in the sub-plots. In order to supply dual inoculum of the two

species, their inoculum was mixed with the 1:1 ratio (w/w). Sixty g of inocula were placed directly under potato tubers. Water levels were exerted from the flowering time.

Certified potato seeds (*Solanum tuberosum* cv. Agria, a mid to late-maturing cultivar) were supplied from the Center of Agricultural Services, Khodabande city, Zanzan Province, Iran. Average weight of seeds was 70 g. No fungicide or any other kind of pesticides were used before planting the seeds in the experimental site. The two AMF inoculation species, *G. etunicatum* and *G. intraradices* were provided from the Department of Soil Science, Faculty of Agriculture, University of Tabriz, Iran. The inoculums were the mixture

of 2/3 sterile sand, 1/3 sterile soil, spores (60 spores per ml of inoculum), hyphae and infected fragment of sorghum roots.

Plots were prepared after plowing and disk harrowing. Soil sampling at depth of 30 cm showed that its texture was sandy loam having some chemical attributes as follow: pH of 8.2, 550 mg kg<sup>-1</sup> available K, 28.4 mg kg<sup>-1</sup> available P and organic carbon of 0.98%. The field was kept in the fallow condition during the previous year in order to decrease endogenous mycorrhizal fungi, remove the fungi propagules and decompose the root debris of the previous crop. Potato seeds were planted on May 9 with between-row space of 75 cm and within-row space of 25 cm in plots of 4.5m×5.25m size, with 6 rows. Planting depth was 7 cm. Weeding was done by hand, without applying any kind of herbicide.

All physiological attributes were measured from the third leaf situated on the top of the canopy one day before the final irrigation. Stomatal resistance was measured on the basis of sec cm<sup>-1</sup> through a steady state with a poremeter model AP4 (Delta-T Devices, Ltd., Cambridge, UK). For the measurement of leaf relative water content sampled leave were weighted by a digital scale with 0.01 g accuracy to determine the fresh weight ( $W_f$ ). The leaves were subsequently saturated in the distilled water for 12 h, re-weighted to determine saturated weight ( $W_s$ ), and weighted again to determine dried weight ( $W_d$ ) after drying at 80°C. Following formula was used to determine leaf relative water content (Cvikrová *et al.* 2013).

$$\text{RWC (\%)} = (W_f - W_d) / (W_s - W_d) \times 100.$$

Leaf chlorophyll content was measured by the chlorophyll meter (CCM-200, Opti-Science, USA). The chlorophyll meter calculates a unit-less chlorophyll content index (CCI) on the basis of absorbance measurement at 655 and 940 nm. At harvesting time (October 7) tubers of three plants in the center rows of each plot were weighed and their mean was recorded.

Statistical analysis of the collected data was carried out using MSTATC software and the means were compared by the Least Significant Difference (LSD) test at the 5% and 1% probability levels.

## Results

### Stomatal resistance

Water level, mycorrhiza inoculation and their interaction had significant effects on stomatal resistance (Table 2). Water deficit stress increased stomatal resistance ( $r_s$ ), as there were significant differences between stomatal resistance of  $S_0$  and all water deficit stress levels. The maximum and the minimum values of  $r_s$  were observed for  $S_3$  and  $S_0$ , respectively.  $S_3$  was 524.84% more than  $S_0$  (Table 3). It is noticeable that  $r_s$  value did not change significantly after  $S_1$ . Table 4 shows that AMF species and their dual inoculation had different effects on stomatal resistance.  $G_{ei}$  had the maximum stomatal resistance which was 45.38% more than  $M_0$  as the minimum. Stomatal resistance of  $M_0$ ,  $G_e$  and  $G_i$  were not significantly different, but their  $r_s$  value were significantly different from  $G_{ei}$ . Table 5 shows that the highest and the lowest  $r_s$  values were observed in  $S_1G_{ei}$  and  $S_0G_e$ , respectively. In both NM and AM plants stomatal resistance of water deficit stress

levels were significantly more than that of  $S_0$  as the control. Stomatal resistance of  $S_3$  decreased in AM plants as compared to NM. This decrease was significant for  $G_i$  and  $G_{ei}$ . It is noticeable that stomatal behavior at  $S_3$  was nearly similar for  $M_0$  and  $G_e$ .

### Relative water content

Based on Table 2, water level, mycorrhiza inoculation and their interaction had significant effects on leaf RWC. The maximum and the minimum RWC was observed for  $S_0$  and  $S_1$ , respectively. At  $S_1$ , RWC decreased 14.32% as compared to  $S_0$ . There were significant differences between RWC of water deficit stress levels and  $S_0$  as the control, but RWC values of  $S_1$ ,  $S_2$  and  $S_3$  did not differ significantly (Table 3). In AM plants, RWC was more than NM, as the highest and the lowest RWC were recorded in  $G_{ei}$  and  $M_0$ , respectively. RWC of dual inoculated plants was 8.99% more than  $M_0$ . There was not

significant difference between RWC of  $G_e$  and  $M_0$ , but both  $G_i$  and  $G_{ei}$  were significantly different from  $M_0$ . Also, there was not significant difference between RWC values of  $G_i$  and  $G_{ei}$  (Table 4). Table 5 shows that among the combinations of water and AMF inoculation levels, the maximum and the minimum RWC were obtained from  $S_0M_0$  and  $S_3M_0$ , respectively. It is obvious that gradient of RWC decrease in  $M_0$  plants was more pronounced than AM as water stress intensity increased from  $S_0$  to  $S_1$ . RWC values of  $M_0$  plants did not significantly change as the water level changed from  $S_1$  to  $S_3$ . This condition was also observed for  $G_i$  and  $G_{ei}$ . In  $G_e$  plants the differences of  $S_0$  level with  $S_1$  and  $S_3$  were significant. However,  $S_1$  and  $S_3$  did not differ significantly. It is noticeable that the differences of RWC value of  $S_2$  plants with  $S_1$  and  $S_3$  were significant and non-significant, respectively.

**Table 2. Analysis variance of the effects of water stress level and mycorrhiza inoculation on the studied attributes of potato**

Source of variation	df	Mean squares			
		Stomatal resistance	Leaf relative water content	Chlorophyll content	Tuber fresh weight
Replication	2	7.30	5.94	4.36	3574.87
Water level (S)	3	3161.48**	248.68**	316.43**	210262.44**
Main plot error	6	16.46	4.52	23.83	1860.80
Mycorrhiza inoculation (M)	3	390.88**	61.01**	240.52**	82320.28**
S×M	9	350.18**	45.92**	160.68**	8542.24*
Sub-plot error	24	32.83	4.85	13.61	3425.38
CV (%)		18.76	3.66	6.95	16.16

\*:  $P \leq 0.05$ , \*\*:  $P \leq 0.01$ , CV: Coefficient of Variation, df: degrees of freedom

### Chlorophyll content

Water level, mycorrhiza inoculation and their interaction had significant effects on chlorophyll

content (Table 2). Table 3 shows that the differences of chlorophyll content of  $S_0$  with other water application levels were not significant, but a

significant difference was observed between  $S_1$  and  $S_2$ . On the basis of Table 4, maximum and minimum chlorophyll content were observed in  $G_e$  and  $M_0$  plants, respectively. In  $G_e$  plants it was 21.20% more than  $M_0$ . Chlorophyll content of

AM plants was significantly higher than NM plants. However, there were not significant differences among chlorophyll content of AM plants.

**Table 3. Means of potato traits under study at different water levels**

Water level	Stomatal resistance (sec cm <sup>-1</sup> )	Leaf relative water content (%)	Chlorophyll content (CCI)	Tuber fresh weight (g)
$S_0$	6.48	66.89	52.85	551.12
$S_1$	40.17	57.31	59.67	349.16
$S_2$	34.99	59.04	47.13	304.04
$S_3$	40.49	57.41	52.59	244.31
LSD (1%)	6.14	3.22	7.38	65.29

$S_0$  represents water application after 60 mm evaporation from class A pan as control.  $S_1$ ,  $S_2$  and  $S_3$  show water stress levels after 90, 120 and 150 mm evaporation from the pan, respectively. LSD: Least significant difference.

On the basis of Table 5, chlorophyll content of  $S_1G_e$ , having the maximum value, and that of  $S_2G_{ei}$ , having the minimum value, were 62.88% higher and 5.89% lower than  $S_0M_0$  condition, respectively. There were not significant differences among chlorophyll content of all water levels in  $M_0$  plants. In  $G_e$  and  $G_{ei}$  plants, the chlorophyll content was increased significantly as the water level changed from  $S_0$  to  $S_1$ . The chlorophyll content of  $S_0$  was not significantly different from those of  $S_2$  and  $S_3$  as inoculated with *G. etunicatum*. This status was also observed between  $S_0$  and  $S_2$  levels in the condition of dual inoculation of AMF. Chlorophyll content was decreased with the increase of water deficit from  $S_1$  to  $S_2$  for both  $G_e$  and  $G_{ei}$ . In  $G_i$  plants, chlorophyll content of all water deficit stress levels were less than  $S_0$  as the control. Moreover, the difference between  $S_0$  and  $S_2$  was significant in  $G_i$  plants.

### Tuber fresh weight

Effects of water levels, mycorrhiza inoculation and their interaction were significant on tuber fresh weight (Table 1). Tuber fresh weight was decreased with the increase in water stress intensity as the maximum and minimum values were observed at  $S_0$  and  $S_3$ , respectively. At  $S_3$  it was 55.67% less than  $S_0$ . No significant differences were observed between  $S_1$  and  $S_2$  and between  $S_2$  and  $S_3$  conditions. But, the difference between  $S_1$  and  $S_3$  was significant (Table 3). Inoculation with AMF increased tuber fresh weight significantly as compared to  $M_0$  plants. The lowest and the highest values of tuber fresh weight were recorded for  $M_0$  and  $G_i$ , respectively. Inoculation with *G. intraradices* showed 77.80% more tuber fresh weight than  $M_0$ . There was no significant difference between tuber fresh weight of  $G_e$  and  $G_{ei}$ . However, the differences of  $G_i$  with  $G_e$  and  $G_{ei}$  were significant (Table 4). Tuber fresh weight of NM and AM plants decreased significantly at  $S_1$ ,  $S_2$  and  $S_3$  as compared to  $S_0$

condition. This happened when the plants were inoculated with *G. etunicatum* until the  $S_2$  level, but it stayed nearly constant at  $S_3$ . In all water levels the maximum tuber fresh weight was recorded when inoculated with *G. intraradices*. The highest and the lowest values of tuber fresh

weight were observed for  $S_0G_i$  and  $S_3G_{ei}$ , respectively (Table 5). Tuber fresh weight for  $S_0G_i$  was 84.26% more than  $S_0M_0$ . Also, at  $S_3G_{ei}$  condition it was 4.29% higher and 54.05% lower than  $S_3M_0$  and  $S_0M_0$ , respectively.

**Table 4. Means of potato traits under study at different arbuscular mycorrhizal fungi types**

Arbuscular mycorrhizal fungi	Stomatal resistance (sec cm <sup>-1</sup> )	Leaf relative water content (%)	Chlorophyll content (CCI)	Tuber fresh weight (g)
M <sub>0</sub>	26.46	57.28	46.50	260.83
G <sub>e</sub>	26.29	59.57	56.36	356.07
G <sub>i</sub>	30.92	61.36	55.38	463.76
G <sub>ei</sub>	38.47	62.43	54.00	367.96
LSD (1%)	6.54	2.51	4.21	66.83

M<sub>0</sub>, G<sub>e</sub>, G<sub>i</sub> and G<sub>ei</sub> represent no application of AMF, inoculation with *G. etunicatum* and *G. intraradices* and their dual inoculation, respectively. LSD: Least significant difference

**Table 5. Means of studied attributes as affected by water levels and arbuscular mycorrhizal fungi**

Water level	Arbuscular mycorrhizal fungi level	Stomatal resistance (sec cm <sup>-1</sup> )	Leaf relative water content (%)	Chlorophyll content (CCI)	Tuber fresh weight (g)
S <sub>0</sub>	M <sub>0</sub>	0.87	71.52	45.80	362.38
	G <sub>e</sub>	0.81	63.91	53.00	580.38
	G <sub>i</sub>	4.84	64.53	62.01	667.75
	G <sub>ei</sub>	19.4	67.59	50.62	593.97
S <sub>1</sub>	M <sub>0</sub>	30.5	53.53	49.88	272.89
	G <sub>e</sub>	31.67	54.43	74.60	320.45
	G <sub>i</sub>	41.67	59.03	54.27	432.24
	G <sub>ei</sub>	56.83	62.24	59.92	371.06
S <sub>2</sub>	M <sub>0</sub>	23.47	52.07	45.94	234.11
	G <sub>e</sub>	25.67	62.27	50.49	261.50
	G <sub>i</sub>	42.67	60.55	48.99	380.24
	G <sub>ei</sub>	48.17	61.26	43.10	340.32
S <sub>3</sub>	M <sub>0</sub>	51.00	52.00	44.41	173.97
	G <sub>e</sub>	47.00	57.68	47.35	261.95
	G <sub>i</sub>	34.50	61.34	56.24	374.84
	G <sub>ei</sub>	29.47	58.64	62.37	166.49
LSD		13.08	5.03	8.42	98.63
		P≤0.01	P≤0.01	P≤0.01	P≤0.05

S<sub>0</sub>, S<sub>1</sub>, S<sub>2</sub> and S<sub>3</sub> represent water level after 60, 90, 120 and 150 mm evaporation from class A pan, respectively. M<sub>0</sub>, G<sub>e</sub>, G<sub>i</sub> and G<sub>ei</sub> show no application of AMF, inoculation with *G. etunicatum* and *G. intraradices* and their dual inoculation, respectively. LSD: Least significant difference

## Discussion

Water deficit increased stomatal resistance as compared to the normal condition (S<sub>0</sub>). It has been reported that stomatal resistance of potato plants (*Solanum tuberosum* cv. Agria) was increased with the enhancement of water deficit stress (Khosravifar *et al.* 2008). Also, Kawakami *et al.*

(2006) showed that in dry plots, stomatal conductance of two potato cultivars (Norin 1 and Konafubuki) derived from both microtubers and conventional seeds, were less than wet plots. It has been suggested that stomatal resistance increase is one of the drought avoidance mechanisms that is used by plants as a strategy to

resist against drought stress (Levitt 1980; Kozłowski and Pallardy 2002; Poorter and Markesteijn 2008). It seems that the potato plant uses this mechanism to withstand drought via avoidance of water waste by stomata.

This study showed that AMF inoculation increased stomatal resistance as compared with  $M_0$ . Ruiz-Lozano *et al.* (1995) suggested that AMF may affect the stomatal sensitivity via increasing abscisic acid production. With the exception to  $S_0G_e$ , stomatal resistance of AM plants was more than NM plants as water level changed from  $S_0$  to  $S_2$ . At  $S_3$ , the stomatal resistance of AM plants was less than  $M_0$ . At all water levels, the stomatal behavior was similar in  $M_0$  and  $G_e$ . There are evidences that AM plants can overcome to water deficit stress through stomatal regulation (Levy and Krikun 1980; Stevens and Peterson 1996). However, it was shown that stomatal conductance of lettuce plants (*Lactuca sativa* L. cv. Romana) colonized with *Glomus deserticola*, *Glomus fasciculatum*, *Glomus mosseae*, *Glomus etunicatum*, *Glomus intraradices*, *Glomus caledonium* and *Glomus occultum* were more than NM plants in the well-watered condition. This status was also observed in the drought stress condition except for *G. occultum* (Ruiz-Lozano *et al.* 1995). Our results showed that AM potato plants may not overcome the water deficit stress at  $S_3$  through increase of the stomatal resistance.

Levels of water deficit stress significantly decreased RWC, as compared to  $S_0$ . Results of Khosravifar *et al.* (2008) on potato (*Solanum tuberosum* cv. Agira) showed that RWC decreased with the enhancement of water deficit

stress. Our study showed that RWC of AM plants was more than NM plants. Gholamhoseini *et al.* (2013) also, reported that RWC of AM leaves was significantly more than NM leaves in sunflower (*Helianthus annuus* cv. Alestar) in 2006 but not significant in 2007. In our study application of AMF at  $S_0$  condition could not increase the RWC as compared with  $M_0$ . Hernandez-Sebastia *et al.* (1999) showed that RWC of leaf discs of in-vitro strawberry plantlets was not affected by AMF under high humidity levels. Furthermore, Abo-Ghalia and Khalafallah (2008) reported that the shoot water content of mycorrhizal wheat plants (inoculated by mixture of *Glomus* spp.) was increased as compared to NM plants in the well water condition but the difference was not significant. Our results showed that AMF application may not be effective in the improvement of RWC at  $S_0$  condition, but, in all water stress levels, the RWC of AM plants was more than NM plants. Increasing of leaf RWC in AM potato plants under water deficit condition indicates that these plants have high root hydraulic conductivity at this condition. The root conductance improvement can be related to the longer root and an alteration in the root system induced by mycorrhizae (Kapoor *et al.* 2008). Aliasghar zad *et al.* (2006) reported that in all soil moisture levels with the field capacity (FC) of 0.85, 0.7 and 0.6, leaf RWC of the inoculated soybean plants with *Glomus mosseae* and *Glomus etunicatum* was more than NM plants at both flowering and maturation stages.

The chlorophyll content of water stress treatments was not significantly different from the control. There are different reports about the

effect of water deficit on chlorophyll content in different plants. Khosravifar *et al.* (2008) showed that chlorophyll content of potato plants increased with the enhancement of water deficit stress. Also, Heidari and Golpayegani (2012) reported that chlorophyll content of basil (*Osimum basilicum* L.) increased with changing of soil humidity from 80% field capacity to 40%. However, on the base of other studies, chlorophyll content of *Erythrina variegata* (Manoharan *et al.* 2010) and marigold (Asrar *et al.* 2011) decreased with increasing of water deficit.

Our study showed that chlorophyll content was obviously increased in the AM potato plants as compared to NM plants. This increase was also reported by Mathur and Vyas (2000) in AM of *Ziziphus mauritiana* as compared to NM. Moreover, results of Meenakshisundaram and Santhaguru (2010) revealed that chlorophyll content in the leaves of sorghum (*Sorghum bicolor* L.) plants colonized with *Glomus fasciculatum*, *Glomus mosseae*, *Glomus dimorphicum* and *Glomus tubaeformis* was significantly more than NM plants. Also, there are other reports about the increase of chlorophyll a and b content in the leaves of AM *Rosa hybrida* L. (Auge *et al.* 1987), pepper (Demir 2004) and wheat (Abo-Ghaila and Khalafallah 2008). The carbon derived from photosynthesis to maintain the performance of AM symbioses is often referred to the “cost”, and the nutrients obtained through the symbiosis are often referred to the “benefit” of the symbiosis (Koide and Elliot 1989). The increase in chlorophyll content by AMF inoculation may improve the carbon requirement as the cost of AM symbiosis and

consequently improves the symbiosis especially in the drought condition. Furthermore, it has been suggested that high chlorophyll content of AM plants as compared to NM plants, is sometimes related to high photosynthesis rate or enhancement in nitrogen and magnesium content (as major components of chlorophyll molecules) in the AM plants (Mathur and Vyas 1995). With the exception to S<sub>2</sub>G<sub>ei</sub>, chlorophyll content of AM plants was more than M<sub>0</sub> in all water levels. It has been reported that in both water deficit and well-watered conditions, chlorophyll a and b content of AM (inoculated with *Glomus constrictum*) was more than AM plants in marigold (Asrar *et al.* 2011). It has been suggested that AM fungi can help plants to confront photoinhibition and photodestruction of pigments under stress condition through the increase of carotenoids. Carotenoids protect photosynthetic apparatus of plants in the drought condition dealing with oxygen radical as agent of photoinhibitory damaging (Foyer and Harbinson 1994).

Water deficit had adverse effect on tuber fresh weight. Khosravifar *et al.* (2008) reported that water deficit stress decreased tuber fresh weight of potato plants (*Solanum tuberosum* L. cv Agria). Lahlou *et al.* (2003) showed that final tuber fresh yield of different potato cultivars decreased by drought stress in the field. Potato yield decrease in the drought condition can be mediated by the decrease in plant leaf size as the first morphological change (Jefferies and MacKerron 1987) and consequently decrease dry matter accumulation in tubers as a result of decrease in radiation interception (Jefferies 1993).



Our study revealed that AMF inoculation increased tuber fresh weight. However, this increase was different at AMF levels. Yao *et al.* (2002) also reported the increase of tuber fresh weight in micropropagated potato plantlets through inoculation with *Glomus intraradices* and *Glomus etunicatum*, but the increase was more pronounced in *G. etunicatum*. Duffy and Cassells (2000) suggested that mycorrhizal fungi can increase or decrease crop yield depending on the mycorrhizal isolate and host genotype. Our results showed that tuber fresh weight of all NM and AM plants decreased with the enhancement of water deficit intensity as compared to S<sub>0</sub>. Also, in all water levels tuber fresh weight of AMF potato plants was more than NM plants, except for the dual inoculation of AMF at the S<sub>3</sub> level. Guarav *et al.* (2010) showed that potato tuber yield of Kufri Badshah cultivar decreased in both AM and NM plants under water deficit stress, but the yield was higher in the stressed and non-stressed AM plants as compared to their related NM controls. Some

researchers suggested that the increase of crop yield by AMF colonization under drought stress may occur through high root surface absorption area and densely proliferate root growth (Kothari *et al.* 1990) or differences in the root systems hydraulic attribute (Auge *et al.* 1994).

### Conclusion

The results indicated that the enhancement of water deficit stress increased stomatal resistance and decreased relative water content and tuber fresh weight. For both stomatal resistance and relative water content, the higher values were observed with the dual inoculation of *Glomus etunicatum* and *Glomus intraradices*. Also, the maximum chlorophyll content and tuber fresh weight were recorded when inoculated with *Glomus etunicatum* and *Glomus intraradices*, respectively. In both control and water deficit conditions, the inoculation with *Glomus intraradices* had the best effect on potato tuber fresh weight.

### References

- Abo-Ghaila HH and Khalafallah AA, 2008. Responses of wheat plants associated with arbuscular mycorrhizal fungi to short-term water stress followed by recovery at three growth stages. *Journal of Applied Sciences Research* 4: 570-580.
- Aliasgharzad N, Neyshabouri MR and Salimi G, 2006. Effects of arbuscular mycorrhizal fungi and *Bradyrhizobium japonicum* on drought stress of soybean. *Biologia, Bratislava* 19: 324-328.
- Al-Karaki GN and Clark RB, 1998. Growth, mineral acquisition, and water use by mycorrhizal wheat grown under water stress. *Journal of Plant Nutrition* 21: 263-276.
- Allen MF, 1982. Influence of vesicular-arbuscular mycorrhiza on water movement through *Buteloua gracilis* (H.B.K.) Lax Ex Steud. *New Phytologist* 91: 191-196.
- Asrar AWA, Khalid A and Elhindi M, 2011. Alleviation of drought stress of marigold (*Tagetes erecta*) plants by using arbuscular mycorrhizal fungi. *Saudi Journal of Biological Sciences* 18: 93-98.
- Auge RM, Duan X, Ebel RC and Stodola AJ, 1994. Nonhydraulic signaling of soil drying in mycorrhizal maize. *Planta* 193: 74-82.
- Auge RM, Schekel KA and Wample RL, 1987. Leaf water and carbohydrate status of VA mycorrhizal rose exposed to drought stress. *Plant and Soil* 99: 291-302.
- Boyer JS, 1982. Plant productivity and environment. *Science* 218: 443-448.

- Cvikrova M, Gemperlova L, Martincova O and Vankova R, 2013. Effect of drought and combined drought and heat stress on polyamine metabolism in proline-over-producing tobacco plants. *Plant Physiology and Biochemistry* 73: 7-15.
- DeFauw SL, He Z, Larkin RP and Mansour SA, 2012. Sustainable potato production and global food security. In: He Z, Larkin R and Honeycutt W (Eds). *Sustainable Potato Production: Global Case Studies*. Pp. 3-19. Springer.
- Dell'Amico J, Torrecillas A, Rodriguez P, Morte A and Sanchez-Blanco MJ, 2002. Responses of tomato plants associated with the arbuscular mycorrhizal fungus *Glomus clarum* during drought and recovery. *Journal of Agricultural Science* 138: 387-393.
- Demir S, 2004. Influence of arbuscular mycorrhiza on some physiological growth parameters of pepper. *Turkish Journal of Biology* 28: 85-90.
- Duffy EM and Cassells AC, 2000. The effect of inoculation of potato (*Solanum tuberosum* L.) microplants with arbuscular mycorrhizal fungi on tuber yield and tuber size distribution. *Applied Soil Ecology* 15: 137-144.
- Echave M, Conti M, Clúa A, Ruscitti M and Beltrano J, 2005. Responses of mycorrhizal infection in the drought resistance and growth of *Lotus glaber*. *Lotus Newsletter* 35: 182-186.
- Ellis JR, Larsen HJ and Boosalis MG, 1985. Drought resistance of wheat plants inoculated with vesicular mycorrhizae. *Plant and Soil* 86: 369-378.
- Faber BA, Zasoski RJ, Munns DN and Schakel K, 1991. A method for measuring hyphal nutrient and water uptake in mycorrhizal plants. *Canadian Journal of Botany* 69: 87-94.
- Fitter AH, 1985. Functioning of vesicular-arbuscular mycorrhizas under field conditions. *New Phytologist* 99: 257-265.
- Foyer CH and Harbinson J, 1994. Oxygen metabolism and the regulation of photosynthetic electron transport. In: Foyer CH and Mullineaux PM (Eds). *Causes of Photooxidative Stress and Amelioration of Defense System in Plants*. Pp. 1-4. CRS Press, Boca Raton.
- Gaurav SS, Sirohi SPS, Singh B and Sirohi P, 2010. Effect of mycorrhiza on growth, yield and tuber eformity in potato (*Solanum tuberosum* L.) grown under water stress condition. *Progressive Agriculture* 10: 35-41.
- Gholamhoseini M, Ghalavand A, Dolatabadian A, Jamshidi E and Khodaei-Joghan A, 2013. Effects of arbuscular mycorrhizal inoculation on growth, yield, nutrient uptake and irrigation water productivity of sunflowers grown under drought stress. *Agricultural Water Management* 117: 106-114.
- Guenthner J, 2010. Production by region and season. In: Bohl WH and Johnson SB (Eds). *Commercial Potato Production in North America*. Pp. 13-16. USDA Agricultural Research Service.
- Hamel C and Strullu DG, 2006. Arbuscular mycorrhizal fungi in field crop production: potential and new direction. *Canadian Journal of Plant Science* 86: 941-950.
- Hardie K, 1985. The effect of removal of extra radical hyphae on water uptake by VAM plants. *The New Phytologist* 101: 677-684.
- Hardie K and Leyton L, 1981. The influence of vesicular-arbuscular mycorrhizae on growth and water relations of red clover. *New Phytologist* 89: 599-608.
- Heidari M and Golpayegani A, 2012. Effects of water stress and inoculation with plant growth promoting rhizobacteria (PGPR) on antioxidant status and photosynthetic pigments in basil (*Ocimum basilicum* L.). *Journal of the Saudi Society of Agricultural Sciences* 11: 57-61.
- Hernandez-Sebastia C, Piche Y and Desjardins Y, 1999. Water relations of whole strawberry plantlets in vitro inoculated with *Glomus intraradices* in a tripartite culture system. *Plant Science* 143: 81-91.
- Hoerling M and Kumar A, 2003. The perfect ocean for drought. *Science* 299: 691-694.
- Huang RS, Smith WK and Yost RE, 1985. Influence of vesicular arbuscular mycorrhizae on growth, water relation and leaf orientation in *Lcucaena icucocephala* (Lam.) de Wit. *New Phytologist* 99: 229-243.
- Jefferies RA, 1993. Responses of potato genotypes to drought. I. Expansion of individual leaves and osmotic adjustment. *Annals of Applied Biology* 122: 93-104.
- Jefferies RA and Mackerron DKL, 1987. Responses of potato genotypes to drought. II. Leaf area index, growth and yield. *Annals of Applied Biology* 122: 105-112.
- Kapoor R, Sharma D and Bhatnagar AK, 2008. Arbuscular mycorrhizae in micropropagation systems and their potential applications. *Scientia Horticulturae* 116: 227-239.

- Kawakami J, Iwama K and Jitsuyama Y, 2006. Soil water stress and the growth and yield of potato plants grown from microtubers and conventional seed tubers. *Field Crops Research* 95: 89–96.
- Khalafallah AA and Abo-Ghaila HH, 2008. Effect of arbuscular mycorrhizal fungi on the metabolic products and activity of antioxidant system in wheat plants subjected to short-term water stress, followed by recovery at different growth stages. *Journal of Applied Sciences Research* 5: 559-569.
- Khosravifar S, Yarnia M, Khorshidi Benam MB and Hosseinzadeh Moghbeli AH, 2008. Effect of potassium on drought tolerance in potato cv. Agria. *Journal of Food, Agriculture & Environment* 6: 236-241.
- Koide R, 1985. The nature of growth depressions in sunflower caused by vesicular arbuscular infection. *New Phytologist* 99: 449-462.
- Koide R and Elliot G, 1989. Cost, benefit and efficiency of the vesicular-arbuscular mycorrhizal symbiosis. *Functional Ecology* 3: 252–255.
- Kothari SK, Marschner H and George E, 1990. Effects of VA mycorrhizal fungi and rhizosphere microorganisms on root and shoot morphology, growth and water relations in maize. *New Phytologist* 116: 303-311.
- Kozlowski T and Pallardy S, 2002. Acclimation and adaptive responses of woody plants to environmental stresses. *The Botanical Review* 68: 270-334.
- Lahlou O, Ouattar S and Ledent JF, 2003. The effect of drought and cultivar on growth parameters, yield and yield components of potato. *Agronomie* 23: 257-268.
- Levitt J, 1980. Stress terminology. In: Tuner NC and Kramar PJ (Eds). *Adaptation of Plants to Water and High Temperature Stress*. Pp. 437-439. Wiley, New York.
- Levy Y and Krikun J, 1980. Effect of vesicular-arbuscular on *Citrus jambhiri* water relations. *New Phytologist* 85: 25-31.
- Manoharan PT, Shanmugaiah V, Balasubramanian N, Gomathinayagam S, Sharma MP and Muthuchelian K, 2010. Influence of AM fungi on the growth and physiological status of *Erythrina variegata* Linn. grown under different water stress conditions. *European Journal of Soil Biology* 46: 151-156.
- Marschner H and Dell B, 1994. Nutrient uptake in mycorrhizal symbiosis. *Plant and Soil* 159: 89-102.
- Mathur N and Vyas A, 1995. Influence of VA mycorrhizae on net photosynthesis and transpiration of *Ziziphus mauritiana*. *Journal of Plant Physiology* 147: 328–330.
- Mathur N and Vyas A, 2000. Influence of arbuscular mycorrhizae on biomass production, nutrient uptake and physiological changes in *Ziziphus mauritiana* Lam. under water stress. *Journal of Arid Environments*. 45: 191-195.
- Meenakshisundaram M and Santhaguru K, 2010. Studies on association of arbuscular mycorrhizal fungi with *Gluconacetobacter diazotrophicus* and its effect on improvement of *Sorghum bicolor* (L.). *International Journal of Current Scientific Research* 1: 23 – 30.
- Moawad AM, 1979. Ecophysiology of VAM in tropics. In: Harley JL and Russell RS (Eds). *The Soil- Root Interface*. Pp. 197-209. Academic Press, London.
- Poorter L and Markesteijn L, 2008. Seedling traits determine drought tolerance of tropical tree species. *Biotropica* 40: 321-331.
- Puppi G and Bras A, 1990. Nutrient and water relations of mycorrhizal white clover. *Agriculture, Ecosystems & Environment* 29: 317-322.
- Quilambo OA, 2003. The vesicular-arbuscular mycorrhizal symbiosis. *African Journal of Biotechnology* 2: 539-546.
- Ramakrisnan B, Jorri BN and Gupta RK, 1990. The response of mycorrhizal maize plants to variations in water potentials. *Current Trends in Mycorrhizal Research* 1: 61-62.
- Ruiz-Lozano JM, Azcon R and Gomez M, 1995. Effects of arbuscular-mycorrhizal *Glomus* species on drought tolerance: physiological and nutritional plant responses. *Applied and Environmental Microbiology* 61: 456-460.
- Ruiz-Lozano JM, Roussel H, Gianinazzi S and Gianinazzi-Perason V, 1999. Defense genes are differentially induced by a mycorrhizal fungus and *Rhizobium* sp. in a wild-type and symbiosis-defective pea genotypes. *Molecular Plant Microbe Interaction* 12: 976-984.
- Sanchez-Diaz M and Honrubia M, 1994. Water relations and alleviation of drought stress in mycorrhizal plants. In: Gianninazi S and Schuepp H (Eds). *Impact of Arbuscular Mycorrhizas on Sustainable Agriculture and Natural Ecosystems*. Pp. 167-178. Birkhauser Basel.

- Sharif M and Moawad AM, 2006. Arbuscular mycorrhizal incidence and infectivity of crops in North West Frontier Province of Pakistan. *World Journal of Agricultural Sciences* 2: 123-132.
- Smith SE and Read D, 2008. *Mycorrhizal Symbiosis*. 3rd edition. Academic Press.
- Stevens KJ and Peterson RL, 1996. The effect of a water gradient on the vesicular-arbuscular mycorrhizal status of *Lythrum salicaria* L. (Purple loosestrife). *Mycorrhiza* 6: 99-104.
- Sylvia DM, Hammond LC, Bennett JM, Haas JH and Linda SB, 1993. Field response of maize to a VAM fungus and water management. *Agronomy Journal* 85: 193-198.
- Vonreichenbach HG and Schonbeck F, 1995. Influence of VA-mycorrhiza on drought tolerance of flax (*Linum uaitissimum* L). 2. Effect of VA-mycorrhiza on stomatal gas exchange, shoot water potential, phosphorus nutrition and the accumulation of stress metabolites. *Journal of Applied Botany* 69: 183-88.
- Xoconostle-Cazares B, Ramirez-Ortega FA, Flores-Elenes L and Ruiz-Medrano R, 2010. Drought tolerance in crop plants. *American Journal of Plant Physiology* 5: 241-6.
- Yao MK, Tweddell RJ and Desilets H, 2002. Effect of two vesicular arbuscular mycorrhizal fungi on the growth of micropropagated potato plantlets and on the extent of disease caused by *Rhizoctonia solani*. *Mycorrhiza* 12: 235-242.