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# Response of Sunflower Plant to the Application of Certain Vitamins and Arbuscular Mycorrhiza Under Different Water Regimes

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**Abstract:** The aim of the present work is to study the effect of application of α-tocopherol, nicotinamide and fungi (mycorrhiza) at different water holding capacities (WHCs) on growth, some biochemical aspects and yield components of sunflower plants and also to raise the efficiency of sunflower plants to resist water stress and to reduce the amount of water used for irrigation. The experiment was carried out in the greenhouse of the National Research Centre using the sunflower (Giza 102). Sandy soil was brought from Nubariya farm of National Research Centre El-Behaira Governorate, Egypt and the proportion of sand is about 94%. The recommended doses of mycorrhizal fungi were applied on soil before planting. Different concentrations of α-tocopherol and nicotinamide were sprayed after 20 days of cultivation. The plants were raised at different levels of WHC 80%, 60% and 30%. Our results highlight that cultivation of sunflower plant in the presence of mycorrhiza and either α-tocopherol or nicotinamide led to increases in growth parameters and yield components concomitantly with an increase in the levels of IAA, GA, photosynthetic pigments, total carbohydrates, total nitrogen, some minerals (K, Ca, Mg, P) and a decrease in ABA content. Moreover, the same treatments induced a potent effect in regulating the stomatal aperture in plants under different levels of WHCs when compared to plants grown without mycorrhiza. As a conclusion, cultivation of sunflower plant in the presence of mycorrhiza with α-tocopherol was the most effective particularly in the presence of 60% water holding capacity.

**Key words:** Arbuscular mycorrhizal, α-Tocopherol, Sunflower, Nicotinamid , Water regimes, Yield.

## INTRODUCTION

Drought is perhaps the major factor limiting crop production worldwide Jones and Corlett, 1992. Drought stress is characterized by reduction of water content, diminished leaf water potential and turgor loss, closure of stomata and decrease in cell enlargement and growth. Severe water stress may result in the arrest of photosynthesis, disturbance of metabolism and finally the death of plant Jaleel *et al.,* 2008 a. Water stress inhibits cell enlargement more than cell division. It reduces plant growth by affecting various physiological and biochemical processes, such as photosynthesis, respiration, translocation, ion uptake, carbohydrates, nutrient metabolism and growth promoters Jaleel *et al*., 2008 b.

Sunflower is a major oil seed crop worldwide, and it is also an important crop in Mediterranean areas and in the Arab Republic of Egypt where water stress is an increasing problem Caterina, *et al.,* 2007. Survival under these stressful conditions depends on the plant's ability to perceive the stimulus, generates and transmits signals and instigates biochemical changes that adjust the metabolism accordingly Dolatabadian & Saleh 2009. Reactive oxygen species such as superoxide radical ($O^-_2$), hydrogen peroxide ($H_2O_2$) and hydoxyl ($OH^-$) radical are also produced during water stress, and are responsible for the damage to membranes and other essential macromolecules such as photosynthetic pigments, proteins, DNA and lipids. Out of various compounds exploited to alleviate the plant stress, vitamins from which nicotinamide and α-tocopherol. Exogenous applications of vitamins have been reported to successfully mitigate the adverse effects of water stress on plants Khan *et al*., 2006. Vitamins are organic compounds which are required in trace amount to maintain normal growth and proper development of all organisms; these compounds act as coenzymes and thus take essential part in the regulation of metabolism, vitamins, can be limiting factors in the development of plant Hassanein *et al*., 2009.

Nicotinamide (Vit. B) is a well-characterized constituent of the pyridine dinucleotide coenzymes $NAD^+$& $NADP^+$, which are involved in many enzymatic oxidations - reductions reactions in living cells Berglund, 1994. In addition, nicotinamide is a stress – associated compound that induces and regulate secondary metabolic accumulation and/or the manifestation of defense metabolism in plants Berglund & Ohlsson, 1995.

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Nicotinamide might be a link between various types of stress which leads to an increased frequency of DNA strand breaks, and plant defensive gene transcription Berglund, 1994.

α-tocopherol (Vit. E) is lipophilic antioxidants synthesized by all plants. α-tocopherol protects lipid membranes from oxidative stress because they deactivate singlet oxygen, reduce superoxide radicals, and terminate lipid peroxidation by reducing fatty acyl peroxy radicals Munne-Bosch *et al*., 1999. α-tocopherols interact with the polyunsaturated acyl groups of lipids, stabilize membranes and scavenge and quench various reactive oxygen species (ROS) Cvetkovska *et al*., 2005. Increasing evidence suggests that in higher plants vitamin E might play a protective role to membrane system in cell. This evidence points at α-tocopherol being an important part of the plant defense machinery maintaining the integrity and normal function of the photosynthetic apparatus Collin *et al*., 2007. Arbuscular mycorrhizal fungi symbiosis contributes to enhance growth and vigor of plants, and can alter plant water relations, particularly during water stress periods Augé, 2001. The arbuscular mycorrhizal (AM) fungi enhance tolerance of plant to water deficit through the alteration of plant physiology Ricardo Aroca *et al.*, 2008.The mechanisms involved in water uptake by the AMF symbiosis include regulation of stomatal conductance, an increase in stomatal sensitivity to leaf-air vapor pressure deficit, and lowering leaf osmotic potential for turgor maintenance Sánchez-Blanco *et al.,* 2004. AM fungi enhance the growth of the plant by improving properties of soil in rhizosphere Huixing Song 2005. They absorb P and other nutritional elements and then improve nutritional status of host plant Plassard and Dell 2010. In exchange, the plant supplies the fungus with sugars. Mycorrhizal fungi have been suggested as having a role in mediating the uptake of water at times of drought stress Al-Karaki*et al* 2004.

The aim of the present work is to study the effect of application of α-tocopherol, nicotinamide and fungi (mycorrhiza) at different water holding capacity on growth and yield components of sunflower plants and also to raise the efficiency of sunflower plants to resist water stress and to reduce the amount of water used for irrigation.

## MATERIALS AND METHODS

***Experimental Conditions Plant Materials, Growth and Treatment Conditions:***

One variety of sunflower (*Helianthus annuus L.*) cv. Giza 102 was obtained from Agriculture Research Centre for Oil Crops Research Institute Giza, Egypt. Seeds were grown in Pots (diameter 50 cm); filled with sand soil brought from a Nubariya farm and the proportion of sand equal 94%, after the inoculation the soil by the recommended amount micorrohyzal fungi. The irrigation treatments were given to plants with different levels of water holding capacity 80%, 60% and 30%. Alpha-tocopherol concentrations (0.0, 200 and 400 ppm) and nicotinamide concentrations (0.0, 40 and 80 ppm) were sprayed after 21and 28 days of cultivation. The fertilization with super phosphate (5 g / pot), potassium sulfate (25 g / pot) and urea (6 g / pot) were used. Samples were taken after 37 days from sowing to take the growth parameters and determine different chemical analysis and one plant as been left in each pot to yield determinations.

The treatments were as follows:

| No. | Without micorrhyza | No. | With micorrhyza |
|---|---|---|---|
| 1 | Control at 80% of W.H.C. | 15 | Control at 80% of W.H.C. |
| 2 | 40ppm Nicotinamide + 80% WHC | 17 | 40ppm Nicotinamide + 80% WHC |
| 3 | 80ppm Nicotinamide + 80% WHC | 18 | 80ppm Nicotinamide + 80% WHC |
| 4 | 200ppm α Tocopherol + 80% WHC | 19 | 200ppm α Tocopherol + 80% WHC |
| 5 | 400ppm α Tocopherol + 80% WHC | 20 | 400ppm α Tocopherol + 80% WHC |
| 6 | Control at 60% of W.H.C. | 21 | Control at 60% of W.H.C. |
| 7 | 40ppm Nicotinamide + 60% WHC | 22 | 40ppm Nicotinamide + 60% WHC |
| 8 | 80ppm Nicotinamide + 60% WHC | 23 | 80ppm Nicotinamide + 60% WHC |
| 9 | 200ppm α Tocopherol + 60% WHC | 24 | 200ppm α Tocopherol + 60% WHC |
| 10 | 400ppm α Tocopherol + 60% WHC | 25 | 400ppm α Tocopherol + 60% WHC |
| 11 | Control at 30% of W.H.C. | 26 | Control at 30% of W.H.C. |
| 12 | 40ppm Nicotinamide + 30% WHC | 27 | 40ppm Nicotinamide + 30% WHC |
| 13 | 80ppm Nicotinamide + 30% WHC | 28 | 80ppm Nicotinamide + 30% WHC |
| 14 | 200ppm α Tocopherol + 30% WHC | 29 | 200ppm α Tocopherol + 30% WHC |
| 15 | 400ppm α Tocopherol + 30% WHC | 30 | 400ppm α Tocopherol + 30% WHC |

***Experimental Design, Growth and Yield Analyses:***

The pot experiment was conducted in the greenhouse of Botany Department, National Research Center. Experimental design was split- split plot design. Where, the inoculation with micorrohyzal fungi was put in the main plot, the irrigation with different water holding capacities was put in the sub plot and the nicotinamide and α-tocopherol treatments were put in the sub-sub plot. Samples were taken after 37 days after sowing to analyse the crop performed in terms of growth parameters, endogenous phytohormones (auxins, gibberellins, and abscisic acid), photosynthetic pigment (chlorophyll a, chlorophyll b and carotenoids), carbohydrate fractions (polysaccharides and total carbohydrates), nitrogen components (protein nitrogen and total nitrogen)and the element compositions (Potassium, Sodium, Calcium, Magnesium and Phosphorus) were also determined. Each

treatment was replicated four times and each replicate had five plants. Single healthy plant was left in each pot to determine the yield components and oil % of the produced seeds. Plants were grown under naturally illuminated environmental conditions of the net house. The pots were watered regularly according to their water holding capacities.

***Stomatal Opening Area:***

Direct microscopic measurements were carried out according to the method described by El-Shahaby (1994). On leaf epidermal strips which were immediately immersed in solute alcohol for fixation and preservation as well. A standardized linerocular micrometer was used in order to measure the area of stomatal opening and calculated by multiplying the tow axes (length and width) by the factor 2/3.

***Endogenous Phytohormones:***

***Extraction, Separation and Determination of Phytohormones:***

The method of hormone extraction was essentially similar to that adopted by Wasfy and Orrin, (1975) and the Methylation process was carried out according to the method described by Vogel (1975). Identification and determination of auxins, gibberellins, and abscisic acid were carried out by Helwett Packered gas liquid chromatography (5890) with a flame ionization detector (Shindy and Smith, 1975). The gas liquid chromatographic conditions for isothermal work was as follows: The chromatograph was fitted and equipped with HP–130 mmX0.32 mm X 0.25 µm capillary column coated with methyl silicon. The column oven temperature was programmed at 10C º/min from 200 Cº (5 min) to 260Cº and kept finally to 10 min. Injector and detector temperatures were 260 and 300Cº respectively. Gases flow rates were 30, 30, 300 cm/sec for $N_2$ , $H_2$ and air, respectively and flow rate inside column was adjusted at 2 ml / min. Standards of IAA, GA and ABA were used.

***Total Chlorophyll and Carotenoids Contents:***

The Photosynthetic Pigments in the fresh fully expanded leaves (chlorophyll a, chlorophyll b and carotenoids) were determined spectrophotometrically by the method recommended by Metzner *et al,.* 1965.

***Total Soluble Sugars (TSS):***

Total soluble carbohydrates (TSS) were extracted by overnight submersion of dry tissue in 10 ml of 80% (v/v) ethanol at 25°C with periodic shaking, and centrifuged at 600g. The supernatant was evaporated till completely dried then dissolved in a known volume of distilled water to be ready for determination of soluble carbohydrates Homme *et al.* 1992. TSS were analyzed by reacting of 0.1 ml of ethanolic extract with 3.0 ml freshly prepared anthrone (150 mg anthrone + 100 ml 72% H2SO4) in boiling water bath for ten minutes and reading the cooled samples at 625 nm using Spekol SpectrocololourimeterVEB Carl Zeiss Yemm and Willis,1994.

***Total Carbohydrate Contents:***

Total carbohydrate content was determined colorimetrically according to Dubois *et al.*, 1956.A known weight of the dry powdered plant was placed in a sugar tube. 20 ml of 1 N $H_2SO_4$ was then added. The tube was boiled under a reflux condenser in boiling water bath for six hours. The combined filtrate was completed to the mark with distilled water. Total sugars were determined colorimetrically as follows: a known volume (1 ml) of the sugar solution was quantitatively transferred into a test tube and 1 ml of 5 % aqueous phenol solution was added followed by 5 ml of concentrated analar sulphuric acid. Measurements of the intensity of the yellow – orange colour was carried out using Spekol Spectrocolourimeter VEB Carl Zeiss at 490 nm. A standard experiment was carried out using distilled water (1 ml) instead of the sugar solution.

***Nitrogenous Contents:***

The total soluble-N (TSN) and total-N (TN) were determined by the conventional micro–Kjeldahl method (Pirie, 1955). A known sample of hot water extract (TSN determination) or dry powderd plant (for TN determination) was put into a digestion flask. Sulphate mixture followed by 1 ml ammonia-free water and 3 ml of ammonia-free $H_2SO_4$ were added. The sample was then incinerated, ammonia distilled off and nitrogen determined as mentioned before. The protein-N content was calculated from the difference between total-N and total soluble-N.

***Mineral Ions:***

Element contents were determined according to the method described by Chapmen & Pratt 1978. P was determined using Spekol Spectrocololourimeter VEB Carl Zeiss. While, estimation of K, Na ,Ca and contents were estimated using atomic absorption spectrophotometer.

***Oil Determination:***

The oil of sunflower seeds were extracted according to Kates and Eberhardt (1957), the powdered seeds is shaken overnight with isopropanol : chloroform (1:1). The solvent were evaporated under reduced pressure of CO2 atmosphere. The lipid residue is taken up in a chloroform : methanol (2:1 v/v) and given a folch wash, the dissolved total oils were purified by washing with 1% aqueous saline solution. The aqueous phases were washed with chloroform that was combined with the pure oil solution. Chloroform was evaporated and the total pure oil was weighed.

***Statistical Analysis:***

The results were statistically analyzed using MSTAT-C (1988) software. The mean comparisons among treatments were determined by Duncan's multiple range test at 5 $P \leq 0.05$.Gomez and Gomez, 1984.

**Results:**

***Plant Growth:***

Table (1) shows the influence of different concentrations of nicotinamide or α-tocopherol on growth criteria of sunflower plants subjected to different levels of water holding capacity in soils amended with microhyzza. Exposure of plants to the 60% and 30% of water capacity in the field conditions leads to a marked decrease in all morphological parameter studied (plant height, area of leaves per plant, stem diameter, fresh and dry weight of shoots) when compared to plants grown under the level of 80% water holding capacity. While progressive increases were observed for the plants cultivated in the presence of mycorrhizal fungi compared to plants cultivated without mycorrhiza. Treatment of sunflower plants with different concentrations of α-tocopherol or nicotinamide increased all growth criteria in the presence and absence of mycorrhiza under different water levels. The maximum increases in all the growth criteria were obtained by using 80% water holding capacity.

***Yield and Yield Components:***

Results in Table (2) illustrates the behaviour of sunflower plant grown under different water regimes andthe effect of foliar application of different concentrations of α-tocopherol or nicotinamide with or without mycorrhiza on yield and yield components. Increasing drought effect resulted in reduction of yield components (head diameter (cm), seeds weight (g)/head, 100 seeds weight (g) and the oil %. The results showed an increase in all yield components in response to inoculation with mycorrhiza. Spraying sunflower plant with α-tocopherol or nicotinamide showed that different concentrations of both used vitamins increased yield components of the sunflower as compared with the corresponding water stress levels particularly in the presence of mycorrhiza. High concentration of either α-tocopherol (400 mg/l) or nicotinamide (80 mg/l) were more effective than low concentrations respectively, under different water holding capacity levels. The maximum increase in oil % (43.28%) was recorded in plants treated with α-tocopherol (400 mg/l) at 60% WHC in the presence of mycorrhiza when compared with either 80 % or 30 % WHC.

***Stomata Opening Area:***

Table (3) illustrates the effect of foliar application of either α-tocopherol or nicotinamide on the area of stomata opening for sunflower plants under different levels of WHC in soils amended with mycorrhiza. The results showed that, the reduction of the WHC caused marked significant decreases the area of stomata opening on both upper and lower epidermis of sunflower plant in the presence and absence of mycorrhizal fungi. Maximum reduction was notice at 30% WHC and the average of reduction 62%, 61%, 50% and 57% in the upper and lower epidermis in the absence and presence of mycorrhiza respectively. The results showed that growing plants in the presence of mycorrhizal fungi led to an increase in stomatal area at all the different levels of WHC compared to the corresponding plants grown in the absence of mycorrhiza. Spraying sunflower plant with different concentrations α-tocopherol or nicotinamide, increased the area of stomata opening on both upper and lower epidermis of the sunflower as compared with the corresponding water stress levels particularly in the presence of mycorrhiza.

***Endogenous Phytohormone:***

Data in Figure (1) show the variation in growth promoters (indole and gibberellins) in response to spraying sunflower plant with different concentrations of either α-tocopherolor nicotinamide which subjected to different levels of WHC in soils amended with microhyzza. The WHC raised at 30% caused marked significant decreases in both GA and IAA and increased ABA content, as compared with those of the corresponding controls. While the reduction of the WHC up to 60% led to significant increases in both GA and IAA and decreased ABA content. There is significant increase in growth promoters (indole and gibberellins) combined with reduction in inhibitor (ABA) in response to inoculating plant with mycorrhiza. Spraying sunflower plants with different concentrations of either α- tocopherol or nicotinamide increased significantly growth promoters accompanied with reduction in ABA as compared with control plant with or without inoculation with mycorrhiza.

**Table (1): Effect of different concentrations of nicotinamide or α-Tocopherol on morphological criteria of sunflower plants subjectedto different levels o holding capacity in absence or holding capacity in absence or presence of microhyzza.**

| Treatment | | Shoot length (cm) | | Area of leaves (cm2) | | Circumferance (cm) | | Fresh weight of shoot (g) | | Dry weight of shoot (g) | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | Material mg/l | - | + | - | + | - | + | - | + | - | + |
| 80 % of water holding capacity | Control (0.00) | 46.5 ± 0.29 e | 47.8 ± 0.15 c | 127.5 ± 0.29 o | 133.7 ± 0.13 n | 1.5 ± 0.00 ghi | 1.6 ± 0.00 e | 14.9 ± 0.04 g | 15.6 ± 0.02 de | 2.1 ± 0.05 hi | 2.2 ± 0.01 fg |
| | Nicotinamide 40 | 47.1 ± 0.12cd | 49 ± 0.58 b | 192.2 ± 0.14 a | 174.1 ± 0.21 b | 1.6 ± 0.06 e | 2.2 ± 0.00 a | 15.7 ± 0.06 de | 17.2 ± 0.01 a | 2.0 ± 0.02 i | 2.3 ± 0.04 bc |
| | Nicotinamide 80 | 51 ± 0.30 a | 49.4± 0.15 b | 172.9 ± 0.15 c | 155.2 ± 0.14 h | 1.7 ± 0.03 d | 1.8 ± 0.06 c | 15.6 ± 0.03 ef | 16.4 ± 0.07 b | 2.2 ± 0.02 ef | 2.3 ± 0.01 cd |
| | α-Tocopherol 200 | 42 ± 0.00g | 49.3 ± 0.17 j | 159.5 ± 0.29 f | 155.6 ± 0.29 h | 1.8 ± 0.00 c | 1.8 ± 0.00 c | 15.3 ± 0.04 f | 16.2 ± 0.34 cd | 2.3 ± 0.02 de | 2.3 ± 0.02 c |
| | α-Tocopherol 400 | 41 ± 0.58 h | 49 ± 0.00 de | 155.3 ± 0.14h | 161.5 ± 0.29 e | 1.9 ± 0.00 b | 1.9 ± 0.00 b | 15.0 ± 0.02 g | 15.9 ± 0.05 c | 2.3 ± 0.04 cd | 2.4 ± 0.05 b |
| 60% of water holding capacity | Control (0.00) | 38.6 ± 0.26 k | 39 ± 0.00 j | 124.8 ± 0.18 q | 126.5 ± 0.02 p | 1.4 ± 0.01 jk | 1.5 ± 0.00 ghi | 11.7 ± 0.09 m | 13.2 ± 0.13 kl | 2.0 ± 0.01 i | 2.1 ± 0.03 i |
| | Nicotinamide 40 | 43.4 ± 0.23 f | 44 ± 0.14 f | 148.9 ± 0.31 m | 163.0 ± 0.08 d | 1.5 ± 0.01 hij | 1.5 ± 0.01 ghi | 13.0 ± 0.08 l | 13.4 ± 0.09 jk | 2.1 ± 0.00 hi | 2.1 ± 0.01 gh i |
| | Nicotinamide 80 | 39.4 ± 0.06 j | 40.6 ± 0.23 i | 149.9± 0.07 l | 154.1 ± 0.14 i | 1.5 ± 0.01 hij | 1.6 ± 0.01 ef | 13.5 ± 0.06 j | 14.6 ± 0.04 h | 2.4 ± 0.01 bc | 2.2 ± 0.03 fg h |
| | α-Tocopherol 200 | 41.6 ± 0.35 gh | 45.8 ± 0.12 ef | 151.6 ± 0.29 k | 152.5 ± 0.09 j | 1.5 ± 0.01 fgh | 1.6 ± 0.00 e | 14.5 ± 0.06 h | 15.7 ± 0.05 cde | 2.4 ± 0.04 bc | 2.5 ± 0.01 a |
| | α-Tocopherol 400 | 39.6 ± 0.18 j | 40.6 ± 0.23 i | 157.6 ± 0.07 g | 154.2 ± 0.05 i | 1.6 ± 0.01 e | 1.9 ± 0.02 b | 14.2 ± 0.04 i | 14.6 ± 0.03 h | 2.3 ± 0.02 c | 2.4 ± 0.01 bc |
| 30 % of water holding capacity | Control (0.00) | 25.1± 0.30p | 27 ± 0.00l | 84.7 ± 0.26 z | 89.7 ± 0.14 y | 1.3 ± 0.00 m | 1.4 ± 0.03 lm | 7.3 ± 0.08 t | 8.2 ± 0.02 s | 1.5 ± 0.01 p | 1.6 ± 0.01 no |
| | Nicotinamide 40 | 36.1 ± 0.03 1 | 36.8 ± 0.12 k | 91.4 ± 0.05 x | 98.8 ± 0.14 v | 1.4 ± 0.01 ijk | 1.6 ± 0.03 efg | 9.3 ± 0.06 q | 9.7 ± 0.07 p | 1.8 ± 0.04 kl | 1.8 ± 0.01 jk |
| | Nicotinamide 80 | 36.3 ± 0.07 lm | 37.8 ± 0.12 k | 104.7 ± 0.07 u | 115.4 ± 0.06 r | 1.3 ± 0.00 m | 1.4 ± 0.02 ijk | 9.6 ± 0.06 p | 10.3 ± 0.06 o | 1.6 ± 0.04 no | 1.7 ± 0.00 lm |
| | α-Tocopherol 200 | 35.5 ± 0.00m | 36.4 ± 0.23 l | 91.6 ± 0.06 x | 108.5 ± 0.11 t | 1.4 ± 0.01 kl | 1.5 ± 0.01 hij | 10.4 ± 0.09 no | 10.7 ± 0.05 n | 1.6 ± 0.02 m n | 1.7 ± 0.02 m n |
| | α-Tocopherol 400 | 34.6 ± 0.23 n | 36.2 ± 0.07 l | 94.9 ± 0.07 w | 113.3 ± 0.06 s | 1.4 ± 0.00 kl | 1.5 ± 0.01 fgh | 8.5 ± 0.05 r | 11.8 ± 0.09 m | 1.5 ± 0.01 p | 1.8 ± 0.01 j |

***Photosynthetic Pigments:***

Data in Figure (2) show the response of photosynthetic pigment of sunflower leaves sprayed with different concentrations of either α-tocopherol or nicotinamide and subjected to different levels of WHC in soils amended with mycorrhiza. The reduction of the WHC up to 60% led to significant increases in (chlorophyll a and chlorophyll b and carotenoids contents and total pigments) while 30 % WHC led to significant decreases in all photosynthetic fractions when compared to 80 %. The results showed that growing plants in the presence of mycorrhizal fungi led to an increase in photosynthetic pigments at all the different levels of water stress when compared to plants grown in the absence of mycorrhiza. Results also reveal that, significant increase in all photosynthetic pigment contents in response to treatment with either α-tocopherol or nicotinamide, under normal condition or under water stress in the presence or absence of mycorrhiza.

**Table 2:** Effect of different concentrations of nicotinamide or α-Tocopherol on yield components of sunflower plants subjected to different levels of water

| Treatment | | Head diameter (cm) | | Seed weight / head (g) | | 100 - seed weight (g) | | Oil % | |
|---|---|---|---|---|---|---|---|---|---|
| | Material mg/l | - | + | - | + | - | + | - | + |
| 80 % of water holding capacity | Control (0.00) | 11.3 ± 0.17 c-g | 12.5 ± 0.17 c-g | 12.4 ± 0.02 i | 14.5 ± 0.06 f | 3.8 ± 0.03 hi | 4.1 ± 0.04 g | 21.1 ± 0.13 v | 26.9 ± 0.11 q |
| | Nicotinamide 40 | 12.5 ± 0.17 a-d | 13.7 ± 0.06 a-d | 14.0 ± 0.02 gh | 17.8 ± 0.04 b | 4.3 ± 0.03 fg | 4.8 ± 0.04 ab | 25.2 ± 0.09 t | 28.8 ± 0.15 n |
| | Nicotinamide 80 | 13.1 ± 0.17 ab | 13.5 ± 0.00 ab | 14.8 ± 0.04 f | 17.4 ± 0.06 c | 4.8 ± 0.06 abc | 4.9 ± 0.06 a | 28.4 ± 0.19 o | 32.4 ± 0.28 h |
| | α-Tocopherol 200 | 12.3 ± 0.12 a-d | 13.1 ± 0.12 a-d | 17.0 ± 0.00 d | 17.7 ± 0.04 bc | 4.6 ± 0.06 cd | 4.7 ± 0.09 cd | 23.4 ± 0.15 u | 36.9 ± 0.31 d |
| | α-Tocopherol 400 | 12.5 ± 0.17 a-d | 12.8 ± 0.00 a-d | 15.7 ± 0.06 e | 20.4 ± 0.23 a | 4.2 ± 0.12 fg | 4.9 ± 0.12 bc | 25.7 ± 0.11 s | 37.5 ± 0.27 c |
| 60% of water holding capacity | Control (0.00) | 10.5 ± 0.00 e-h | 10.9 ± 0.14 e-h | 10.3 ± 0.07 k | 14.0 ± 0.00 g | 3.6 ± 0.03 ij | 3.7 ± 0.06 hi | 25.8 ± 0.14 r | 29.2 ± 0.19 m |
| | Nicotinamide 40 | 11.3 ± 0.00 b-f | 12.4 ± 0.14 b-f | 11.9 ± 0.06 j | 14.8 ± 0.12 f | 4.1 ± 0.06 fg | 4.3 ± 0.01 ef | 29.5 ± 0.14 l | 32.7 ± 0.31 g |
| | Nicotinamide 80 | 11.6 ± 0.12 b-f | 12.2 ± 0.09 b-f | 13.5 ± 0.03 h | 15.7 ± 0.06 e | 4.5 ± 0.06 de | 4.6 ± 0.02 de | 30.5 ± 0.05 j | 33.5 ± 0.26 e |
| | α-Tocopherol 200 | 11.4 ± 0.06 b-f | 11.6 ± 0.12 b-f | 12.3 ± 0.12 i | 14.8 ± 0.00 f | 3.9 ± 0.04 hi | 4.1 ± 0.04 fg | 31.0 ± 0.16 i | 40.3 ± 0.29 b |
| | α-Tocopherol 400 | 11.4 ± 0.00 b-f | 12.0 ± 0.00 b-f | 13.5 ± 0.03 h | 13.6 ± 0.03 h | 4.2 ± 0.09 fg | 4.1 ± 0.03 fg | 33.1 ± 0.22 f | 43.3 ± 0.35 a |
| 30 % of water holding capacity | Control (0.00) | 5.2 ± 0.17 j | 5.8 ± 0.17 j | 6.8 ± 0.07 p | 7.6 ± 0.06 o | 2.5 ± 0.00 n | 2.8 ± 0.03 m | 12.5 ± 0.15 ] | 17.0 ± 0.07 y |
| | Nicotinamide 40 | 7.2 ± 0.00 fgh | 7.4 ± 0.12 fgh | 8.6 ± 0.06 mn | 8.8 ± 0.09 m | 3.1 ± 0.03 l | 3.3 ± 0.00 kl | 14.4 ± 0.11 / | 20.2 ± 0.19 w |
| | Nicotinamide 80 | 7.2 ± 0.12 fgh | 7.4 ± 0.00 fgh | 9.4 ± 0.07 l | 9.6 ± 0.06 l | 3.1 ± 0.08 l | 3.2 ± 0.04 kl | 16.1 ± 0.08 [ | 23.4 ± 0.22 u |
| | α-Tocopherol 200 | 6.8 ± 2.00 j | 6.9 ± 0.14 j | 9.3 ± 0.04 l | 9.3 ± 0.14 l | 3.2 ± 0.06 kl | 3.4 ± 0.06 jk | 16.8 ± 0.14 z | 28.0 ± 0.22 p |
| | α-Tocopherol 400 | 6.6 ± 0.00 hi | 6.9 ± 0.00 hi | 8.0 ± 0.01 o | 8.3 ± 0.17 n | 3.1 ± 0.06 j | 3.2 ± 0.03 l | 18.5 ± 0.17 x | 29.8 ± 0.25 k |

***Carbohydrate Contents:***

Data in Figure (3) demonstrate that, field capacity up to 30% led to marked decrease in total carbohydrates compared to plants grown under 80% of field capacity. On the other hand cultivation of sunflower in the presence of mycorrhiza led to marked increase in total carbohydrates when compared with plants cultivated without micrrohyza and those grown under water stress. The results also illustrate the effect of both α-tocopherol and nicotinamide on plants under normal and under different levels of water stress on total carbohydrates in the presence or absence of micrrohyza compared to untreated plants.

***Nitrogen Contents:***

Figure (3) show that the decrease of WHC of soil led to marked decreases in total soluble nitrogen and total nitrogen contents compared to plants grown under 80% of WHC. Cultivation of sunflower in the presence of micrrohyza led to significant decrease in total soluble content compared to those of other plants without mycorrhiza and under different water stress levels. The opposite trend was observed at protein nitrogen and total

nitrogen contents under the same treatment. The results showed also stimulatory effects of α -tocopherol and nicotinamide under normal or under different levels of water stress on total soluble nitrogen, protein nitrogen and total nitrogen contents in presence or absence of mycorrhiza compared to untreated plants.

**Table (3): Effect of different concentrations of nicotinamide or α-Tocopherol on stomatal number of sunflower plants subjectedto different levels of water holding capacity in absence or holding capacity in absence or presence of microhyzza.**

| Treatment | | | Upper | | Lower | |
|---|---|---|---|---|---|---|
| | Material mg/l | | - | + | - | + |
| 80 % of water holding capacity | Control (0.00) | | 45.40 ± 0.23 h | 56.00 ± 0.51 ab | 40.40 ± 0.19 f | 54.10 ± 0.41 a |
| | Nicotinamide | 40 | 48.30 ± 0.22 e | 56.30 ± 0.53 a | 41.80 ± 0.24 e | 38.30 ± 0.21 h |
| | | 80 | 43.50 ± 0.40 i | 53.50 ± 0.32 c | 39.00 ± 0.15 g | 39.30 ± 0.24 g |
| | α-Tocopherol | 200 | 43.30 ± 0.12 i | 50.20 ± 0.42 d | 42.00 ± 0.29 e | 52.70 ± 0.79 b |
| | | 400 | 47.50 ± 0.46 f | 55.50 ± 0.38 b | 48.30 ± 0.24 c | 44.80 ± 0.39 d |
| 60% of water holding capacity | Control (0.00) | | 33.60 ± 0.53 n | 41.50 ± 0.52 j | 32.00 ± 0.18 j | 38.00 ± 0.12 h |
| | Nicotinamide | 40 | 37.50 ± 0.22 m | 48.30 ± 0.31 e | 29.50 ± 0.31 k | 39.00 ± 0.18 g |
| | | 80 | 38.00 ± 0.18 l | 48.60 ± 0.23 e | 30.00 ± 0.17 k | 41.60 ± 0.24 e |
| | α-Tocopherol | 200 | 39.30 ± 0.23 k | 44.90 ± 0.21 h | 34.00 ± 0.19 i | 41.50 ± 0.29 e |
| | | 400 | 37.30 ± 0.18 m | 46.30 ± 0.23 g | 34.00 ± 0.07 i | 39.00 ± 0.05 g |
| 30 % of water holding capacity | Control (0.00) | | 17.60 ± 0.23 u | 28.30 ± 0.11 p | 15.90 ± 0.16 q | 23.00 ± 0.14 m |
| | Nicotinamide | 40 | 21.00 ± 0.10 t | 27.70 ± 0.12 pq | 19.60 ± 0.09 p | 21.50 ± 0.14 n |
| | | 80 | 21.00 ± 0.11 t | 27.10 ± 0.12 q | 20.00 ± 0.07 p | 22.60 ± 0.09 m |
| | α-Tocopherol | 200 | 25.90 ± 0.22 r | 32.00 ± 0.23 o | 25.20 ± 0.06 l | 20.70 ± 0.11 o |
| | | 400 | 28.00 ± 0.23 p | 22.00 ± 0.17 s | 21.00 ± 0.17 no | 22.40 ± 0.08 m |

***Elements:***

Figure (4) the decrease of WHCto60% led to significant increases in both potassium and magnesium, while 30% WHC led to a marked decrease in potassium content compared to plants grown under 80% of WHCin presence and absence of mycorrhiza. Water stress caused significant decreases in phosphorous and calcium contents of sunflower cultivars with the increases in soil WHC levels in the presence and absence of mycorrhiza (Figure 4) as compared with the corresponding control. Cultivation of sunflower in the presence of mycorrhiza led to a marked increase in the absorption of the elements (potassium, calcium, magnesium and phosphorus) when compared to plants cultivated without micrrohyza and under water stress. The results showed (Figure 4) the foliar spraying of α -tocopherol and nicotinamide led to significant increase in (potassium, calcium, magnesium and phosphorus), either in normal conditions or under different water stress treatments.

***Discussion:***

***Plant Growth and Yield:***

Plant responses to water stress include growth parameters and biochemical changes that lead first to acclimation and later, as water stress become more severe leading to damage and the loss of plant parts Chaves *et al*., 2003.Water deficits affect plants in very different ways, slowly developing water deficits decrease

growth, by slowing rates of cell division and expansion due to loss of turgor and increased synthesis of abscisic acid (Figure 1) Lawlor and Cornic, 2002 and/or resulted from the osmotic effect of water stress which caused disturbances in water balance of stressed sunflower plant leading to stomatal closure (Table 3), reduction in photosynthesis (Figure 2) and consequently a retarded growth rate (Table 1). The decrease in dry weight of shoots by increasing the water stress could be ascribed to the decrease in photosynthetic output as indicated by the significant decreases of chlorophylls and total carbohydrates (Figure 3) in water stressed sunflower plants. Other authors concluded that, reduction of dry weight might be due to a turgor limitation Farooq *et al*., 2009 or cell wall hardening by limited extension growth Chazen and Neumann, 1994.The reasons may be the non-availability of nutrients and the expenditure of energy to counteract the toxic effects of stress. During the acclimation phase, water stress typically results in slower growth rates because of the inhibition of cell expansion, the reduction in carbon assimilation and the resultant effect on carbon partitioning Hsiao and Xu, 2000. Water stress reduced the head diameter, 100- seed weight, yield per plant and Oil% in sunflower. Similar results were obtained by Tahir & Mehid, 2001.

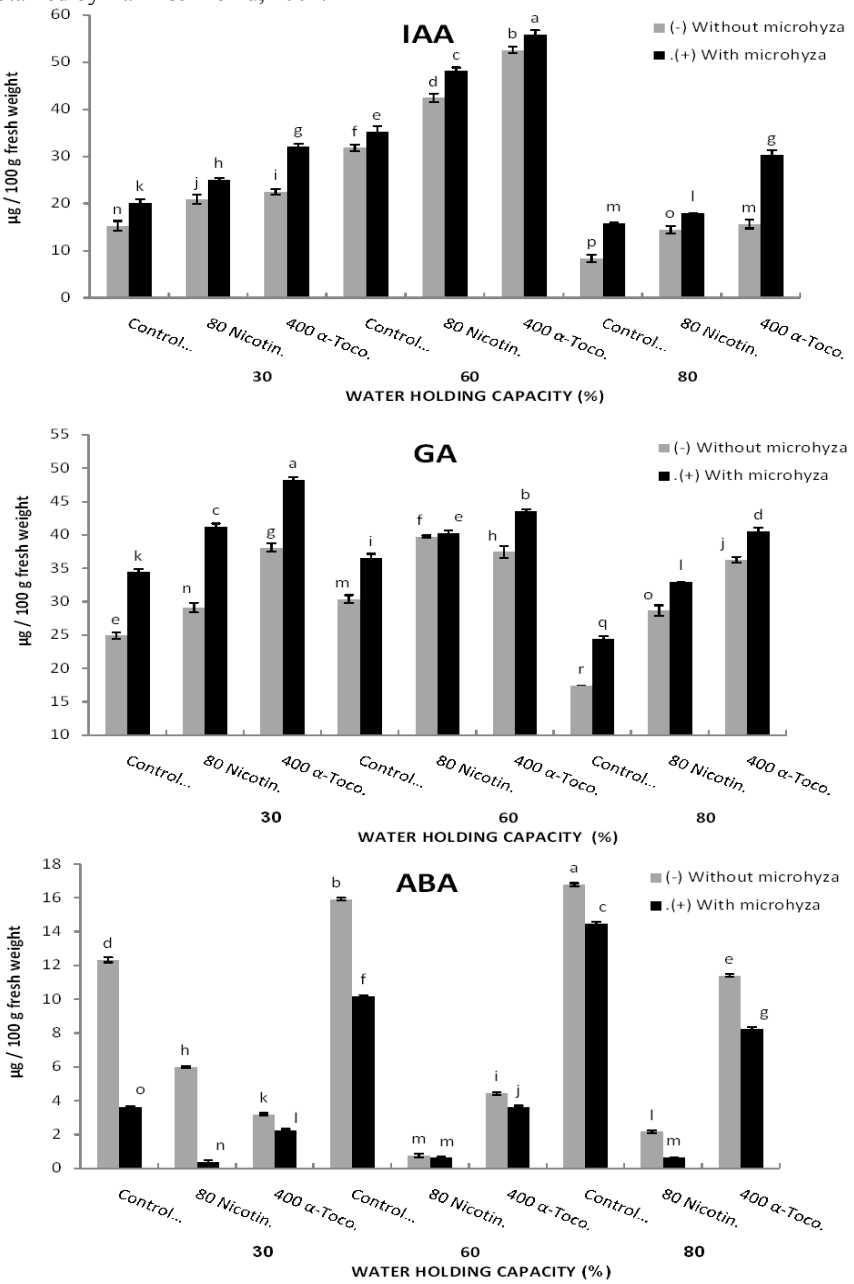


**Fig. 1:** Effet of different concentrations of nicotinamide or α-Tocopherol on endogenous phytohormones of sunflower shoots subjected to different levels of water holding capacity in absence or presence of microhyzza.

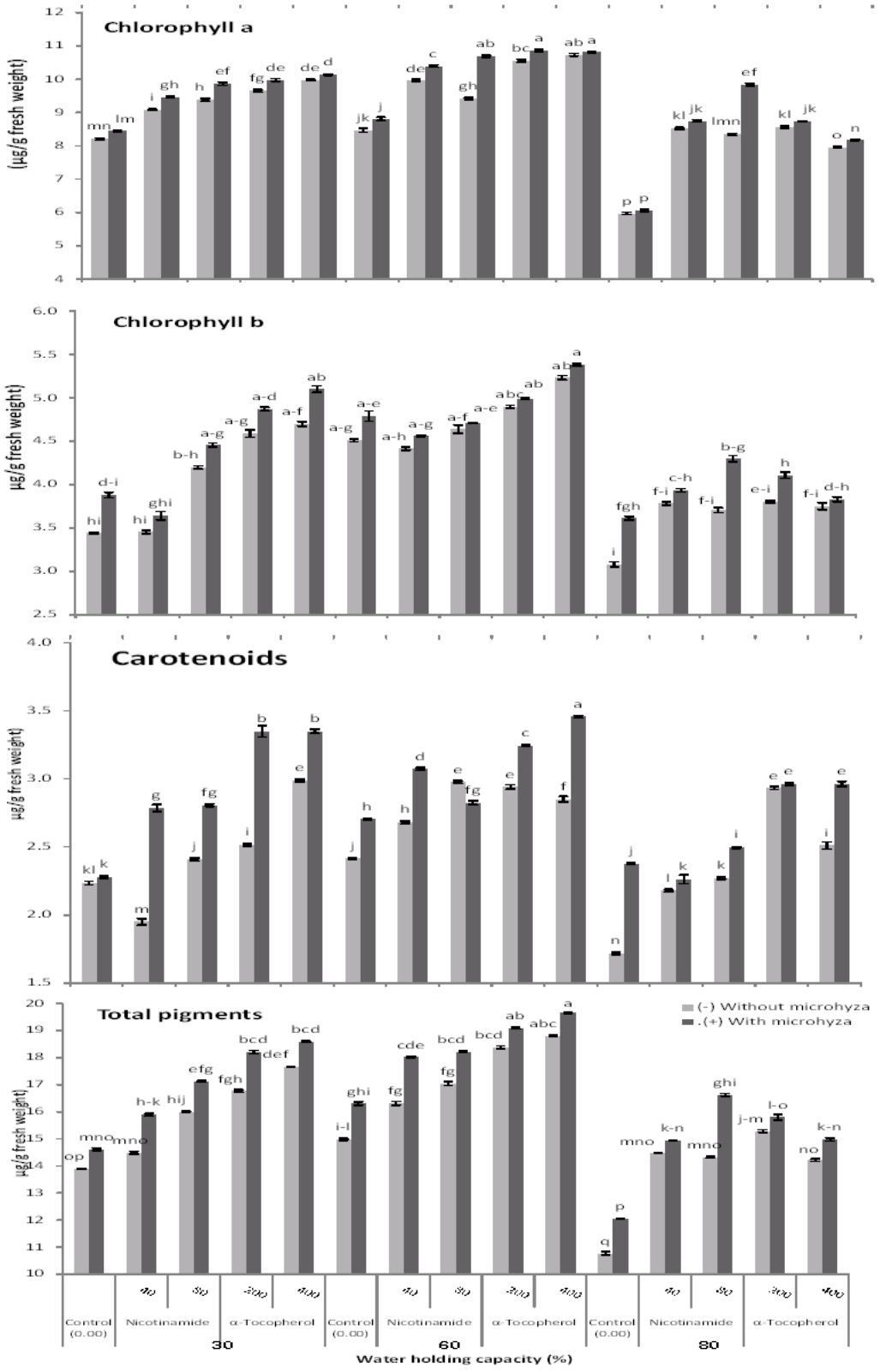


**Fig. 2:** Effect of different concentrations of nicotinamide or α-Tocopherol on photosynthetic pigment contents of sunflower shoots subjected to different levels of water holding capacity in absence or presence of microhyzza.

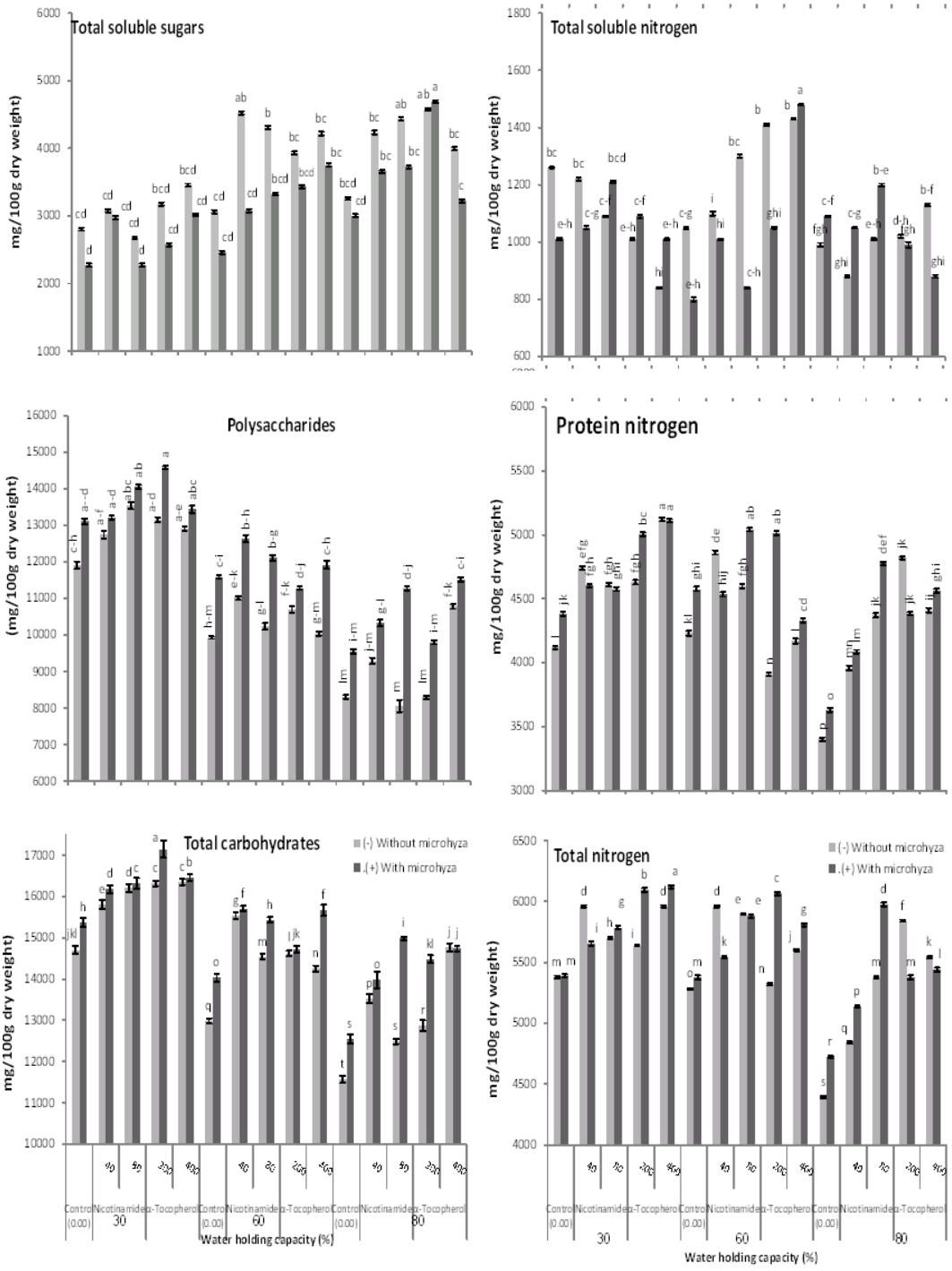


**Fig. 3:** Effect of different concentrations of nicotinamide or α-Tocopherol on photosynthetic pigment contents of sunflower shoots subjected to different levels of water holding capacity in absence or presence of microhyzza.

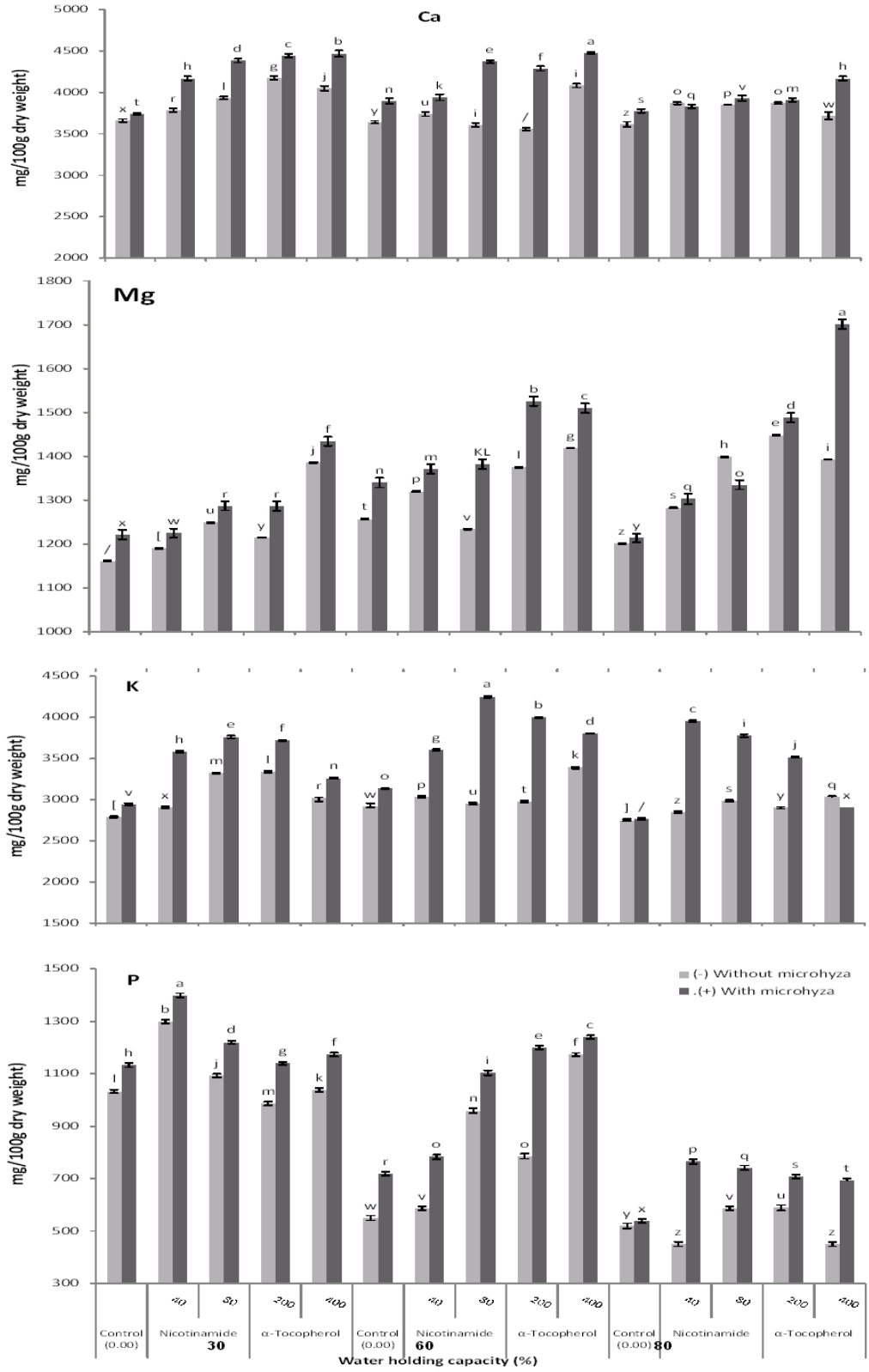


**Fig. 4:** Effect of different concentrations of nicotinamide or α-Tocopherol on minerals content of sunflower shoots subjected to different levels of water holding capacity in absence or presence of microhyzza.

Mycorrhizal plants often have greater tolerance to drought than non mycorrhizal plants. The improved growth, yield and nutrient uptake in sunflower plants reported here demonstrate the potential of mycorrhizal inoculation to reduce the effects of drought stress on sunflower grown under drought stress. Similar results were obtained in tomato grown under field conditions in semiarid areas of the world Al-Karaki*et al* 2004. Mycorrhizal symbiosis has been shown to benefit plants that may suffer from growth inhibition. This inhibition may be due to either chemical or physical soil manipulations at pre-planting stages of agronomical management, shortage in water supply, low quality of irrigation water, high day temperatures with high evapotranspiration and to serve as a biofertilizer enhancing nutrient transfer and bioprotectant activity against environmental stresses, including drought and salinity that can enhance growth and development of crop productivity Bolandnazar *et al*., 2007. The application of AMF inoculums to the soil has been shown to be effective at improving plant growth and enhancing plant resilience to abiotic and biotic stresses Meir*et al.*,2010. Giri *et al*., 2007 reported that mycorrhiza had higher shoot dry weight and fresh yield, than the non-mycorrhizal. This increase in weight can be resulted from the effects of mycorrhiza fungus on absorbing various nutriments such as nitrogen, calcium, potassium, copper, zinc, sulphur and especially better P nutrition Sharifi *et al*., 2007.Using mycorrhiza fungus increases plant growth and affects devoting and transferring nutriments between stem and root so that dry weight of shoot is increased by increasing absorption of nutriment and their transfer. Similar results were also obtained about mycorrhizal plants exposed to osmotic constraints generally perform better than non-mycorrhizal plants Langenfeld-Heyser *et al.,* 2007.On the global scale, osmotic stresses caused by soil salinization and water limitations are the most important factors reducing plant production. Plants and fungi interact naturalistically, while the plant receives mineral nutrients and water through the fungus, the fungus is supplied with carbohydrates by its host Smith and Read, 2008.However, mycorrhization was found to increase the fitness of the host plant by enhancing its growth and biomass. Colla *et al.*(2008 reported that mycorrhiza improved growth, yield, water status, nutrient content and quality of seeds of sunflower plants when exposed to water stress.

Application of α-tocopherol or nicotinamide in the present study improved growth and yield in the sunflower plants by causing significant increases in the values of the above parameters of stressed sunflower (Tables1&2). Similar results were obtained by El–Bassiouny *et al*. 2005 using nicotinamide& α-Tocopherol and Hassanein *et al*. 2009 using nicotinamide .α-tocopherol or nicotinamide may act as growth stimulants which can play a role in mitigating the adverse effect of water stress on metabolic activities relevant to growth through increasing the efficiency of water uptake and utilization as well as protecting the photosynthetic pigments, and the photosynthetic apparatus or increasing IAA content which enhancing cell division and/or cell enlargement. These were further corroborated by the significantly higher levels of carbohydrates observed generally in the vitamins treated plants El-Bassiuony, 2005. The increases in yield and its components might be due to the effect of vitamins on enhancing protein synthesis and delaying senescence El–Bassiouny *et al*., 2005.

***Stomata Opening Area:***

In the present study, leaf stomatal area decreased with drought, which is consistent with the results of sunflower leaves (Table 3). Many studies have shown that water deficit leads to an increase in stomatal density and a decrease in stomatal size Xu and Zhou,2008, indicating this may enhance the adaptation of plant to drought Martinez *et al.,* 2007. An early response to water deficit is a reduction in leaf area and plant growth, which allows plants to reduce their transpiration, thus increasing water use efficiencies (WUE) and promoting interspecies competition capacity underdrought Aguirrezabal *et al*., 2006. Arbuscular mycorrhizal fungi symbiosis contributes to enhance growth and vigor of plants, and can alter plant water relations, particularly during water stress periods Augé, 2001. The arbuscular mycorrhizal (AM) fungi enhance tolerance of plant to water deficit through the alteration of plant physiology Ricardo *et al*., 2008. The mechanisms involved in water uptake by the AMF symbiosis include regulation of stomatal conductance, an increase in stomatal sensitivity to leaf-air vapor pressure deficit, and lowering leaf osmotic potential for turgor maintenance Sánchez-Blanco *et al.,* 2004. In addition to promoting stomatal opening during soil drying and hence carbon gain to lower soil water potential and leaf, AM fungi can also affect stomatal response when soil is lowered osmotically Augé *et al*. 1992. Alpha-tocopherol or nicotinamide application to sunflower plants significantly increase stomatal opening.The increases in stomatal openingmight be due to the effect of vitamins on decreasing the ABA in sunflower shoot Hassanein *et al*., 2009.

***Endogenous Phytohormones:***

Results of the present work (Figure 1) show that decrease in the water holding capacity of the soil caused marked significant decreases in both GA and IAA and increased ABA content, as compared withthose of the control. Similar results were obtained by Liu *et al*. 2000.As soil dries out and soil water potential becomes more negative, plants must activate their defense system to be sure to absorb water as much as they can. It is not known the exact mechanism that defense system is activated. But plant hormone is thought to be an important factor. During drought, Duan *et al*. 1996 found concentrations of abscisic acid (ABA) in xylem sap were lower

in VAM than in NM plants, which was also proved by Liu *et al*. 2000 found the inoculation of VA fungi significantly increased the contents of zeatin, indole acetic acid (IAA) and gibberellin (GA), reduced ABA levels in leaves and roots of corn and cotton under drought conditions. There was a correlation between ABA contents and stomatal resistance. It was suggested that endogenous hormone balance changed by VA fungi colonization contributed to the enhancement of plant drought resistance.

It has been documented that mycorrhization can alter the ABA levels in the host plant Estrada-Luna and Davies, 2003. Jahromi *et al.*(2008 reported lower ABA levels in lettuce plants colonized by Glomus intraradices than in the non-AM plants, indicating that AM plants are less strained by imposed water stress than non-AM plants and, hence, accumulated less ABA.AMF can also benefit plants by stimulating the production of growth regulating substances, increasing photosynthesis, improving osmotic adjustment under drought and salinity stresses and increasing resistance to pests and soil borne diseases Al-Karaki, 2006. These benefits are mainly attributed to improved phosphorous nutrition Plenchette *et al.,* 2005. In view of the results obtained from this study, foliar application of α -tocopherol and nicotinamide counteracted water stress induced decline in the concentration of IAA& $GA_3$ in sunflower shoots. Both chemicals reduced the accumulation of ABA as compared with the corresponding untreated plants (Figure 1). Kodandaramaiah, and Gopala Rao, 1985.The increase in IAA and GA contents in shoot tissues treated with nicotinamide or α -tocopherol concurrent with the increase in growth rate (Table1) indicates the role of the endogenous hormones in stimulation the cell division and/or cell enlargement and subsequently growth Taiz and Zeiger, 1998. Previous published results indicate that exogenous application of nicotinamide leads to IAA and $GA_3$ accumulation and ABA reduction, which was responsible for changes in growth, development and yield of *Zea mays*plant Hassanein *et al* ., 2009.

***Photosynthetic Pigments:***

Water stress may reduce leaf net photosynthetic assimilation by both stomatal and metabolic limitations Ripley *et al*., 2007. In addition, many studies have reported that stomatal effects are major under moderate stresses, but biochemical limitations are quantitatively important during leaf ageing or during severe drought Sheng *et al.*, 2008. Increasing drought causes a reduction in chlorophyll contents (Fig. 1). These results are in good harmony with those obtained by Mittler 2002 who suggested that, the source of reducing energy for ROS scavenging during stress accompanied by suppression of photosynthetic apparatus. In addition,Murkute *et al*., 2006 attributed the reduction in chlorophyll content to the suppression of specific enzymes that are responsible for the synthesis of photosynthetic pigments. The reduction in chlorophyll contents in response to drought stress are mainly due to the stomatal closure due to ABA increased El-Basssiouny 1997. Huixing 2005 VAM plants often display higher rate of photosynthesis than NM counterparts do, which is consistent with VAM effects on stomatal opening. Most of the researchers suggested that VAM symbiosis increased the units of photosynthesis, and so as to increase the rates of photosynthetic storage and export at the same time Augé, 2001. It has been proved that concentration of chlorophyll in VAM plants was higher than their control NM plants Mathur and Vyas, 1995. Morover,the increases in chlorophyll contents as a result of Mycorrhiza ,treatments concomitantly with increasing in Mg levels (Figure 4) in differently WHC levels could be attributed to the role of Mg as an essential for chlorophyll synthesis and hence improving photosynthetic efficiency and plant growth, similar results were reported by Giri *et al.* 2003.

Alpha-tocopherol or nicotinamide application to sunflower plants significantly increased chlorophyll a and chorophyll b content, with no significant change in carotenoid level under low WHC environment. These results are in agreement with those obtained by El–Bassiouny *et al*., 2005 on faba bean. Alpha- tocopherol or nicotinamide may interfere with the protection of chloroplast and their membrane against water stress and the maintaining their integrity Hassanein *et al*., 2009 or vitamins protect chloroplast from oxidative damage Munne–Bosch *et al*., 2001 Also, nicotinamide has a role in activation of enzymes that regulate photosynthetic carbon reduction Taylor *et al*., 1982.

***Total Sugars:***

The increase in total carbohydrates is positively correlated with mycorrhiza of the host plant as reported by Thomson *et al.* 1990. Porcel and Ruiz-Lozano 2004 reported that the positive correlation between sugar content and mycorrhiza is due to the sink effect of the fungus demanding sugars from the shoot tissues Augé, 2000. The processes involved in the development of mycorrhiza frequently lead to increased rates of photosynthesis and of carbon compounds to the root systems of host plants Finlay and Söderström, 1992. The increased sugar accumulation may also be due to hydrolysis of starch to sugars in the seedlings inoculated with mycorrhiza Nemec, 1981. Feng *et al*. 2002 studied the prevalence of correlation between P concentration and sugar accumulation in host plants under saline conditions. Conversely, some authors reported negative correlations between AMF colonization and sugar accumulation in host plants.Sharifi *et al.* 2007 observed that co-inoculated plants had the highest total carbohydrates (%). The favourable effect of co-inoculation may be attributed to hydrolysis of starch to sugars in the co-inoculated plants. In addition, favourably adjusting the osmotic balance and increasing the contents of chlorophylls increases the rate of photosynthesis and carbohydrate synthesis

Swaefy *et al.,* 2007. Alpha-tocopherol or nicotinamide application generally stimulated the accumulation of total carbohydrates contents in drought affected sunflower plant (Figure 3). This is either via increasing endogenous levels of phytohormones (Figure 2) or by acting as activators of carbohydrates synthesis Hassanein *et al*., 2009 on maize. Moreover, accumulation of carbohydrate play a key role in alleviating the water stress, either via osmotic adjustment Ackerson, 1985 or by conferring some desiccation resistance to plant cells Srivastava,. *et al*,1995.

***Total Nitrogen:***

Frechill *et al.*, 2001 reported that water stress interferes with nitrogen (N) acquisition and utilization by influencing different stages of N metabolism, such as, NO uptake and reduction and protein synthesis and the application of AMF can help in better assimilation of nitrogen in the host plant. Furthermore, nicotinamide or α-tocopherol application generally stimulated the accumulation of total nitrogen contents in drought affected sunflower plant (Figure 3). It could be concluded that the inhibitory effect of water stress on the sunflower plant was alleviated by treatment of vitamins through enhancing the biosynthesis of free amino acids and their incorporation into protein. These results added support to the results obtained by Bassouny *et al*., 2008. Thus, it can be concluded that vitamins treatments not only alleviated the inhibitory effect of water stress, via osmotic adjustment or by conferring some desiccation resistance to plant cell, but also stimulated the accumulation of nitrogen constituents over those in the control plants. Moreover, vitamins might act as activators of protein synthesis via significant alteration in the enzymes related to protein metabolism Kodandaramaiah, 1983.

***Inorganic Solute Composition:***

AMF have been shown to have a positive influence on the composition of mineral nutrients (especially poor mobility nutrients such as phosphorus) of plants grown in water-stress conditions Al-Karaki *et al*., 2004 by enhancing and/or selective uptake of nutrients. This is primarily regulated by the supply of nutrients to the root system Giri and Mukerji, 2004 and increased transport (absorption and/or translocation) by AMF (Sharifi *et al*., 2007).

However, P, K Caand Mg% were significantly higher in co-inoculated plants at all WHC levels compared to un-inoculated plants. Increased nutrients uptake in co-inoculated plants may be due to in N metabolism brought about by changes in the enzymes associated with N metabolism, enhancing its uptake facilitated by the extensive hyphae of the fungus which allows them to explore more soil volume than the non-inoculated plants. Co inoculation strongly affects Ca in the plants. Moreover, high Ca was also found to enhance colonizationand sporulation of AMF Shokri and Maadi,2009. Co-inoculated (AMF+R) stressed plants were able to maintain a higher osmotic potential of cells leading to the significantly rapid growth, enhanced nodulation parameters, N, P, K, Ca, total carbohydrates percentages and chlorophyll contents as well as proline in leaves, and significantly reduced the Na percentage (Soliman*et al.,*2012).Potassium plays a key role in plant metabolism. It activates a range of enzymes, and plays a key role in plant water stress and has been found to be the cationic solute which is responsible for stomatal movement in response to changes in bulk leaf water status. Accordingly, the response of AM-infected plants to stress and $K^+$ content are closely related. The adjustment of leaf osmotic potentials requires intracellular osmotic balance Naidoo,1986.

Lee and George 2005 showed that mycorrhizal hyphae of *G. mosseae* had a significant contribution in the uptake of P, Zn and Cu by inoculated cucumber plants resulting in a increased concentration of those nutrients in the plant shoots. Wang *et al*. 2008 also showed an increased N, P, Cu and Zn uptake in inoculated cucumber plants. There is a great correlation between nutritional status of plant and its drought resistance, while VAM changed the nutritional status of its host plant. P concentrations themselves may affect host water balance, but it is often fixed in soil and not available to plant. Phosphatase produced by VA fungi play an important role in translating fixed or insoluble into soluble P, which can be used by plant freely. At the same time, hyphae are also important ways of P transported in soil. The absorption of Ca, Si, Ni, Co etc. was also reported increased by VAM symbiosis Gong *et al* 2000. It is still accepted by many that VAM enhances resistance of high stress of host plants by improving their nutritional status. Enhances the absorption of P and other nutritional elements, and then improves nutritional status of host plant Huixing, 2005.

Foliar spray of nicotinamide or α -tocopherol to sunflower plants under the various levels of WHC and in the co-inoculated plants and un-inoculated plants of mycorrhiza caused an increase in the $K^+$, $Ca^{2+}$, $Mg^{2+}$ and P contents as compared with the corresponding WHC levels (Figure 4). El- Bassiouny *et al*. (2005) also report an increased level of P, $K^+$, $Ca^{2+}$ and $Mg^{2+}$ in response to nicotinamide and α -tocopherol application in faba bean plants. Vitamins led to increase in the contents of ions in the main organs of the stressed sunflower plant through their role in increasing osmotolerance and/or through regulating various processes including absorption of nutrients from soil solution Buschmann and Lichtenthaler, 1979.

***Conclusion:***

The experiment has indicated that *arbuscular mycorrhiza* was able to alter water relation of its host sunflower plants. This paper summarizes effects of *mycorrhiza* on morphology, metabolism and protective adaptation of host plants in the condition of drought stress. Mechanism that *mycorrhiza* can enhance resistance of drought stress in host plant may include many possible aspects: *mycorrhiza* improves its efficiency of water absorption; *mycorrhiza* enhances the absorption of P and other nutritional elements, and then improves nutritional status of host plant; activates defense system of host plant quickly; they can increase the resistance of plants to environmental stresses by stimulating growth regulators level which may be induced a potent effect in regulating the stomatal aperture in plants under different levels of field capacity when compared to plants grown without mycorrhiza and involved in protecting the photosynthetic apparatus and consequently increasing the photosynthetic pigments and the photosynthetic machinery and thereby increasing the carbohydrate , nitrogen contents and the growth rate. The most effect treatment was observed when cultivation of sunflower plant in the presence of mycorrhiza with nicotinamide or α-tocopherol particularly in the presence of 60% water holding capacity.

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