Pak. J. Bot., 51(3): 809-815, 2019. DOI: 10.30848/PJB2019-3(4)

ROLE OF POTASSIUM, ZINC AND GIBBERELLIC ACID IN INCREASING DROUGHT STRESS TOLERANCE IN SUNFLOWER (*HELIANTHUS ANNUUS* L.)

AMIN ULLAH JAN[1, 2*], FAZAL HADI[2], FAZAL AKBAR[3] AND ABDULLAH SHAH[1]

[1]*Department of Biotechnology, Faculty of Science, Shaheed Benazir Bhutto University Sheringal Dir Upper, Pakistan*
[2]*Department of Biotechnology, Faculty of Biological Science, University of Malakand, KPK, Dir Lower, Pakistan*
[3]*Center for Biotechnology and Microbiology, University of Swat, KPK, Pakistan*
**Corresponding author's email: aminjan@sbbu.edu.pk*

Abstract

Nutrients and plant hormone are very essential and play a key role in drought stress tolerance. The current studies was conducted in pots, to investigate the role of potassium, zinc and gibberellic acid in biosynthesis of free proline, total phenolic, antioxidant compounds and drought stress tolerance in sunflower varieties. Plants were grown in glass house and foliar sprays of potassium Nitrate (KNO_3), zinc sulphate ($ZnSO_4$) and gibberellic acid GA_3 were applied. Varieties were evaluated for drought tolerance through leaf disc assay and polyethylene glycol PEG6000 was used to induce in-vitro drought stress. Zinc significantly increased the RWC% and maximum value was noted in *HS-K6* followed by *Rising Sun*. Potassium increased the relative water content (RWC %) and the highest RWC % was found in *Hysun-33* followed by *Rising Sun*, *SMH-0907* and *US-444*. The biosynthesis of free Proline and total phenolic contents was significantly increased with Zn and K treatments. Superoxide dismutase, catalase, ascorbate peroxidase and ascorbic acid biosynthesis were significantly increased by Zn followed by potassium. The Zn treatment significantly reduced the MDA content in sunflower varieties. A significant positive correlation was found between RWC % with proline ($R^2 = 0.92^{***}$) and total phenolic ($R^2 = 0.95^{***}$) respectively. Antioxidant compounds also showed significant positive correlation with relative water content. Conclusively; drought tolerance in sunflower might be increased by foliar application of Zn, K and GA_3. Further investigation is suggested to study the effect of Zn, K and GA_3 on drought tolerant genes expression.

Key words: PEG6000, Membrane damage, Antioxidant.

Introduction

Sunflower (*Helianthus annuus* L.) is the most important oilseed crop in the world and belongs to family Asteraceae. Due to its significant share in vegetable oil production of Pakistan emerged as an economically important crop of the country. But its sensitivity to water stress during the growing season constraints its seed yield with significant reductions. Water availability is one of the warning factors for plant survival in natural ecosystems. Water scarcity is one of the major yield-limiting factors for major crops and significantly reduces the plant growth (Zhang *et al.,* 2014; Qamar *et al*., 2018). Mineral nutrients play important role in plant drought stress. Potassium is one of the most important macronutrients, motivates growth of plants, decreases transpiration and increases the maintenance of water in plants (Umar & Moinuddin, 2002). It also sustains the turgidity, relative water content and osmotic potential of the cell and regulates the opening and closing of stomata under drought condition (Aslam *et al.,* 2014). Micronutrient such as Zn can minimize the adverse effects of water deficiency and improve the production of growth regulator in plants. Its application diminishes the activity of membrane-bound NADPH oxidase, which may lead to decrease the production of reactive oxygen species. Certain plant growth regulator, particularly gibberellic acid promote phloem loading, increases the amount of sucrose phosphate synthase and fructose-1, 6-biphosphatase and inhibit oxidative and heat stresses in plant (Iqbal *et al.,* 2011). GA3 may lead to increase the level of Salicylic acid and plays a vital role in abiotic stress tolerance in plants (Khan *et al.,* 2002). Proline is a free amino acid and frequently accumulates during stress condition and act as osmoregulator (Szabados, 2009). Dehydration stress, promotes the production of reactive oxygen species (ROS), which can be detrimental to proteins, lipids, carbohydrates, and nucleic acids. Phenolic compound work as an antioxidant and minimizes the adverse effect of ROS (Michael *et al.,* 2014).The aim of the present research work was to investigate the role of potassium, zinc and gibberellic acid on proline, phenolic production and drought tolerance in sunflower varieties.

Materials and Methods

Plant material and treatments: Seeds of sunflower (vars. *Rising Sun, SMH 0907, Ausigold-7, SMH-0939, US-444, Hysun-33, SMH-0917* and *HS-K6)* were collected from National Agricultural Research Center (NARC) Islamabad. Five seeds of each variety were sown per pot containing fertile soil and 10 pots/replicates were used for each treatment and control. All pots were put in the glass house at 22°C, 16 h photoperiod 180 mmol m^2 s^1 light intensity and 52 % relative humidity. After seeds germination thinning were made, single healthy seedling per pot was kept using completely randomize design. Different treatments of Potassium (50 ppm of KNO_3), Zinc (30 ppm of $ZnSO_4$) and Gibberellic acid (50 ppm of GA_3) were made as (C) Control without treatment, (T_1) Potassium foliar spray, (T_2) Zinc foliar spray and (T_3) Gibberellic acid foliar spray. When plant reached three to four leaf stage, three doses of each treatment were applied to the aerial parts of the plants at the interval of 7 days. During treatments, soil was completely covered with polyethylene sheet, to avoid soil contamination. For each treatment fresh solution was prepared. After one day of each treatment, varieties were analyzed for physiological and biochemical tests.

Drought stress assessment: Drought tolerance evaluation of sunflower varieties was carried out by leaf disc assays. Aqueous solutions of polyethylene glycol (PEG6000) i.e., 0%, 1%, 3% and 6% were made, added with Murashige and Skoog media (4 g. L^{-1}) designated as T0, T1, T2, and T3 respectively. The media pH was adjusted to 5.8 and autoclaved. Fully expanded upper leaves from each varieties were collected from the green-house and surface was sterilized in 70% ethanol for a few seconds and then in 10% bleach for 10 minutes followed by three rinses with sterile distilled water. Leaf discs were cut using a 1.0 cm diameter cork borer from the leaf blade areas and leaf discs of each varieties were transferred to sterilized Petri dish containing sterilized distilled water and allowed to stand overnight at room temperature (25 ^{0}C) in order to become turgid. Next day, the turgor weight (TW) of each disc was recorded. Leaf discs were transferred to sterile Petri plates containing test media and incubated for five days at room. Then the weight of each disc was noted as fresh weight (FW). Leaf disc was then oven dried at 70^{o}C for two hours and dry weight (DW) of each disc was recorded. Percent relative water content (RWC) for each disc was calculated by using the following formula:

$$RWC\ \% = (FW\text{-}DW) / (TW - DW) \times 100$$

Estimation of proline: Proline estimation was carried out following the method of Bates *et al*., (1973). Fresh mature leaves were collected from experimental plants and properly washed with sterile distilled water. Samples (200 mg) were taken and homogenised in 3% sulfosalicylic acid. Centrifuged at 13,000 rpm for 5 min, 300 ml of the supernatant was transferred to each test tube. Glacial acetic acid (Sigma Aldrich) and acid ninhydrin (Sigma Aldrich) were added to each test tube. The reaction mixture was boiled at 100^{o}C for 1 h in water bath and then test tubes were immediately dipped in ice to stop the reaction. One ml toluene (Sigma Aldrich) was added to the reaction mixture and was shaked briskly for 30 second. The chromophore containing toluene was pipetted and the absorbance was read at 520 nm by spectrophotometer (Shimadzu UV-1700) using toluene for a blank. Three replicates were used for each sample.

Estimation of total phenolic: The total phenolic content in sunflower was determined according to the method of Malik & Singh (1980). Samples (200 mg) were ground into powder and transferred into eppendorf tubes. Methanol (2 ml) was added to each sample and kept for four hours. Centrifuged for 6 minutes at 5000 rpm. The supernatant was transferred into fresh tubes. Methanol extract (0.5 ml) was mixed with 2.5 ml of 10% of Folin–Ciocalteu reagent (Sigma) and kept for 5 minutes. Added 2.5 ml (7.5% $NaHCO_3$) and 2.5 ml distilled water and kept in dark for 1 hour. Blue color developed, then the absorbance was measured at 650 nm by spectrophotometer (Shimadzu UV-1700). The concentrations of total phenolic contents in samples were calculated from the calibration plot and expressed as mg galic acid equivalent of phenol/g of sample.

Estimation of antioxidant compounds: Fresh sunflower leaves were collected and homogenized in extraction buffer (5 mL) containing 50 mM sodium phosphate (pH 7.8). The samples were centrifuged and the supernatant was applied to measure the activities of catalase (CAT; EC 1.11.1.6) and superoxide dismutase (SOD; EC 1.15.1.1) following the methods of Wu *et al.,* (2014). The fresh sunflower leaves were homogenized by (5 mL) 50 mM sodium phosphate (pH 5.5) containing 0.2 mM EDTA-Na2. The supernatant was applied to measure the activity of ascorbate peroxidase (APX; EC 1.11.1.11) according to the description of Wu *et al.,* (2014). The ascorbic acid in sunflower was measured applying the descriptions of Li *et al.,* (2000). The measurement of reduced glutathione applied the description of Wu *et al.,* (2014).

Measurement of membrane damage: The contents of malonaldehyde (MDA) were determined following the method of Quan *et al*., (2004). Crown tissue (0.5 g) was homogenized in 3 mL of 10% trichloroacetic acid (TCA) using a pestle and mortar. The homogenates were then centrifuged at 8000g for 10 min. A 1 mL aliquot of the supernatant was incubated with 1 mL of 0.5% 2-thiobarbituric acid (TBA) in 10% TCA in boiling water for 15 min. After centrifugation at 8000g for 10 min, the absorbance of the supernatant was read at 450, 532, and 600 nm respectively.

Results and Discussion

Effect of treatments on relative water contents (%): The RWC % was reduced while increasing PEG concentration showing the dehydration effect of the treatments and there were differences among the varieties. Different treatments (KNO_3, $ZnSO_4$ and GA_3) significantly increased relative water contents in sunflower varieties as compared to control C (Fig. 1). In control plants (without treatments), Rising Sun, SMH-0907 and HS-K6 showed less than 50% relative water content (Fig. 1A). The treatments of potassium and zinc increased up to eight times more RWC % in these varieties. In T_1 treatment (50 ppm of KNO_3), the highest RWC % was noted in Hysun-33 followed by Rising Sun while in T3 treatment (30 ppm of $ZnSO_4$), the highest RWC % was found in the SMH-0917 followed by HS-K6 Fig. 1(B and C). The overall effect of treatments (K, Zn and GA3) on RWC %, $ZnSO_4$ showed high effect on RWC % followed by K and GA3 as compared to Control C i.e. Zn > K > GA_3 > Control C Fig. 1(A-D). The application of K enhanced relative water content (RWC %) under stress conditions (Zhang *et al.,* 2009). It increased the water use efficiency and lowered the rate of transpiration (Maria *et al.,* 2008). Potassium plays a key role in the maintaining of cell turgor and growth of the plant in lack of the water (Umar & Moinuddin 2002). It also helps in retaining equilibrium between osmotic potential of the cell and its surroundings (Mujtaba *et al.,* 2007). The appropriate potassium supply to the plants increased root growth which consequently enhanced the water uptake from the soil. The plants which received a proper amount of Zn had more relatively more water content (Shahri *et al.,* 2012). It was very useful for the growth and development of the plants and its application increased the auxin level which may lead to enhanced root growth consequently improved the drought tolerance (Waraich *et al.,* 2011).

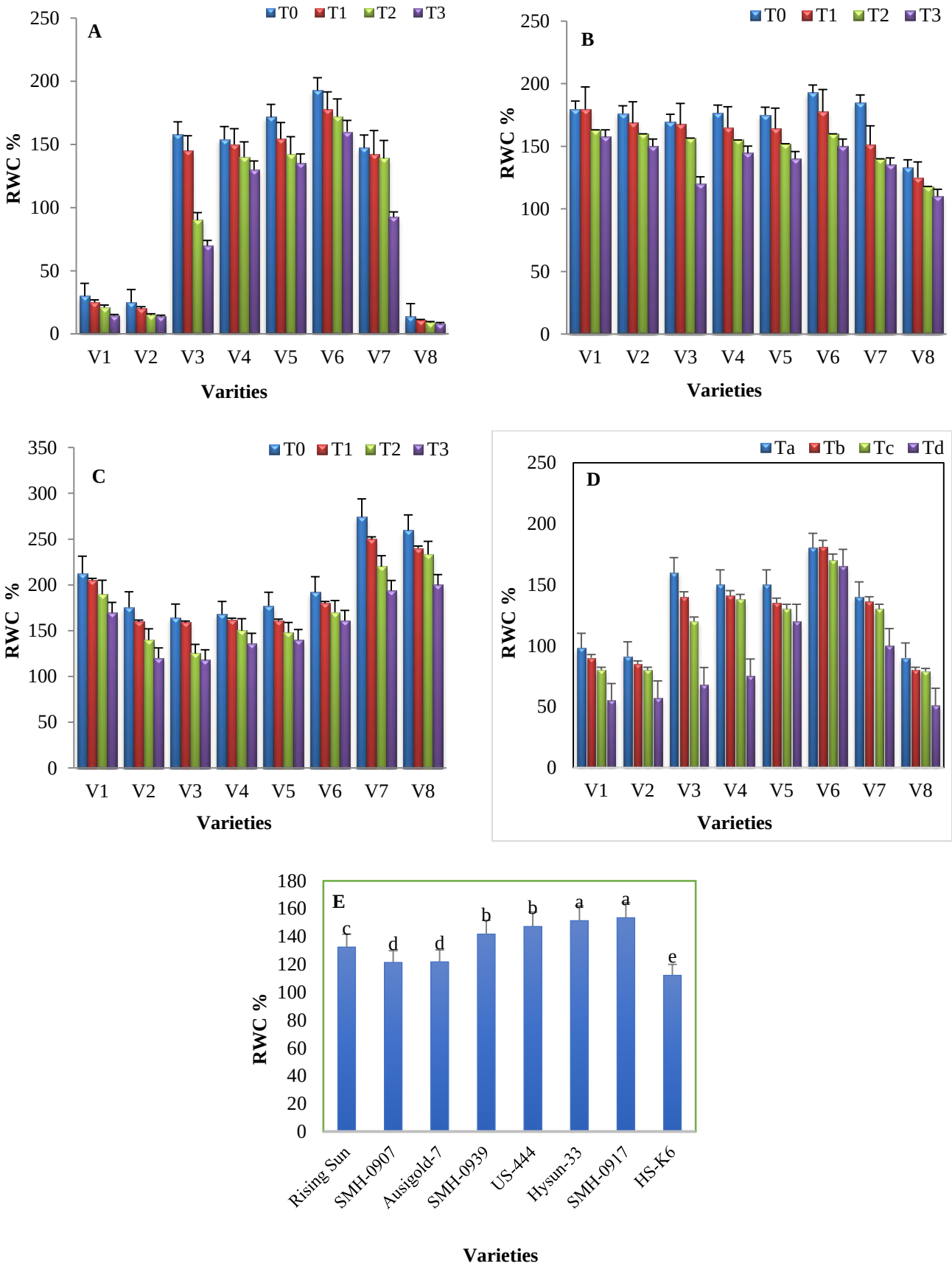

Fig. 1. (A-E): Effect of different treatments (K, Zn and GA3) on the relative water content (RWC %) in leaf discs of sunflower varieties at different concentration of polyethylene glycol in test solutions, T0 = control without polyethylene glycol, T1 = 1% PEG, T2 = 3 % PEG and T3 = 6 % PEG. (A) Control plants without treatment (B) potassium foliar spray (C) Zinc foliar spray (D) GA3 foliar spray (E) Mean of RWC % in all treatments. Alphabets show the significant differences and bars show standard error. Where: V1 = Rising Sun, V2= SMH-0907, V3= Ausigold-7, V4 = SMH-0939, V5 = US-444, V6 = Hysun-33, V7= SMH-0917 and V8= HS-K6

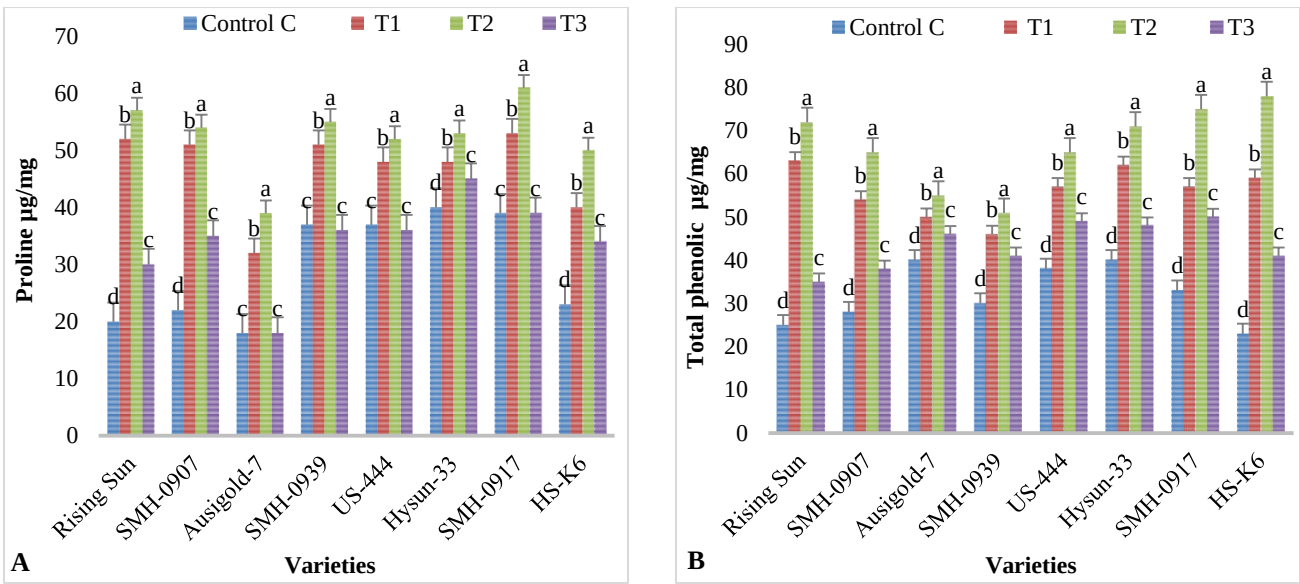

Fig. 2. Effect of different treatments (KNO_3, $ZnSO_4$ and GA_3) on free proline (A) and total phenolic contents (B). Control C (without treatment), T1 treatment (50 ppm of KNO_3), T2 treatment (30 ppm of $ZnSO_4$) and T3 treatment (50 ppm of GA_3). Alphabets show the significant differences and bars show standard error.

Effect of treatments on biosynthesis of free proline: Different treatments (KNO_3, $ZnSO_4$ and GA_3) showed significant ($p \leq 0.05$) effect on biosynthesis of free proline as compared to control C (Fig. 2A). Potassium treatments enhanced synthesis of free proline in plant as compared to control C. The highest free proline was found in SMH-0917 while lowest proline was observed in *Ausigold-7*. Proline works as a buffer agent, cell osmotic potential, and detoxify the toxic ions (Ashraf & Foolad, 2007). During stress condition proline synthesis are increased which provide protection to the macromolecules of the cell. These findings are in line with the work of (Zadehbagheri *et al.*, 2012). In T_2 treatment (30 ppm of $ZnSO_4$) the highest proline was observed in *SMH-0917* while lowest proline level was noted in *Ausigold-7*. Zinc is essential micro-element and work as cofactor of several enzymes. It is a structural component of aldolase and carbonic anhydrase, which are involved in plant carbon fixation. The reactive oxygen species are produced during stress condition. Zinc may act as a scavenger of ROS and provide protection to the cells. The exogenous application of gibberellic acid elevated the biosynthesis of proline in sunflower as compared to control plants (Fig. 2A). Our results are an agreement with the findings of (Naz & Bano, 2012).

Effect of treatments on total phenolic contents: The exogenous application of KNO_3, $ZnSO_4$ and GA_3 significantly ($p \leq 0.05$) increased biosynthesis of total phenolic content in sunflower varieties as compared to control plants (Fig. 2B). In T_1 treatment (50 ppm of KNO_3), the highest total phenolic content was found in *Rising Sun* while lowest total phenolic content was noted in *Ausigold-7*. It significantly diminishes the production of ROS and boost the activities of antioxidant enzymes, i.e., superoxide dismutase, catalase and peroxidase (Umar & Moinuddin, 2002). In T_2 treatment (30 ppm of $ZnSO_4$), the highest total phenolic was found in *HS-K6* while lowest total phenolics was observed in *SMH-0939*. Zinc plays vital role in retaining the bio-membrane structure and for detoxifying of reactive oxygen species. Zinc application increases total phenolic content in plants (Tavallali *et al.*, 2010). GA_3 foliar spray slightly increased the total phenolic contents as compared to control plants (Fig. 2B). Our results were in agreement with the finding who reported GA_3 increased specific phenolic compounds in plants (Anastasia *et al.*, 2012).

Effect of treatments on biosynthesis of anti-oxidant compounds: Different treatments of KNO_3, $ZnSO_4$ and GA_3 significantly increased the biosynthesis of anti-oxidant compounds in sunflower as compared to control (Fig. 3). The exogenous application of treatments increased the biosynthesis of enzymatic antioxidant compounds. The maximum superoxide dismutase (SOD), catalase (CAT), ascorbate peroxidase (APX) was found in the $ZnSO_4$ treatment followed by KNO_3 and GA_3 Fig. 3(A-C). The current findings are in line with the work (Wu *et al.*, 2015). Zn play crucial roles in protecting cytomembranes of plant from impairments caused by ROS under water deficit. The application of zinc sulphate significantly reduced the malonaldehyde (MDA) followed by potassium and gibberellin as compared to control plants (Fig. 3D). These findings are in line with the work of Wu *et al.*, (2015). The biosynthesis of reduced glutathione (GSH) and ascorbic acid (AsA) was significantly increased by zinc sulphate followed by potassium and gibberellin Fig. 3(E and F). GSH and AsA are all non-enzymatic antioxidants in higher plants and are also conducive to alleviate oxidative injuries resulted from water deficit. AsA plays an essential role in removing hydrogen peroxide, and GSH is an antioxidant and redox buffer that scavenges ROS, which has been shown to be involved in the resistance to drought stress in several plants (Thounaojam *et al.*, 2014).

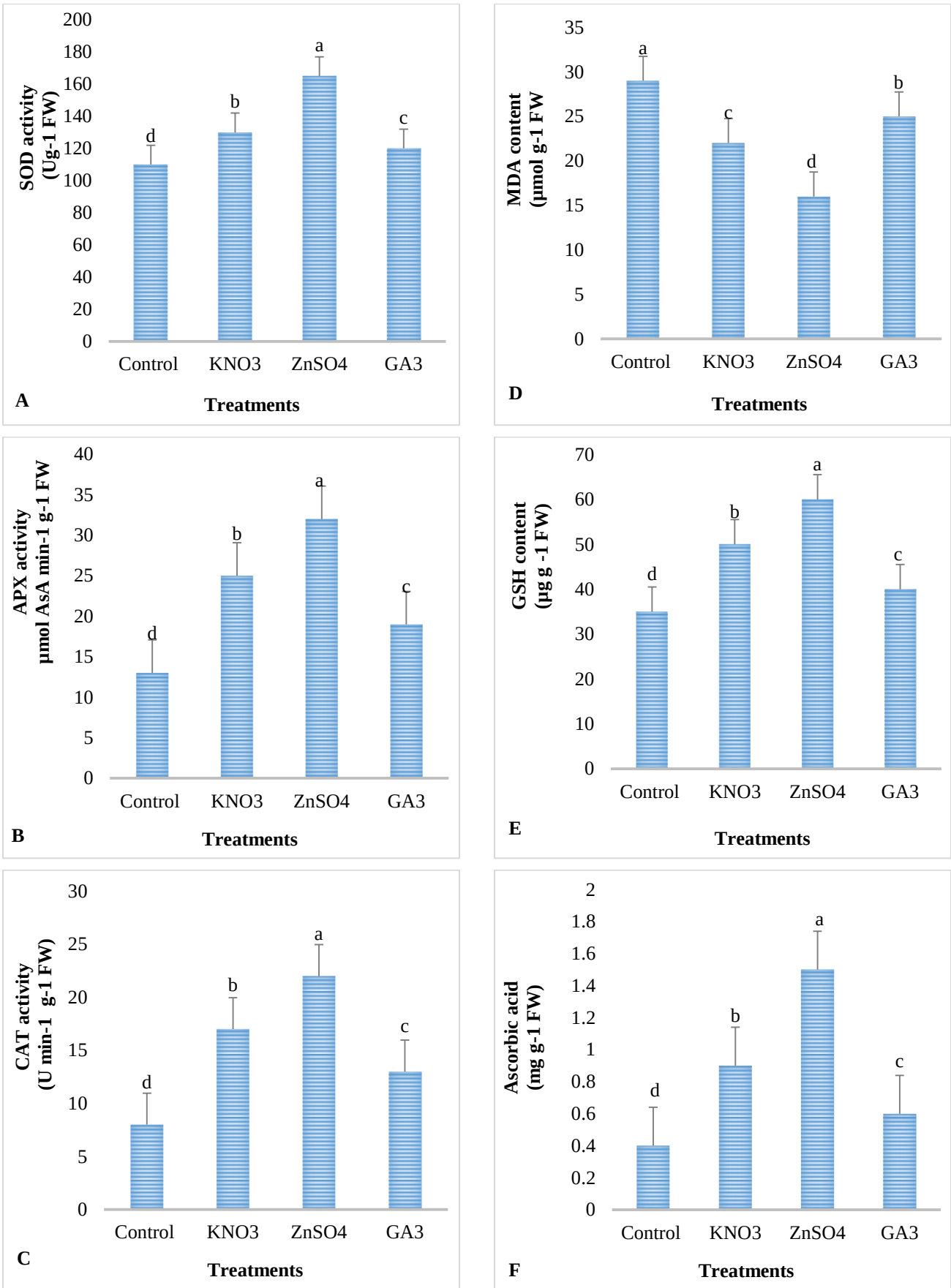

Fig. 3. Effect of different treatments (KNO_3, $ZnSO_4$ and GA_3) on enzymatic and non-enzymatic antioxidant compounds. SOD (A), APX (B), CAT (C), MDA content (D), GSH (E), and Ascorbic acid (F).). Control (without treatment), T1 treatment (50 ppm of KNO_3), T2 treatment (30 ppm of $ZnSO_4$) and T3 treatment (50 ppm of GA_3). Alphabets show the significant differences and bars show standard error.

Table 1. Correlations among different parameters measured in Sunflower varieties.

Parameters	RWC	Superoxide dismutase	Ascorbate peroxidase	Catalase	Malonaldehyde	Ascarbic acid	Proline	Phenolic
RWC	1.00							
Superoxide dismutase	0.95***	1.00						
Ascorbate Peroxidase	0.94***	0.96***	1.00					
Catalase	0.98***	0.95***	1.00	1.00				
Malonaldehyde	-0.94***	-0.84**	-0.96***	-0.97***	1.00			
Ascarbic acid	0.93***	0.99***	0.98***	0.97***	-0.89**	1.00		
Proline	0.93***	0.80**	0.91***	0.90**	-0.94***	0.86**	1.00	
Phenolic	0.95***	0.98***	0.99***	0.99***	-0.93***	0.99***	0.90**	1.00

Correlation of proline, total phenolic and antioxidant compounds with RWC (%): There was a positive correlation between proline and RWC % in sunflower varieties. A positive correlation between proline accumulations and RWC was also observed by (Chutipaijit *et al.,* 2009). Proline has been reported to activate other mechanisms, such as the formations of strong H-bonded water around protein for protecting protein structures and scavenger of free radicals. Highly significant positive correlation was found between total phenolic and relative water content (Table 1). The proline and phenolic play a vital role in the osmotic adjustments which enable plant to maintain turgor pressure during drought condition. Different researchers reported positive correlation between proline level and total phenolic contents with RWC % (Hare & Cress, 1997). The enzymatic antioxidant compounds i.e. superoxide dismutase, ascorbate peroxidase and catalase showed significant positive correlation with relative water content respectively (Table 1). MDA content showed significant negative correlation (R^2 = -0.93) with relative water content. The non-enzymatic antioxidant compound i.e. ascorbic acid showed significant positive correlation (R^2 = 0.94) with relative water content.

References

Anastasia, E., F. Giannakoula, J. Jelena, M. Maksimovic and D.Z. Maksimovic. 2012. The effects of plant growth regulators on growth, yield, and phenolic profile of lentil plants. *J. Food Comp. & Anal*., 28: 46-53.

Ashraf, M. and M.R. Foolad. 2007. Roles of glycine betaine and proline in improving plant abiotic stress resistance. *Envi. Exp. Bot*., 59: 206-216.

Aslam, M., M.S.I. Zamir, I. Afzal and M. Amin. 2014. Role of potassium in physiological functions of spring maize grown under drought stress. *J. Ani. & Pl. Sci*., 24(5): 1452-1465.

Bates, L., R.P. Waldren and I.D Teare. 1973. Rapid determination of free proline for water-stress studies. *Plant Soil*, 39: 205- 207.

Chutipaijit, S., S. Chaum and K. Sompornpailin. 2009. Differential accumulations of proline and flavonoids in indica rice varieties against salinity. *Pak. J. Bot*., 41(5): 2497-2506.

Hare, P.H. and W.D. Cress. 1997. Metabolic implications of stress induced proline accumulation in plants. *Plant Growth Regulator*, 21: 79-102.

Iqbal, N., R. Nazar, M. Iqbal, R. Khan, A. Masood and N.A. Khan. 2011. Role of gibberellins in regulation of source–sink relations under optimal and limiting environmental conditions. *Cur. Sci*., 100(7): 10-14.

Khan, N.A., R. Mir, M. Khan, S. Javid and Samiullah. 2002. Effects of gibberellic acid spray on nitrogen yield efficiency of mustard grown with different nitrogen levels. *Pl. Growth Reg*., 38: 243-247.

Li, H.S., Q. Sun and S.J. Zhao. 2000. Principles and techniques of plant physiological biochemical experiment. Higher Education, Beijing, pp. 138-248.

Malik, C.P. and M.B. Singh. 1980. Extraction and estimation of amino acids and keto acids. In: Plant Enzymology and Histo-enzymology. Kalyani Publishers, New Delhi-Lud Hana, India, p. 286.

Maria, A.M., A.A. Gendy, A.H. Selim and A. Abd-El. 2008. Response of wheat plants grown under water stress in relation to Jasmonic acid. Minufiya. *J. Agri. Res*., 33(6): 1355-1375.

Michael, J.O., S. Leuzinger, D. Christopher, Philipson and A. Hector. 2014. Drought survival of tropical tree seedlings enhanced by non-structural carbohydrate levels. *Nat. Clim. Chang*., 4: 710-714.

Mujtaba, S.M., M.M. Ali, M.Y. Ashraf, B. Khanzada, S.M. Farhan, M.U. Shirazi, M.A. Khan, A. Shereen and S. Mumtaz. 2007. Physiological responses of wheat genotypes under water stress conditions at seedling stage. *Pak. J. Bot*., 39(7): 2575-2579.

Naz, I. and A. Bano. 2012. Assessment of phytohormones producing capacity of stenotrophomonas maltophiliassa and its interaction with *Zea mays*. *Pak. J. Bot*., 44(1): 465-469.

Qamar, R., M. Ghias, F. Hussain, S. Habib, M.K. Razzaq, M. Aslam and I. Habib. 2018. Effect of drought on morpho-physiological traits of sunflower (*Helianthus annuus* L.) hybrids and their parental inbred lines. *Pak. J. Agri. Res*., 31(2): 186-193.

Quan, R., M. Shang, H. Zhang, Y. Zhao and J. Zhang. 2004. Improved chilling tolerance by transformation with betA gene for the enhancement of glycinebetaine synthesis in maize. *Pl. Sci*., 166: 141-149.

Razzaq, H., M.H.N. Tahir, H.A. Sadaqat and B. Sadia. 2017. Screening of sunflower (*Helianthus annus* L.) accessions under drought stress conditions, an experimental assay. *J. Soil Sci. Plant Nutr*., 17(3): 662-667.

Shahri, Z.B., G.R. Zamani and M.H. Sayyari-Zahan. 2012. Effect of drought stress and zinc sulfat on the yield and some physiological characteristics of sunflower. *Adv. Environ. Biol*., 6(2): 518-525.

Szabados, L. and A. Savoure. 2009. Proline: a multifunctional amino acid. *Trends in Pl. Sci*., 15(2): 89-97.

Tavallali, V., M. Rahemi, S. Eshghi, B. Kholdebarin and A. Ramezanian. 2010. Zinc alleviates salt stress and increases antioxidant enzyme activity in the leaves of pistachio seedlings. *Turk. J. Agri. & Forest*, 34: 349-359.

Thounaojam, T.C., P. Panda, S. Choudhury, H.K. Patra and S.K. Panda. 2014. Zinc ameliorates copper-induced oxidative stress in developing rice seedlings. *Protoplasma*, 251: 61-69.

Umar, S. and M. Moinuddin. 2002. The effect of sources and rates of potassium application on potato yield and economic returns. *Better Crop Int*., 15: 13-15.

Waraich, E.A., R. Ahmad, M.Y. Saifullah, Ashraf and Ehsanullah. 2011. Role of mineral nutrition in alleviation of drought stress in plants. *Aust. J. Crop Sci*., 5(6): 764-777.

Wu, S., C. Hu, Q. Tan, L. Li, K. Shi, Y. Zheng and X. Sun. 2015. Drought stress tolerance mediated by zinc-induced antioxidative defense and osmotic adjustment in cotton (*Gossypium hirsutum*). *Acta Physiol. Pl.*, 37: 167.

Wu, S., C. Hu, Q. Tan, Z. Nie and X. Sun. 2014. Effects of molybdenum on water utilization, antioxidative defense system and osmotic-adjustment ability in winter wheat (*Triticum aestivum*) under drought stress. *Pl. Physiol. Biochem*., 83: 365-374.

Zadehbagheri, M., M.M. Kamelmanesh, S. Javanmardi and S. Sharafzadeh. 2012. Effect of drought stress on yield and yield components, relative leaf water content, proline and potassium ion accumulation in different white bean genotypes. *Afr. J. Agri. Res.*, 42: 5661-5670.

Zhang, J., B. Dell, E. Conocono, I. Waters, T. Setter and R. Appels. 2009. Water deficits in wheat: fructan exohydrolase (1-FEH) mRNA expression and relationship to soluble carbohydrate concentrations in two varieties. *New Phytol*., 18: 843-85.

Zhang, L., J. Peng, T.T. Chen, X.H. Zhao, S.P. Zhang, S.D. Liu, H.L. Dong, L. Feng and S.X. Yu. 2014. Effect of drought stress on lipid peroxidation and proline content in cotton roots. *J. Ani & Pl. Sci*., 24(6): 1729-1736.

(Received for publication 7 March 2018)