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Chemical role of α -tocopherol in salt stress mitigation by improvement in morpho-physiological attributes of sunflower (*Helianthus annuus* L.)Irfana Lalarukh^a, Xiukang Wang^{b,*}, Syeda Fasiha Amjad^{c,*}, Rashid Hussain^d, Sunny Ahmar^{e,m}, Freddy Mora-Poblete^f, Shams H. Abdel-Hafez^g, Mustafa A. Fawzy^h, Mohamed H.H. Abbasⁱ, Ahmed A. Abdelhafez^{j,k,*}, Rahul Datta^{l,*}^a Department of Botany, Government College Women University, Faisalabad 38000, Pakistan^b College of Life Sciences, Yan'an University, Yan'an, Shaanxi 716000, PR China^c Department of Botany University of Agriculture Faisalabad, 38000, Pakistan^d Department of Horticultural Sciences, Faculty of Agriculture and Environment, The Islamia University of Bahawalpur, 63100 Punjab, Pakistan^e Institute of Biological Sciences, University of Talca, 1 Poniente 1141, Talca 3460000, Chile^f Institute of Biological Sciences, Campus Talca, Universidad de Talca, Talca 3465548, Chile^g Department of Chemistry, College of Science, Taif University, P.O.Box 11099, Taif 21944, Saudi Arabia^h Biology Department, College of Science, Taif University, P.O.Box 11099, Taif 21944, Saudi Arabiaⁱ Benha University, Faculty of Agriculture, Soils and Water Department, Egypt^j New Valley University, Faculty of Agriculture, Soils and Water Department, Egypt^k National Committee of Soil Science, Academy of Scientific Research and Technology, Egypt^l Department of Geology and Pedology, Faculty of Forestry and Wood Technology, Mendel University in Brno, Zemedelska 1, 61300 Brno, Czech Republic^m Department of Plant Breeding and Genetics, PMAS- Arid Agriculture University, Rawalpindi 46000, Pakistan

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ABSTRACT

Elevated concentrations of salts in soil and water represent abiotic stresses. It considerably restricts plant productivity. However, the use of alpha-tocopherol (α -toc) as foliar can overcome this problem. It can improve crop productivity grown under salinity stress. Limited literature is documented regarding its optimum foliar application on sunflower. That's why the need for the time is to optimize α -toc foliar application rates for sunflower cultivated in salt-affected soil. A pot experiment was performed to select a better α -toc foliar application for mitigation of salt stress in different sunflower cultivars FH (572 and 621). There were 2 levels of salts, i.e., control (no salt stress) and sodium chloride (120 mM) and four α -toc foliar application (0, 100, 200, and 300 mg L⁻¹). Results showed that foliar application of 100 mg/L- α -toc triggered the remarkable increase in fresh shoot weight, fresh root weight, shoot, and root lengths under salinity stress in FH-572 and FH-621 over 0 mg/L- α -toc. Foliar application of 200 mg/L- α -toc was most effective for improvement in chlorophyll *a*, chlorophyll *b*, total chlorophyll and carotenoids compared to 0 mg/L- α -toc. Furthermore, an increase in A was noted in FH-572 (17%) and FH-621 (22%) with α -toc (300 mg L⁻¹) application under saline condition. In conclusion, the 100 and 200 mg/L- α -toc are the best application rates for the improvement in sunflower FH-572 and FH-621 growth, chlorophyll contents and gas exchange attributes. Further investigations are needed to select a better foliar application rate between 100 and 200 mg/L- α -toc at the field level under the different agro-climatic zone and soil types.

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1. Introduction

Salt stress is highly harmful stress among (Ahmed et al., 2020) all abiotic stresses (drought, salinity, heavy metals, nutritional deficiency, imbalance nutrition application) that adversely affect many physiological processes in crops (Abbas et al., 2020; Danish and Zafar-ul-Hye, 2020; Danish et al., 2019; Irfana et al., 2020; Rafiullah et al., 2020; Ullah et al., 2020; Wahid et al., 2020; Yadav et al., 2017; Zafar-ul-Hye et al., 2020, 2018). Its implications on the grown plants arise via inhibition of expansion of young leaves (osmotic phase) and promotion of senescence in mature leaves (ionic phase). Accordingly, plant leaves become thicker and smaller (Zörb et al., 2015). Also, chlorophyll contents decreased considerably in plants, e.g., safflower (Gengmao et al., 2015), linseed (Sh et al., 2014), tomato (Muneer et al., 2014) and cotton (Liu et al., 2014). Moreover, this stress induces the accumulation of abscisic acid (ABA), which, in turn, causes significant reductions in stomatal aperture, stomatal conductance, substomatal CO₂ concentration RuBisCO and hinders the activities of many other enzymes (Chaves et al., 2009). In roots, salt stress promotes lateral root growth while restricting primary roots' growth (Jung and McCouch, 2013). Overall, salt stress leads to metabolic disorders, stunted growth and causes considerable losses in crop productivity. Improving plant growth under such conditions requires adopting appropriate water conservation strategies that control plant transpiration and maintain tissue hydration (Abbasi et al., 2015). Probably, spraying plants with various growth regulators (Yaseen et al., 2021), including vitamins (Lalarukh, 2018), can mitigate salt stress conditions.

Vitamins are the key growth regulators to maintain the proper functioning of plants (Hassanein et al., 2009). Among thirteen essential vitamins needed by living organisms, vitamin E (α -tocopherol) is selected in this study because of its impacts on the detoxification of reactive oxygen species (Mène-Saffrané, 2018); hence, it increases the stability of cell membranes (Hincha, 2008). Its signals the activation of (MAPK) mitogen-activated protein kinases under abiotic stresses, protects photosystem II from photo-inhibition, and assists in phloem loading within plants (Hyun et al., 2011). This enzyme is manufactured in plastids (Hincha, 2008); hence, its insufficient production may subsequently alter plant growth, germination, translocation of photosynthetic products, and leaf senescence (Falk and Munné-Bosch, 2010). Several reports highlighted the beneficial effects of spraying plants with vitamin E (α -toc) on enhancing crop productivity. For example, spraying wheat plants with α -toc raised carbohydrate and protein contents in grains and considerably increased the total yield (Dawood et al., 2014). In mung bean (*Vigna radiate*), spraying this enzyme on plants during the vegetative growth stage raised the activities of (enzymatic) antioxidants, vitamin C, amino acids and protein contents (Sadiq et al., 2016). Foliar spray of onion (*Allium cepa*) with α -toc increased its growth by reducing peroxidation of lipids and hydrogen peroxide production (Semida et al., 2016). Similar results were reported on common bean (*Phaseolus vulgaris*) and soybean (Hemida et al., 2017; Serefioglu et al., 2017). It is, therefore, assumed that foliar application of α -toc could mitigate the harmful effects of salt stress on plants by improving their morpho-physiological features.

Due to the exponential increase in population and demand for food worldwide, much saddle is placed on scientists to introduce new safe techniques for increasing the productivity of crops grown under abiotic stresses (Amjad et al., 2021; Rafeeq et al., n.d.). Sunflower (*Helianthus annuus* L.) is the 4th important vegetable oil in world trade (Fernández-Martínez et al., 2010) and, on the other hand, is considered a moderately salt-tolerant crop (Lalarukh, 2018). It can survive with lesser irrigations and also tolerate salt

stress up to 50 mM (Kumar et al., 2014). Its oil contains a high percentage of polyunsaturated fatty acids with zero cholesterol levels (Anuradha, 2014). Despite the reports that guaranteed growing sunflower in arid and semi-arid regions (Awais et al., 2017), many countries still import edible oils to meet their local market needs. Pakistan, for example, imports about 80% (edible) oil from foreign countries (Keerio et al., 2020), while Egypt is the 7th biggest importer of edible oils worldwide (Mohamed, 2018). It's worth noting that Saudi Arabia is one of the top importers of sunflowers from Ukraine (Kaya et al., 2015).

In arid and semi-arid regions, water scarcity and salinity stresses are major conjugated problems (Garcia-Franco et al., 2021) and the high salinity hazards significantly decrease the productivity of sunflowers (Wang et al., 2017). Accordingly, the current study explores the impact of spraying two sunflower cultivars with vitamin E (α -toc) to alleviate salinity's adverse effect on grown plants. Limited information is documented so far in literature on the exogenous application of α -toc for mitigating salinity stress especially in sunflower for the optimization of foliar application levels. This study will be helpful in covering the knowledge gap regarding the best foliar application rate for the mitigation of salinity stress and improvement in sunflower plant growth, chlorophyll contents and gas exchange attributes. Sunflower was selected in the current study as it is an important economic oil crop and is among the most cultivated plants. It is hypothesized that sunflower plants received foliar application of α -toc might perform better under salt stress compared to untreated.

2. Materials and methods

2.1. Cultivars and location of the experiment

A pot experiment (2016) was conducted in the botanic garden (73° 10'E longitude, 31° 30'N latitude and 213 m altitude) at the University of Agriculture, Faisalabad, Pakistan. Achenes of FH-(572 and 621) sunflower cultivars were acquired from the Oilseed Research Section of Ayub Agricultural Research Institute, Faisalabad, Pakistan. The research was performed from February to June 2016 to study the influence of α -toc foliar application on morpho-physiological attributes of sunflower in salt stress. Surfactant Tween 20 (0.1 %) was used in all levels of α -toc.

2.2. Experimental layout

Soil sampling was done according to standard protocol (Marfo et al., 2019a, 2019b). Sixty-four (plastic) pots (27.94 cm height × 24.5 cm width) were filled with 10 kg sand portions. Ten healthy achenes of sunflower were sown in each pot, and only six healthy plants were kept after three weeks of cultivation per pot. All pots received two-liters full-strength Hoagland solution (-/+ 120 mM NaCl) at fourteen days after seed sowing until maturity. Four α -toc concentrations (0, 100, 200 and 300 mg L⁻¹ were prepared and added at a rate of 25 mL pot⁻¹ (38 days after achene sowing) at the vegetative stage. Leaves of sunflower plants were sampled at the beginning of the reproductive stage (three weeks after foliar spray) for appraisal of physiological features.

2.3. Experimental design and environmental conditions

The experiment was laid according to factorial Completely Randomized Design (CRD) with four replicates (2 cultivars × saline irrigation water × foliar spray of vitamin E). The trial was led in an ambient environment with the sunshine of 5.6 to 10.4 h, tem-

perature 16.5 to 36.1 °C, rainfall 67.9 to 11.6 mm and relative humidity 66 to 39% from February to June, respectively.

2.4. Morphological parameters

Sunflower plants were uprooted easily as grown in the sand, so no damage occurred to the roots. Roots were then washed with tap water several times to remove dirt, deionized water, and dried. Root and shoot (fresh) weight was recorded immediately after plant harvest. Lengths of roots and shoots were recorded using a measuring tape.

2.5. Gas exchange attributes

IRGA (infrared gas analyzer) was used for the recording of gas exchange parameters. Net CO₂ assimilation rate, transpiration rate, sub-stomatal (internal) CO₂ concentration and stomatal conductance values of 3rd leaf (from the top) were recorded. Leaf chamber showed values of ambient pressure 98.8 kPa, rate of gas flow 351 μmol s⁻¹, ambient CO₂ conc. 350 μmol mol⁻¹, temperature 32.4 to 36.1 °C and PAR (active photosynthetic radiation) value was most up to 1796 μmol m⁻² s⁻¹ during data recording from 11:00 am to 1:00 pm.

2.6. Chlorophyll contents

Quantification of carotenoids, chlorophyll *a* and *b* were done following the procedures of Arnon (1949). Lamina (0.5 g) of fresh leaf third from the top was chopped into tiny bits and immersed in (10 mL) 80% acetone at 0–4 °C overnight. The absorbance of the mixture (supernatant) at 645, 663 and 480 nm wavelengths were recorded with the spectrophotometer (UV-2550; Shimadzu, Kyoto, Japan).

2.7. Data analysis

The data were tested using a two-way ANOVA (Analysis of Variance) to compare the means of different treatments by finding significance values. Also, LSD at 5% significance level ($p \leq 0.05$) was applied to compare the treatments in detail. Then, a significant difference at $p \leq 0.05$ with various letters showed in each treatment. To compute associations between various measured variables, Pearson's correlation analysis was implemented. The graphical representation of data was carried out by Origin 2021 software.

3. Results

3.1. Morphological attributes

Effect of different levels of α -tocopherol (α -toc) was significant for shoot fresh weight (Fig. 1A), root fresh weight (Fig. 1B), shoot length (Fig. 1C) and root length (Fig. 1D) of different sunflower varieties (FH-572 and FH-621) under salt stress. No significant change was noted in shoot fresh weight of FH-572 + C and FH-621 + C compared to FH-572 + S and FH-621 + S. Treatment 100 mg/L- α -toc and 300 mg/L- α -toc, caused significant improvement in shoot fresh weight of FH-572 + C, FH-572 + S and FH-621 + S over 0 mg/L- α -toc. No significant change was noted in shoot fresh weight of FH-572 + C and FH-621 + C, FH-572 + S and FH-621 + S were noted in 200 mg/L- α -toc compared to 0 mg/L- α -toc. On average, genotype FH-621 performance was significantly better than FH-572 under salinity stress. For root fresh weight, FH-572 + C, FH-572 + S, FH-621 + C and FH-621 + S did not differ significantly at 0 mg/L- α -toc. Application of 100 mg/L- α -toc differed significantly for an increase in root fresh weight in

FH-572 + C and FH-621 + C than 0 mg/L- α -toc. However, no significant change in root fresh weight was observed at 200 and 300 mg/L- α -toc among FH-572 + C, FH-572 + S, FH-621 + C and FH-621 + S over 0 mg/L- α -toc. Change in shoot length was non-significant among FH-572 + C, FH-572 + S, FH-621 + C and FH-621 + S under 0 and 100 mg/L- α -toc. Treatment 200 mg/L- α -toc differed significantly over 0 mg/L- α -toc for improvement in shoot length among FH-572 + C, FH-572 + S, FH-621 + C and FH-621 + S. FH-572 + C and FH-572 + S shoot length was significantly higher at 300 mg/L- α -toc compared to 0 mg/L- α -toc. It was noted that root length of FH-621 + C and FH-621 + S differed significantly better in 100 mg/L- α -toc over 0 mg/L- α -toc. Treatments 200 and 300 mg/L- α -toc did not differ significantly for root length of FH-572 + C, FH-572 + S, FH-621 + C and FH-621 + S from 0 mg/L- α -toc.

3.2. Photosynthetic pigments

The impact of different levels of α -toc was significant for chlorophyll *a* (A), chlorophyll *b* (B), total chlorophyll (C) and carotenoids (D) of FH-572 and FH-621 under salt stress. A significant decrease was noted in chlorophyll *a* of FH-572 + S and FH-621 + S over FH-572 + C and FH-621 + C. Treatment 100 mg/L- α -toc and 300 mg/L- α -toc, caused significant improvement in chlorophyll *a* of FH-572 + S over 0 mg/L- α -toc. A significant enhancement was noted in chlorophyll *a* of FH-572 + C and FH-621 + C, FH-572 + S and FH-621 + S were noted in 200 and 300 mg/L- α -toc compared to 0 mg/L- α -toc. In the case of chlorophyll *b*, FH-572 + C, FH-572 + S and FH-621 + S remained significantly better at 100 mg/L- α -toc over 0 mg/L- α -toc. Addition of 200 and 300 mg/L- α -toc also remained significantly better for an increase in chlorophyll *b* in FH-572 + C, FH-572 + S and FH-621 + C, FH-621 + S than 0 mg/L- α -toc. Change in total chlorophyll was significant among FH-572 + S, FH-621 + C and FH-621 + S where 100 mg/L- α -toc was applied over 0 mg/L- α -toc. Treatment 200 and 300 mg/L- α -toc remained significantly better over 0 mg/L- α -toc for an increase in total chlorophyll among FH-572 + C, FH-572 + S, FH-621 + C and FH-621 + S. Results showed that FH-572 + C, FH-572 + S and FH-621 + S carotenoids were significantly enhanced at 100, 200 and 300 mg/L- α -toc than 0 mg/L- α -toc (see Fig. 2).

3.3. Gas exchange attributes

The influence of variable levels of α -toc was significant for net CO₂ assimilation rate (A), transpiration rate (E), stomatal conductance (gs), Sub-stomatal CO₂ concentration (Ci), relative internal CO₂ concentration (Ci/Ca) and water use efficiency (A/E) of FH-572 and FH-621 under salt stress. Treatment 100 mg/L- α -toc caused significant improvement in A compared to 0, 200 and 300 mg/L- α -toc in FH-572 + C. However, 300 mg/L- α -toc remained significantly best for enhancement in A of FH-572 + S than 0, 100 and 200 mg/L- α -toc. Addition of 200 and 300 mg/L- α -toc differed significantly better for an increase in A of FH-621 + C and FH-621 + S over 0 and 100 mg/L- α -toc. A maximum increase of 17 and 22% was noted in FH-572 + S and FH-621 + S where 300 mg/L- α -toc was applied than 0 mg/L- α -toc respectively. In the case of E, the interactive effect (α -toc \times V) and the main effect of α -toc were non-significant. For A/E, 100 mg/L- α -toc remained significantly best for the significant increase in A/E over 0 mg/L- α -toc in FH-572 + C. All treatments remained statistically alike to each other for A/E in FH-572 + S and FH-621 + C. Application of 300 mg/L- α -toc in FH-621 + S differed significantly for A/E over 0 mg/L- α -toc. For gs, 300 mg/L- α -toc was significantly different compared to 0 mg/L- α -toc in FH-572 + C. Treatment 100 mg/L- α -toc differed significantly for gs than 0 mg/L- α -toc in FH-572 + S. However, 200 and 300 mg/L- α -toc

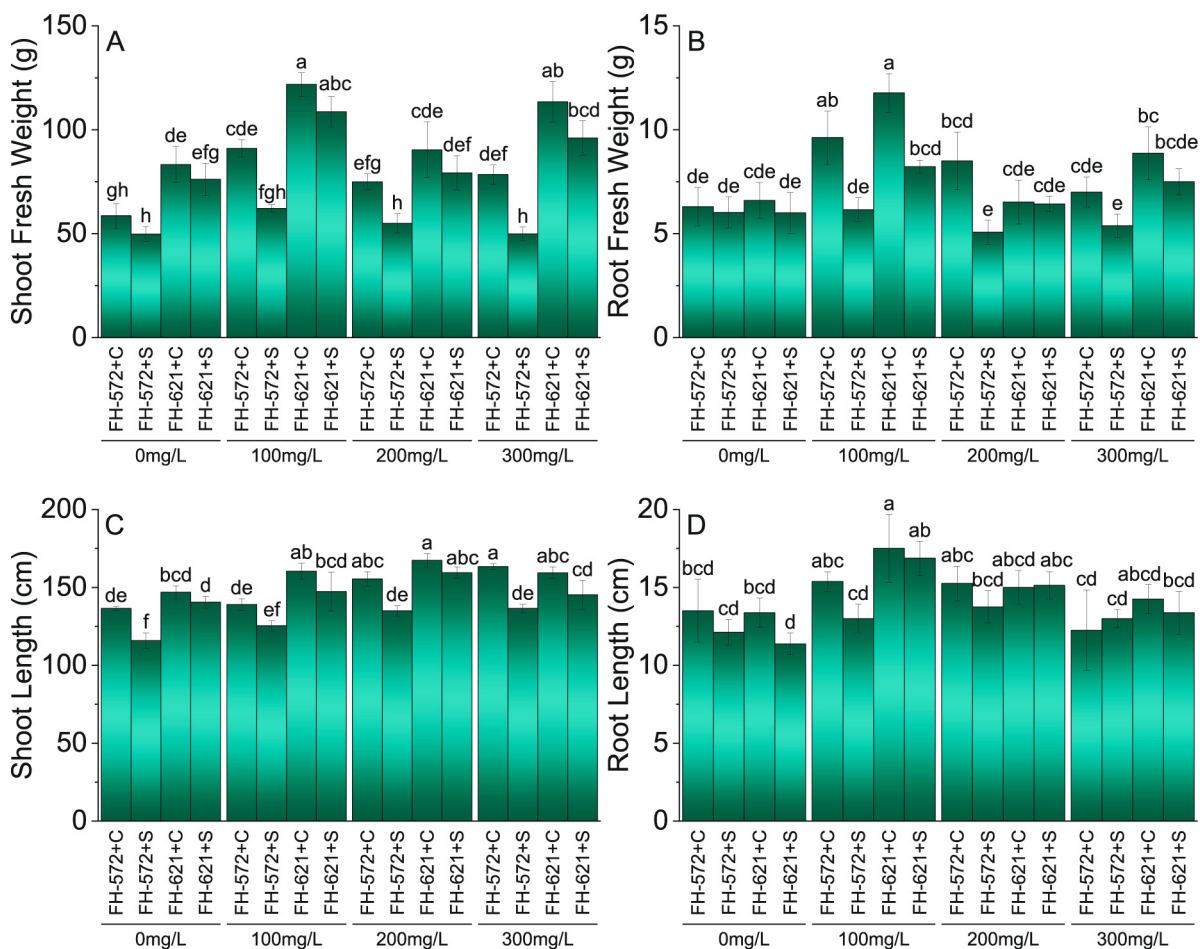


Fig. 1. Impact of α -toc foliar application on shoot fresh weight (A), root fresh weight (B), shoot length (C) and root length (D) of different sunflower varieties (FH-572 and FH-621) under salt stress and non-stressed conditions. Different letters at bars showed significant change at $p \leq 0.05$. Values 0, 100, 200 and 300 mg/L are showing the levels of α -toc concentrations. C = non saline (control); S = Salinity stress (120 mM NaCl).

was significantly better than 0 mg/L- α -toc for gs in FH-572 + S and FH-621 + C. In the case of Ci, the interactive effect (α -toc \times V) and the main effect of varieties were non-significant. In Ci/Ca, 100 mg/L- α -toc remained significantly better compared to 0 mg/L- α -toc in FH-572 + S. In FH-621 + C and FH-621 + S no significant improvement was noted by the addition of 100, 200 and 300 mg/L- α -toc over 0 mg/L- α -toc (Table 1).

Table 1. The data are given as mean of four replicates. Different letters showed significant change at $p \leq 0.05$. Values 0, 100, 200 and 300 mg/L are showing the levels of α -toc concentrations. Non-significant interaction (α -toc \times V) did not have any letter. C = non saline (control); S = Salinity stress (120 mM NaCl) [A = net CO₂ assimilation rate; E = Transpiration rate; gs = Stomatal conductance; Ci = Sub-stomatal CO₂ concentration; Ci/Ca = Relative internal CO₂ concentration; WUE (A/E) = Water use efficiency]. ns = non-significant; * = significant.

3.4. Pearson correlation

Significant positive correlations exist among genotype (variety), shoot length, A/E and gs. However, genotype (variety) was significant negative in correlation with Ci/Ca. The application of different levels of α -toc were significant and positive in correlation with shoot length, chlorophyll a, chlorophyll b, total chlorophyll, carotenoids, gs and Ci. It was observed that a significant negative correlation was existed among E and shoot fresh weight, root fresh weight, shoot length, root length, chlorophyll a, chlorophyll b, total

chlorophyll and carotenoids. On the other hand, shoot fresh weight, root fresh weight, shoot length, root length, chlorophyll a, chlorophyll b, total chlorophyll and carotenoids were significantly positive in correlation with Ci (Fig. 3).

4. Discussion

Salt stress negatively affected the fresh weights of sunflower root and shoot, especially in FH- 572 cultivar. Similar results reported the drastic effect of salt stress on the growth of *Vigna unguiculata* (Patel et al., 2010), sunflower (Kumar et al., 2014), bean and wheat (Radi et al., 2013). The growth inhibition under salt stress conditions is probably related to the adverse water relation and the influence of ion imbalance on plant metabolic functions (Lalarukh and Shahbaz, 2020). However, the adverse impact of NaCl stress on inhibiting plant growth can be mitigated by α -toc foliar application, which can accelerate the activities of enzymatic antioxidants, increase osmolytes production and improve ion homeostasis (Sadiq et al., 2019). Results obtained herein indicate that the foliar application of α -toc leads to notable increases in the growth of roots and shoots. Previous studies also reported significant improvements in the growth of plants owing to α -toc foliar application, e.g., *Oryza sativa* (Mohammed and Tarpley, 2011), *Triticum aestivum* (Dawood et al., 2014), *Vicia faba* (Orabi and Abdelhamid, 2016), citrus (Kostopoulou et al., 2014) and *Allium cepa* (Semida et al., 2016). Probably such increases occurred due to enhanced division and extension growth

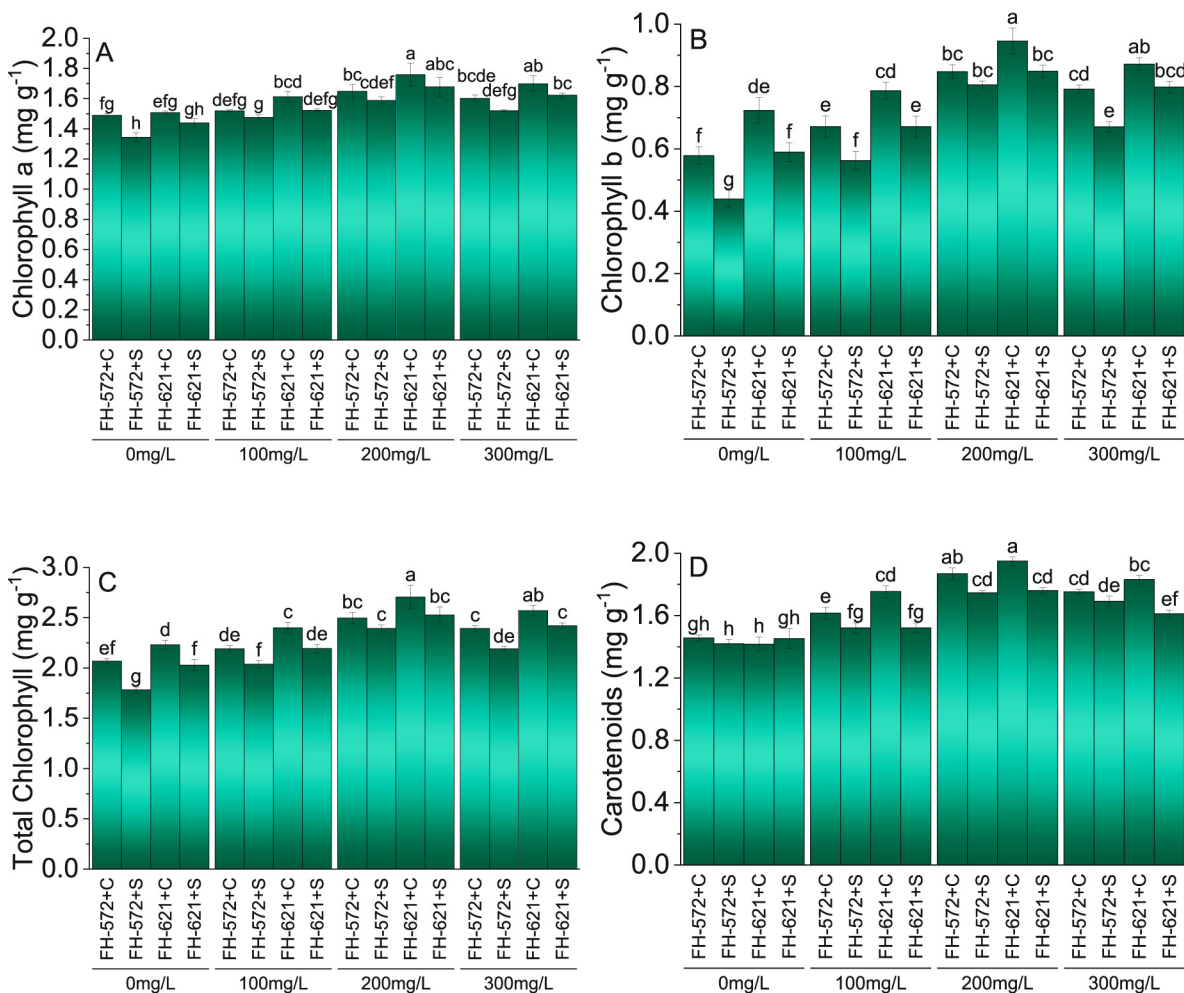


Fig. 2. Impact of α -toc foliar application on chlorophyll a (A), chlorophyll b (B), total chlorophyll (C) and carotenoids (D) of different sunflower varieties (FH-572 and FH-621) under salt stress and non-stressed conditions. Different letters at bars showed significant change at $p \leq 0.05$. Values 0, 100, 200 and 300 mg/L are showing the levels of α -toc concentrations. C = non saline (control); S = Salinity stress (120 mM NaCl).

Table 1

Mean values \pm SD of data for gas exchange characteristics of sunflower plants upon foliar application of α -tocopherol under salt stress and non-stress conditions.

Treatment	Sunflower varieties	Main Effect of Treatments (Mean \pm SD)					
		A ($\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$)	E ($\text{mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$)	A/E ($\mu\text{mol CO}_2/\text{mmol H}_2\text{O}$)	gs ($\text{mmol m}^{-2} \text{ s}^{-1}$)	Ci ($\mu\text{mol mol}^{-1}$)	Ci/Ca
C-0 mg/L	FH-572	32.42c-f	2.74	10.09e-g	458bc	160	0.71c-e
C-100 mg/L	FH-572	37.28ab	2.51	15.07ab	328c-e	225	0.64de
C-200 mg/L	FH-572	30.89d-g	3.07	10.46e-g	360c-e	268	0.76a-c
C-300 mg/L	FH-572	26.52 h	2.29	9.63e-g	223e	255	0.71c-e
S-0 mg/L	FH-572	29.78e-h	3.66	8.99 fg	477a-c	154	0.67c-e
S-100 mg/L	FH-572	29.59e-h	2.93	10.37e-g	223e	211	0.85a
S-200 mg/L	FH-572	28.80f-h	2.93	8.54 g	543ab	247	0.75a-d
S-300 mg/L	FH-572	34.92b-d	2.81	11.42d-g	620ab	236	0.74a-d
C-0 mg/L	FH-621	31.09d-g	2.31	13.78a-d	260e	224	0.74b-d
C-100 mg/L	FH-621	30.98d-g	2.17	15.25ab	273de	255	0.47f
C-200 mg/L	FH-621	39.77a	2.77	14.59a-c	478a-c	259	0.82ab
C-300 mg/L	FH-621	36.01a-c	1.96	12.40b-e	610ab	217	0.62e
S-0 mg/L	FH-621	27.51gh	3.06	11.78c-f	360c-e	200	0.69c-e
S-100 mg/L	FH-621	31.52d-g	2.68	12.17b-e	443b-d	250	0.48f
S-200 mg/L	FH-621	34.40b-d	2.33	14.13a-d	573ab	255	0.61e
S-300 mg/L	FH-621	33.47b-e	2.84	16.39a	648a	234	0.62e
p-values	α -tocopherol	0.0160*	0.0750 ns	0.0673 ns	0.0001*	0.0000*	0.0003*
	Varieties	0.0049*	0.0019*	0.0000*	0.0043*	0.1111 ns	0.0000*
	α -toc \times V	0.0000*	0.2061 ns	0.0049*	0.0009*	0.117 ns	0.0000*

in cells by increasing IAA (plant hormone) level (Dawood et al., 2014).

Further adverse effects for salinity stress were detectable on (1) decreasing plant pigments (carotenoids and Chl. b) and (2) the drop in relative water content percentage (RCW%) of leaves. Previ-

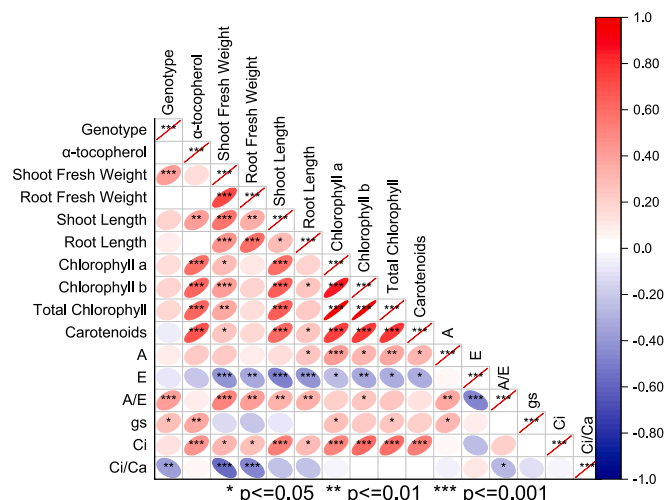


Fig. 3. Pearson correlation of different sunflower attributes under salt stress and non-stress conditions. Intensity of blue color showed negative correlation while red color showed positive correlation.

ous studies highlighted the significant reductions that occurred in photosynthetic pigments under salt (160 mM NaCl) stress conditions in bean and wheat (Radi et al., 2013). Likewise, Gengmao et al. (2015) recorded significant reductions in Chl. a and b contents of safflower leaf under (100 mM) NaCl stress while detecting no significant changes in leaf carotenoids. Probably, reduction in chlorophyll a and b affect negatively energy balance conservation (Björkman, 1981). On the other hand, Semchuk et al. (2012) found that carotenoid pigment and antioxidants increased under salt stress in *Arabidopsis thaliana*. In the case of the reductions that occurred in water relation parameters under salt stress conditions, our results were confirmed by Koyro (2006), who reported that salt stress-induced drop in osmotic and water potential of leaf in *Plantago coronopus* and *Brassica juncea* whereas, Shaheen et al. (Shaheen et al., 2013) expressed an increase in turgor potential of eggplant leaf. Probably, sunflower cultivars exhibit higher salinity tolerance than many other crops because plants contain higher contents of glycine betaine (GB), free proline and (total) free amino acids that increased the negative osmotic and water potential values of leaf (Irfana et al., 2020).

Our findings showed that spraying tolerant sunflower cultivars with α -toc significantly improved plant pigments contents (chlorophyll and carotenoids) and increased leaf cells' turgor potential. These results were confirmed in *Vicia faba* (El Bassiou et al., 2005), indicating protection/stability from photo-oxidation to photosynthetic apparatus. Furthermore, foliar spray of α -toc can maintain osmolality, stabilizing proteins and increasing the rate of photosynthesis (Ashfaque et al., 2014). It is worth mentioning that leaf stomata are turgor-operated valves (Franks and Brodribb, 2005) that regulate the uptake of carbon dioxide needed for photosynthesis. Thus, this foliar spray significantly improved sunflower photosynthetic rate and stomatal conductance (g_s). A decrease in gas exchange characteristics was reported under salt stress by many researchers in sunflower (Akram et al., 2012), wheat (Kanwal et al., 2011) and canola (Shahbaz et al., 2013). It seems that reductions in transpiration rates of stressed plants were probably due to the reduction that occurred in plant growth (Tian et al., 2020), rather than being an effective adaptive mechanism to increase plant tolerance when grown under salinity stress conditions. Spraying plants with α -toc improved this adaptation mechanism by decreasing further plant transpiration rate; despite that, it lessens stomatal conductance (g_s). This may occur via induction of rapid stomatal closure. As mentioned earlier, the

results support the study's primary assumption as the foliar application of α -toc seemed to be an effective technique in mitigating the inhibitory effect of salt stress on sunflower plants by improving gas exchange attributes and hence improving plant vigor and tolerance. In the present study, foliar spray of α -toc and salt stress showed a significant impact on the osmotic, turgor and water potential of the leaf.

5. Conclusions

A positive effect of alpha-tocopherol foliar application on morpho-physiological features of two sunflower cultivars was noted. The spray i.e., 100 and 200 mg/L- α -toc are the best application rates had a remarkable influence on the majority of study attributes in both FH-572 and 621 cultivars in salt stress. Sunflower cv. FH-621 seemed to be more tolerant to salt stress than FH-572. Therefore, in conclusion, the adverse impact of salt stress on sunflower can be mitigated by the foliar application of α -toc (100 and 200 mg/L- α -toc) are the best application on a sunflower at earlier stages. In future, more studies are recommended at the field level under different agro-climatic zones and salinity levels to declare 100 or 200 mg/L- α -toc as best foliar application rates for salinity stress alleviation.

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CRedit authorship contribution statement

Irfana Lalarukh: Conceptualization, Methodology, Software, Validation, Formal analysis, Investigation, Resources, Data curation, Supervision. **Xiukang Wang:** Conceptualization, Methodology, Software, Validation, Formal analysis, Investigation, Resources, Data curation. **Syeda Fasiha Amjad:** Conceptualization, Methodology, Software, Validation, Formal analysis, Investigation, Resources, Data curation. **Rashid Hussain:** Writing – original draft. **Sunny Ahmar:** Writing – original draft. **Freddy Mora-Poblete:** Writing – review & editing. **Shams H. Abdel-Hafez:** Writing – review & editing. **Mustafa A. Fawzy:** . **Mohamed H.H. Abbas:** Writing – review & editing. **Ahmed A. Abdelhafez:** Writing – review & editing. **Rahul Datta:** Writing – original draft, Writing – review & editing.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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