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RESEARCH ARTICLE

Comparative Study of Salinity-induced Drastic Effects on Morphological and Physiological Attributes of Okra Cultivars

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ABSTRACT

Okra (*Abelmoschus esculentus L. Moench*) is a common vegetable consumed in almost all parts of Pakistan that can grow in diverse climatic conditions and is coming across the problems of biotic and abiotic stress, resulting in ionic imbalance and nutritional deficiencies in the okra plant. The pot experiment was conducted to see the comparative response of two salt-tolerant (OH-713 and OH-139) and two salt-sensitive (Sitara-9101 and Okra-7080) okra genotypes under salt stress conditions applied to the leaf water status, physiological, enzymatic and ionic attributes. The treatment levels were 2, 4, 6, and 8 dS.m^{-1} and a control in which nothing was supplied except water and Hogland solution. Results showed that salinity affected all parameters of the plant in the form of reduction in salt sensitive species as compared to that of salt tolerant species of okra. The transpiration rate was reduced to 48% and 61.15%, respectively in salt tolerant as compared to that of salt sensitive that was 70% and 80 %. Water use efficiency was also increased in response to salinity that was 11.60% and 10.10% in salt tolerant genotypes while in salt sensitive it was about 50% and 70%, respectively as compared to that of control. Nitrate reductase activity (NRA) was reduced to 20.66% and 24.50% in salt sensitive genotypes while in those of salt tolerant genotypes; it was observed 47.10% and 55.66% as compared to that of control. It was inferred that tolerant cultivars stored high contents of Mg^{2+} and K^+ in their leaves as compared to that of roots which possibly made them able to withstand under saline environment.

INTRODUCTION

Plants have to cope with various abiotic and biotic stress factors during their life span and being tolerant or resistant against these unfavorable conditions ensure their survivability. In salt delicate species of plants, the stress possessions fluctuate significantly reliant on the degree and interval of the salt stress but also on influences concerning to plant individualities (Tarchoun et al., 2021). Abiotic stresses comprised of either excess or deficit situation of water, salts, nutrients and temperature etc. Salinity is among the natural severe disasters, restricting plant growth and productivity thus poses to be an obstacle in attaining sustainability in agriculture (Allakhverdiev et al., 2000). In saline surroundings, growth of plant is badly affected by multifarious collaboration of osmotic effects, hormones, precise ion effects and nutritional disparities, perhaps all happen instantaneously (Mahougnon et al., 2021) The increment in salinity level of cultivated soils is partly contributed by irrigation water, technique and frequency (Rubio et al., 2009). It is assessed that approximately 7% of the world land is pretentious by salinity and around 20% of 230 million ha moistened land is salt affected (Ucarli, 2020). Salinity of soil is being considered responsible for producing the effects on 7% and 5% of cultivated land including rainfed and irrigated areas (Ghassemi et al., 1995 and Hillel, 2000).

The salt stress is known as one of the main abiotic stress dynamics restraining the output of numerous agronomic plant species. Though, as a sessile creature, plant might regulate their metabolism reprograming several diverse complex passageway targeting tolerate such diverse stresses by initiating transcriptional and genetic elements (Corte-Real et al., 2020). Salinity is considered as a chief risk to agriculture, amongst other abiotic stresses; presently, 20% or more of agricultural acreage is disturbed by salinity, which is intensifying with the passage of time and already disturbs nearly 954 million hectares of the overall land area of the world (Saddiq et al., 2021). The declining effects of high salt level in soil solution create prominent responses in plant genotypes. These responses (morphological, physiological, biochemical, ionic and molecular) ensure their sustainability in salt stress regimes. (Rahnama and Ebrahimzadeh, 2004) articulated that high salt levels in the sand owing to elevated levels of ions specially sodium and chloride exerts negative effects on plants and thus play a role lower productivity (Abbas et al., 2010 and Gorai et al., 2010). Such increase in ions of soil solution grounds more uptakes of these ions and results in ionic imbalance/toxicity and nutritional deficiencies owing to nutrient replacement by them. Alteration in plant water relations has been reported as an important outcome of salt stress (Yeo et al., 1985). A high concentration of salts in soil solution leads to higher uptake of salt ions (sodium and chloride), which raises the cellular osmotic potential thus altering the water potential (Zribi et al., 2009 and Hayat et al., 2010). Such alteration leads to various responses at both cell and whole plant level (Hasegawa et al., 2000). Regulation of ionic homeostasis is an important step in high osmotic stress, which equips the plant to survive in stressed condition (Xiong and Zhu, 2002).

Great intensities of salinity badly affect the germination, reproducibility and plant growth through badly affecting the various metabolic activities as respiration, photosynthesis, transpiration, nutrient balance, membrane properties cellular homeostasis, enzymatic activities and hormone imbalance (Bakhoum et al., 2020). Plant photosynthetic mechanism is a chief physiological mechanism that is affected by high salt level in plant growing conditions. Salt stress causes adverse changes in this process by imposing stern threats to the photosynthetic apparatus. (Dobrota, 2006) explained that such alterations disrupt the progression of ATP synthesis. (Garcia-Legaz et al., 1993) reported a converse relation between elevated chloride and sodium contents and gas exchange attributes including stomatal conductance, photosynthesis rate and transpiration.

Salt stress inflicts the plant with serious nutritional deficiencies but the extent of this deficiency is determined by the type and nature of the plant as well as the salinity level, nutrient availability in soil solution and growing conditions (Grattan and Grieve, 1999). An efficient nutrient supplementation program for the salt stressed plant can act as a rejuvenating factor which aids in improving its survivability (Hu and Schmidhalter, 2005). Amongst numerous field harvests, usually, wheat is extra delicate to salinity that hinders the growth and development of plant, clues to low production or even whole crop failure under dangerous levels of salinity (EL Sabagh et al. 2021). Okra is a very popular vegetable, grown as an annual crop (Chauhan, 1972). Keeping in view, this study was conducted to characterize the available okra cultivars on the basis of their response under salt stressed conditions, so that we may be able to utilize the large saline area for its production.

MATERIALS AND METHODS

Plant material and growth conditions

Seeds of two salt tolerant (OH-713 and OH-139) and two salt sensitive (Sitara 9101 and Okra-7080) genotypes (previously tested for their salt tolerance potential) were sown in plastic pots filled with Astatula fine sand as a growth medium. The sand with field capacity 7.2%, pH ranges from 6.0-6.5, and incipient wilting @ 1.2 % was used. Half strength Hoagland solution was employed as nutrient medium.

Salinity applications

Sodium chloride (NaCl) @ 0, 2, 4, 6 and 8 $dS.m^{-1}$ was applied 20 days after sowing. In order to avoid osmotic shock, the salinity levels were attained gradually in 48 hours.

Measurement of physiological attributes

Photosynthesis (Pn), transpiration (*E*) and stomatal conductance (g_s)

For the measurement of photosynthesis rate (Pn), transpiration rate (*E*) and stomatal conductance (g_s) three leaves $plant^{-1}$ (two plants in each replication $treatment^{-1}$) were selected. These leaves were placed one by one in the chamber of portable apparatus termed as Infra-Red Gas Analyzer (IRGA) (Analytical Development Company, Hoddesdon, England) and data was recorded following the protocols described by (Zekri, 1991 and Moya et al., 2003).

Water use efficiency (WUE)

It is the ratio between photosynthesis (Pn) and transpiration (E) and was measured using the equation:

WUE=Pn/E

Measurement of enzymatic activity

The nitrate reductase activity was measured by following the protocol of (Sym, 1984 and Ramarao et al., 1983), respectively.

Measurement of ionic contents

Potassium contents in leaf and root samples were measured using the procedure described by Wolf (1990) for sample digestion followed by data recording using Flame Photometer (Jenway PFP-7, UK). $Mg^{2+}$was determined titrimetrically against EDTA solution (0.01 *N*) as a standard solution using EBT and calcon indicators as described in US-Staff Hand Book-60 (1962).

Measurement of plant water relations

Leaf water potential (-ψ_w)

After 45 days of plant growth, a razor was used to cut the fully flourished leaf of six plants (two from each replicate treatment^{-1}) and placed in the gasket of pressure chamber (Model, 615, USA) to calculate the ψ_w.

Leaf osmotic potential (-ψ_s)

The same leaf that was used in pressure chamber for ψ_w was placed in a plastic bag and kept at low temperature (-20^{o}C) in a freezer for 7days. The frozen leaf material was then thawed at room temperature for half an hour and cell sap was extracted with the help of a disposable syringe. The 10 µL of extracted sap was placed on osmometer (Wescor Model-5500) with the help of plastic syringe and ψ_s measurement was taken.

Leaf turgor potential (ψ_t)

Turgor potential (ψ_t) signifies the difference between ψ_w and ψ_s, therefore was calculated as: $(\Psi_t) = (-\Psi_w) - (-\Psi_s)$

Relative water contents (RWC)

Three mature leaves were detached from randomly selected six plants (two plants from each replication treatment^{-1}). Tap water was used to wash the sample leaves for at least five minutes and then tissue paper was used to absorb the excessive moisture from the leaves. After staining, the leaves were weighed and dipped in water for 24 hours, so that they attain their turgidity again. After measuring the turgid weight, leaves were oven dried at 72 ^{o}C for 48 hrs and their dry weight was measured employing digital balance (Bosch AE-160, Germany). The method reported by Wheatherly and Barrs (1962) was used to calculate the average RWC replicate^{-1}.

Relative water contents = Fresh weight-Dry Weight/Turgid Weight-Dry Weight X100

Statistical analysis

The present experiment was a two-factor factorial (genotypes and salinity). The treatment means were grouped on the basis of HSD (Tucky's test) @ 0.05 level of probability by using Statistix 8.1.

RESULTS

Physiological Attributes

Salt stress significantly altered the physiological processes (Fig. 1) Photosynthetic activity (Pn) of all investigated cultivars was reduced in response to applied salinity levels. The plants submitted to non-saline conditions gave the highest values for Pn as compared to those under saline conditions. Maximum decline in Pn was recorded in plants treated with high NaCl concentration (8.0 dS.m^{-1}), followed by 6.0, 4.0, and 2.0 dS.m^{-1}. The salinized plants of OH-713 and OH-139 exhibited a minimum percent decrease in Pn as compared to Sitara-9101 and Okra-7080 with respect to the control. Salinity significantly reduced the transpiration rate (*E*) in all the tested okra cultivars but plants irrigated with +NaCl solution had a maximum percent reduction in *E* as compared to those treated with –NaCl. At high salt stress (8.0 dS.m^{-1}), all the four cultivars showed maximum reduction in *E* as compared to other salinity levels but OH-139 (21.60%) and OH-713 (28.10%) proved to be highly salt-tolerant because of less reduction in *E* as compared to Sitara-9101 (48.00%) and Okra-7080 (61.15%). Water use efficiency (WUE) was significantly increased in all okra plants submitted to saline regimes. In the case of cultivars, OH-139 and OH-713 had maximum percent enhancement in WUE as compared to the Sitara-9101 and Okra-7080 under saline conditions. At high salt stress (8.0 dS.m^{-1}), all the four cultivars showed enhancement in WUE but OH-713 (11.46%) and OH-139 (10.01%) proved to be highly salt-tolerant because of the high increase in WUE as compared to Sitara-9101 and Okra-7080. Stomatal conductance (g_s) of both tolerant and non-tolerant cultivars was reduced by salt stress but the maximum reduction was recorded in plants grown under 8.0, followed by 6.0, 4.0, and 2.0 dS.m^{-1}NaCl stress. However, salt-tolerant cultivars (OH-713 and OH-139) maintained maximum g_s than non-tolerant cultivars (Sitara-9101 and Okra-7080) under saline conditions.

Enzymatic activity

Salinity caused a marked reduction in enzymatic activity of okra cultivars (Fig. 1). Nitrate reductase activity (NRA) was high in plants grown under control conditions but it was gradually decreased with the increasing salinity levels from 2.0 dS.m^{-1} to 8.0 dS.m^{-1}. The salt tolerant cultivars (OH-713 and OH-139) showed satisfactory performance by maintaining maximum NRA under saline conditions while the lowest NRA was measured in salt susceptible cultivars i.e. Sitara-9101 and Okra-7080. In case of plants grown under 8.0 dS.m^{-1}NaCl, OH-713 and OH-139 exhibited maximum resistance to salinity in terms of less percent reduction in NRA by 20.66% and 24.50%, respectively while Sitara-9101 and Okra-7080 proved to be highly

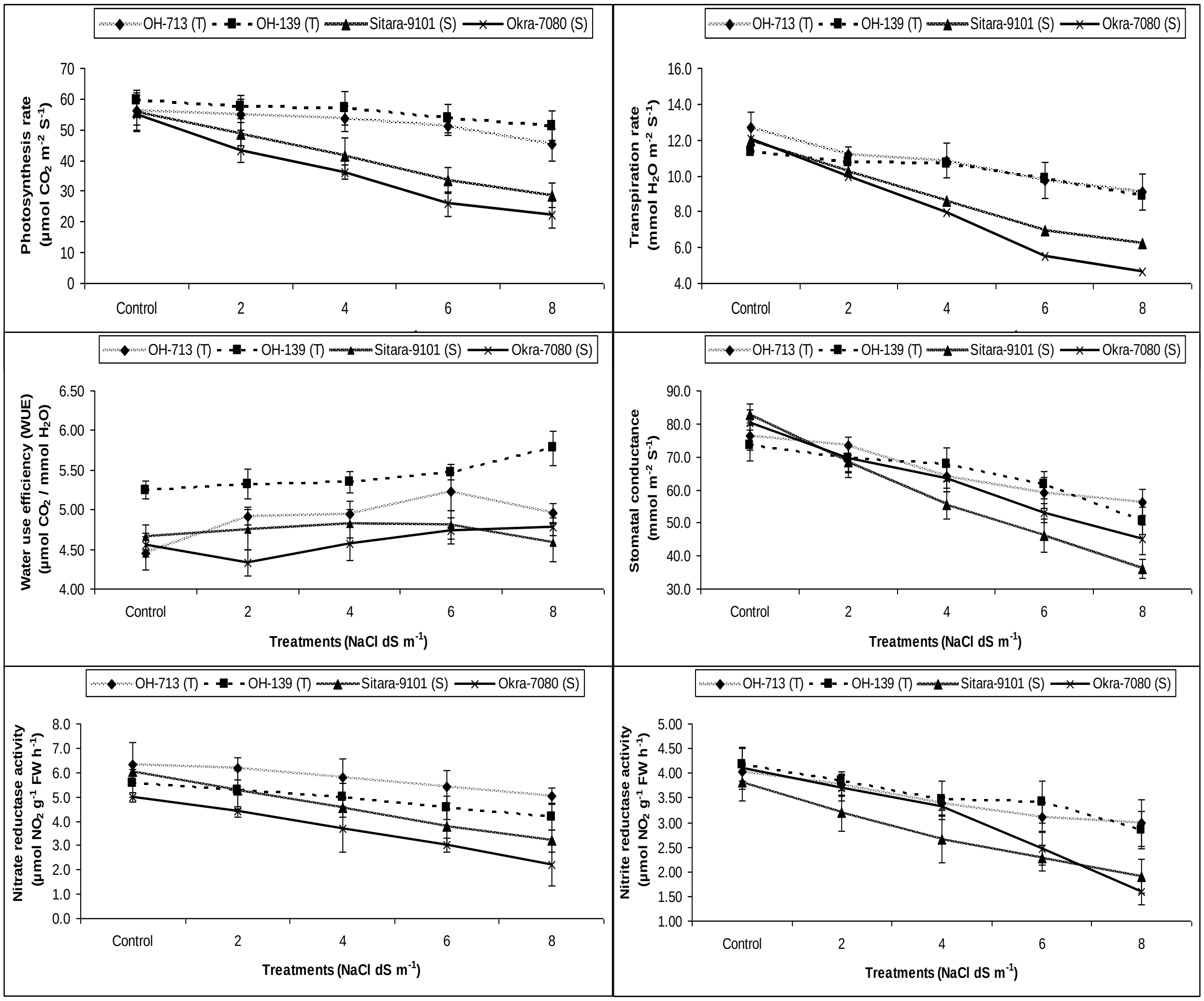

Figure 1: Effect of salt stress on physiological and enzymatic attributes of okra cultivars

salt susceptible due to maximum reduction of 47.10% and 55.62%, respectively in NRA.

Data regarding NRA described that it was reduced in both tolerant and non-tolerant okra cultivars exposed to different NaCl levels. The decline in NRA was maximum at high NaCl concentration i.e. 8.0 $dS.m^{-1}$ while maximum values for this attribute was observed under control conditions (no salinity). Both salinity resistant and non-resistant okra cultivars had varied values for NRA. The salt resistant OH-713 and OH-139 showed splendid performance by having minimum reduction in NRA under salt stressed conditions in comparison to non-resistant Sitara-9101 and Okra-7080. Among four NaCl concentrations applied to okra cultivars, maximum drastic effects on NRA were observed in plants growing under high NaCl stress (8.0 $dS.m^{-1}$) levels. So, it can be concluded that NRA also differentiated the salt tolerant cultivars from non-tolerant ones.

Ionic activity

Salt stress caused a significant reduction in ionic contents in leaves and roots of all okra cultivars (Fig. 3). The plants grown under 8.0 $dS.m^{-1}$NaCl stress demonstrated that OH-713 (31.84%) and OH-139 (40.07%) had high percent decrease values of root Mg as compared to the Sitara-9101 (26.97%) and Okra-7080 (28.19%). Whereas, in case of leaves OH-713 (22.44%) and OH-139 (29.76%) gave the excellent performance than Sitara-9101 (46.05%) and Okra-7080 (59.56%) by showing minimum percent decrease in Mg contents. Both root and leaf Mg showed continuous decrease with salt stress. However, the tolerant cultivars accumulated maximum amounts of Mg in their leaves but minimum in their roots while it was opposite in case of salt sensitive ones. On the basis of Mg contents, it can be concluded that OH-713 and OH-139 maintained high Mg in their leaves in comparison to the sensitive ones which may be linked with their high salt tolerance potential.

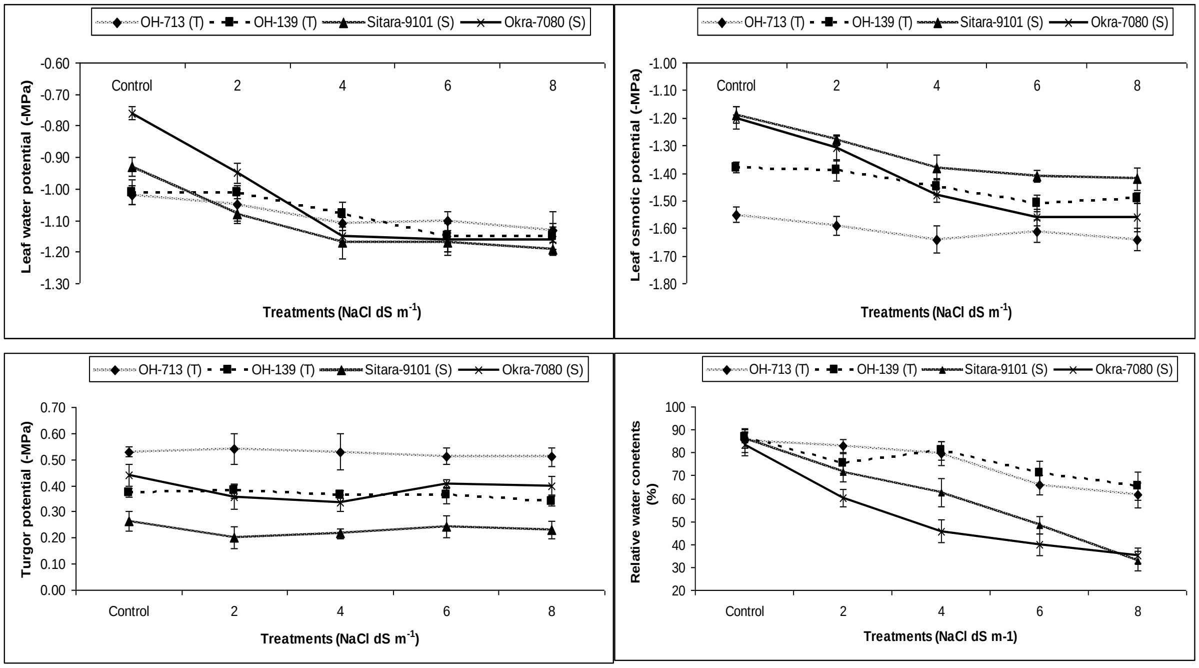

Figure 2: Effect of salt stress on leaf water status of okra cultivars

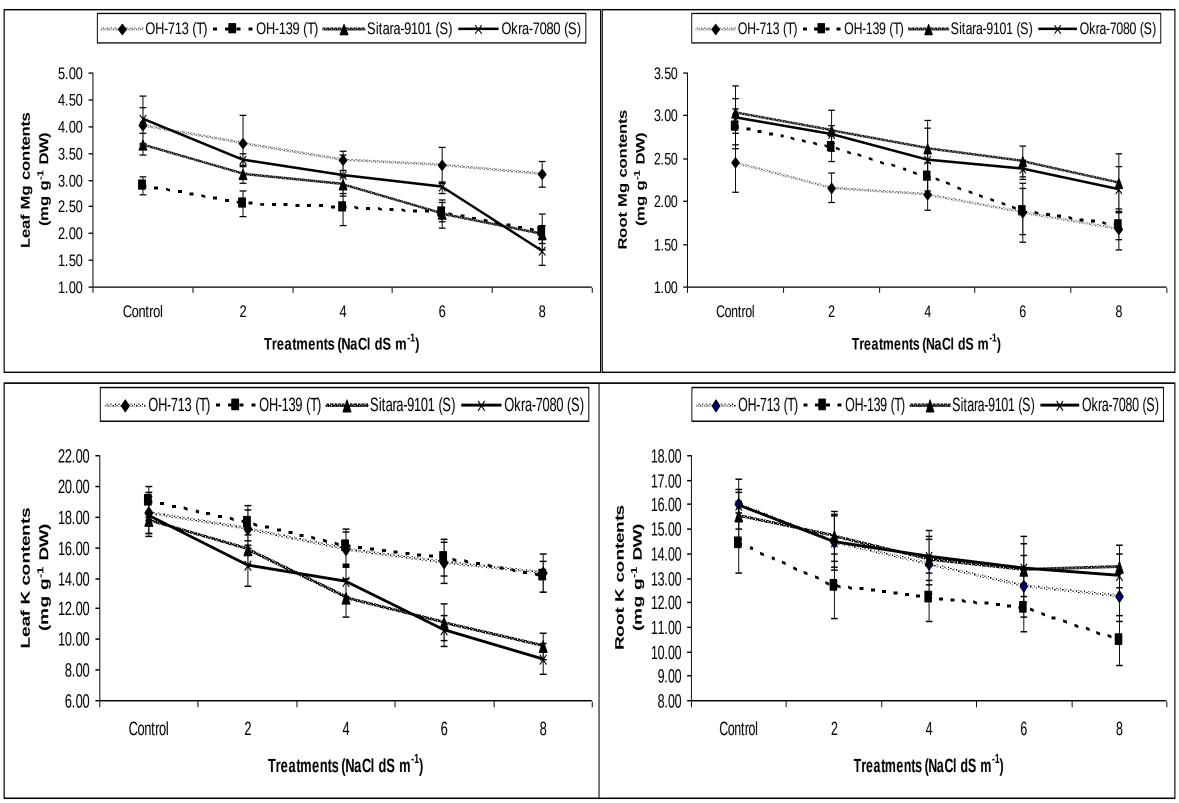

Figure 3: Effect of salt stress on ionic content of leaves and roots of okra cultivars

A significant reduction in potassium (K) contents in leaves and roots of all okra cultivars was observed. The plants grown under highest NaCl stress (8.0 dS.m^{-1}), exhibited that OH-713 (17.47%) and OH-139 (27.53%) had high percent decrease values of root K as compared to the Sitara-9101 (13.44%) and Okra-7080 (18.09%).

Whereas, in case of leaves, OH-713 (21.49%) and OH-139 (25.70%) gave excellent performance than Sitara-9101 (46.35%) and Okra-7080 (51.79%) by showing minimum percent decrease in K contents. Both root and leaf K showed continuous decrease with salt stress. However, the tolerant cultivars accumulated maximum amounts of K in their leaves but minimum in their roots while it was opposite in case of salt sensitive ones.

Water relations

Salt stress induced marked reduction in water status of tested okra cultivars (Fig. 2). Data regarding leaf water potential indicated that salinity had significant decreasing effect on it. Maximum leaf water potential was recorded in the plants grown under non-saline conditions (control), followed by 2.0, 4.0, 6.0 and 8.0 $dS.m^{-1}$NaCl stress. The salt tolerant cultivars (OH-713 and OH-139) gave good performance due to less percentage decrease in leaf water potential in response to NaCl stress. However, the salt sensitive cultivars (Sitara-9101 and Okra-7080) presented maximum percentage decrease in leaf water potential under salt stress. Salt stress induced a significant reduction in leaf osmotic potential of all the tested okra cultivars. Maximum leaf osmotic potential was noted for plants grown under non saline conditions, followed by 2.0, 4.0, 6.0 and 8.0 $dS.m^{-1}$NaCl. Under 8.0 $dS.m^{-1}$NaCl stress, OH-713 (6.03%) and OH-139 (7.97%) showed less percentage reduction while Sitara-9101(18.99%) and Okra-7080 (29.64%) surpassed the tolerant ones in this regard. Salt stress casued the considerable reduction leaf turgor potential of all investigated okra cultivars. The salt tolerant cultivars (OH-713 and OH-139) showed satisfactory performance by maintaining the maximum leaf turgor potential under saline conditions while the lowest leaf turgor potential was measured in salt susceptible cultivars i.e. Sitara-9101 and Okra-7080. In case of plants grown under 8.0 $dS.m^{-1}$ NaCl, OH-713 and OH-139 exhibited maximum resistance to salinity in terms of less percent reduction in leaf turgor potential by 3.77% and 8.93%, respectively while, Sitara-9101 and Okra-7080 proved to be highly salt susceptible due to maximum reduction of 12.66% and 9.9%, respectively in leaf turgor potential. Data regarding the relative water contents (RWC) depicted that it reduced in both tolerant and non-tolerant okra cultivars exposed to different NaCl levels. The decline in RWC was maximum at high NaCl concentration i.e. 8.0 $dS.m^{-1}$ while maximum values for this attribute were observed under control conditions (no salinity). Both salinity resistant and non-resistant okra cultivars had varied values of RWC. The salt resistant OH-713 and OH-139 showed the splendid performance by having minimum reduction in RWC under salt stressed conditions in comparison to non-resistant Sitara-9101 and Okra-7080.

DISCUSSION

Growth parameters of plants comprising stem girth, plant height, number of leaves and leaf area reduced considerably with cumulative salinity concentration. Salt stress also hinders the growth parameters in plants. Dry and fresh weight of plant fragments, number of fruits, total biomass, fresh and dry weights of fruits as well as total chlorophyll of leaves decrease by salt stress (Kekere et al., 2020). Plant growth is directly linked with photosynthesis and salt stress negatively affects the growth by disturbing the functioning of the photosynthetic apparatus (Omoto et al., 2020 and Qu et al., 2012). In the current investigation, salt stress significantly reduced the photosynthetic activity (Pn) in both salt-tolerant (OH-713 and OH-139) and salt-sensitive (Sitara-9101 and Okra-7080) okra cultivars. There are various possible reasons i.e. reduction in leaf area, chlorophyll contents, number of stomata and their size, and reduced transpiration rate that could have limited the photosynthetic activity in tested salt-stressed okra plants. Botanically, stomata are the openings within the leaf and may be in the stem epidermis that is specialized for gaseous exchange. These openings are covered by special structures called guard cells, which regulate the gaseous exchange from stomatal openings (Esau, 1977). It means these guard cells are indirectly associated in controlling the rate of photosynthesis by controlling the passage of carbon dioxide and oxygen, which are the essential components of the photosynthetic reaction. The opening and closing of guard cells is under the control of various ions specifically potassium (K+). The opening of stomatal pores requires the entry of K+ into guard cells (Blatt et al., 1990) along with some anion channels (Negi et al., 2008) activated by calcium ions (Grabov et al., 1997). However, salinity causes ion toxicity by elevating Na and Cl ions while decreasing the availability of calcium, magnesium, and potassium to plant tissues, especially the leaves, therefore, this non-availability of Ca, Mg, and K leads to the malfunctioning of stomata. This reduction in Ca and K ions in plants submitted to saline conditions will disturb the gaseous exchange via stomatal opening; resultantly the rate of photosynthesis will get reduced. So, this kind of mechanism cannot be ignored in our tested okra cultivars and the reduction in Pn in this study may also be due to disturbances in the stomatal functioning through ionic toxicity or deficiency of beneficial ions. The minimum decline in Pn of salt-tolerant (OH-713 and OH-139) as compared to salt-sensitive (Sitara-9101 and Okra-7080) could have been due to less ion-toxicity and maximum transport of beneficial ions (Ca, Mg, K) from rooting zone to the leaves. Furthermore, photosynthetic activity is also dependent on the rate of transpiration (E); thus decrease in E due to the disturbances in stomatal opening and closing under the discussed above facts could be the cause of reduced Pn in salt-stressed okra cultivars. (Shahid et al., 2011 and Balal et al., 2012) studied the pea and citrus rootstocks plants, respectively under saline conditions and observed a significant reduction in number of stomata and stomatal size. Therefore, this reduction in a number of stomata and stomatal size could be another cause of decline in Pn in okra cultivars studied. (Dinakar et al., 2021; Huang et al., 2012; Li et al., 2012; Qu et al., 2012) studied maize, artichoke and rice plants submitted to salt stress and observed a significant reduction in photosynthetic activity.

In present study, salinity also reduced the stomatal conductance (gs) in investigated okra plants of both salt-tolerant and non-tolerant cultivars. It is a vital physiological process that is associated with Pn and plant biomass formation. Stomatal conductance is linked with number of stomata and their size, rate of transpiration and leaf area (Shahid et al., 2011). A positive correlation was observed between the Pn and gs of tested cultivars. Therefore, the cultivars (OH-713 and OH-139) having high gs showed maximum Pn as compared to the Sitara-9101 and Okra-7080. Literature depicted that salinity stress significantly reduced the gs in various plant species such as wheat, grass, strawberry and *Tecticornia indica* (Orsini et al., 2012 and Zheng et al., 2020). All these reports are in agreement with the findings of present study in which salinity decreased the gs in salt-stressed plants of both salt-tolerant and sensitive okra cultivars.

Nitrogen (N) requirements of plant are fulfilled from soil in the form of nitrates. This absorbed nitrate is utilized as N source in the formation of proteins and essential amino acids. The whole process of N reduction is carried out by two catalytic-reactions, which consume only eight electrons in the absentia of ATP. In the first phase nitrate is reduced to nitrite by the enzyme nitrate reductase while in second step nitrite is converted through reduction process to ammonia by another enzyme known as nitrite reductase. This ammonia is used in the formation of amino acids. So, it is evident that both nitrate reductase and nitrite reductase enzymes ensure the availability of N and ammonia required for the formation of amino acids and proteins. The saline conditions, even a low salinity limits the activities of nitrate reductase enzyme (Lingqiang and Scandalions, 2002). In current study, all the tested okra cultivars showed a significant reduction in the nitrate reductase activity with the increasing salt stress. This reduction in nitrate reductase could be attributed to high concentration of toxic salts/ions within the plant tissues because the tolerant cultivars (OH-713 and OH-139) showed the least reduction in nitrate reductase activity as compared to the salt-sensitive (Sitara-9101 and Okra-7080). The minimum reduction in nitrate reductase activity of OH-713 and OH-139 depicted that these cultivars had accumulated less amount of toxic ions in their tissues while Sitara-9101 and Okra-7080 had high accumulation of toxic ions which hindered the functioning of nitrate reductase enzyme. (Khan et al., 1990) submitted salt-tolerant and non-tolerant sorghum varieties to saline conditions and observed that nitrate reductase activity was comparatively high in salt-stressed plants of salt-tolerant variety as compared to the sensitive one. Findings of present study also followed this trend. The reduction in nitrate reductase activity may be due to the reduced transport of NO_3-and NO_2- along with destabilization and disintegrations of membranes under the effect of excessive salts or drastic ions. Salinity decreases the availability of substrate (NO_2-) because of the reduction in nitrate reductase activity (Veuger et al., 2007). Therefore, this reduction in substrate (NO_2-) could be the possible cause of reduced nitrite reductase activity (NRA) in salt-stressed okra plants. Secondly, it is reported that NiR enzyme is present within the chloroplast, where it is activated by an electron donor termed as ferridoxin (Kasukabe et al., 2006). So, it is clear that the activity of NiR enzyme is indirectly linked with the availability or activity of ferridoxin. (Ashraf and Tufail, 1995) reported that salt stress had inhibiting effect of the ferridoxin activity and it may also be attributed to reduce NRA in investigated okra cultivars. The findings of current study are in agreement with the reports of (Surabhi et al., 2008 and Reda et al., 2011).

Plant water relations comprised of leaf water potential, leaf osmotic potential, turgor potential and leaf water contents. The salt stress reduced leaf water potential, osmotic potential and turgor potential both in salt-tolerant and salt-sensitive okra cultivars. Literature depicted that salt stress had reducing effect on water relations (Alvarez et al., 2012 and Munoz-Mayor et al., 2012) and results of current investigation has confirmed these reports. The leaves of salt-tolerant (OH-713 and OH-139) maintained less fluctuation in their water relations because they accumulated less toxic ions in their leaves as compared to salt-sensitive cultivars (Sitara-9101 and Okra-7080). On the other hand, the excessive amounts of salts in the root zone also create a drought condition known as physiological drought. In this condition, water moves from plant body to the root zone because the excessive amounts of salts in the root zone lowers the water potential of rooting medium to a level lower than that of within the plant body, consequently water moves from higher water potential (plant body) to the lower water potential (rooting zone). So, this process may also be responsible for variations in water relations of plants grown under salt stress and this condition is overcome by the osmotic adjustment process. The absolute amount of water contents required by the plant to get full saturation is regarded as relative water contents (RWC) and it is an important indicator that determines the water status in plant tissues. In current investigation salinity induced a significant decline in RWC in leaves of both salt-tolerant and non-tolerant okra cultivars but, salt-tolerant cultivars (OH-713 and OH-139) had less reduction in RWC as compared to salt-sensitive cultivars (Sitara-9101 and Okra-7080). This difference in RWC of salt-tolerant and non-tolerant okra cultivars may be associated with the decrease in the water potential of rooting medium. This decrease in water potential of growing medium resulted in physiological drought, as described earlier. Due to this specific condition, the plants cannot absorb the moisture from the soil or rooting zone so less amount of water is transferred to upper plant parts. The high ratios of RWC in OH-713 and OH-139 indicated that these cultivars had less experienced the mechanism of physiological drought, because the excessive accumulation organic and inorganic osmolytes enhanced their osmotic adjustment potential which created a balance between water potential within the plant body and root zone. Consequently, the leaves of these cultivars retained maximum moisture in their leaves, which is represented as RWC. The findings of this study regarding RWC are in accordance with the reports of (Cambrolle et al., 2011; Karlidag et al., 2011; Chakraborty et al., 2012) who studied poppy, strawberry and brassica plants, respectively under saline conditions and noted a significant decline in leaf RWC. Salinity stress has significantly caused a nutritional imbalance in tested okra cultivars by increasing or decreasing the availability of various nutrients to plants. Salt stress reduced Mg and K contents in both salt-tolerant and non-tolerant okra cultivars. Salt stress markedly reduced the concentration of beneficial ions (K and Mg) both in roots and leaves of investigated okra cultivars. This reduction in beneficial ions could have been due to antagonistic effect of Na with K and Mg. The high ratios of Na in the root zone may hinder the entry of Ca, K and Mg in the roots, thus quantities of these ions may be transported to the upper plant parts especially the leaves. On the other hand, salinity also causes the change in pH of rooting zone which disturbs the nutrient absorbing capacity of roots, consequently the supply of nutrients including K and Mg get reduced.

Conclusion

From the findings of this investigation, it can be extracted that salt stress limits the growth and productivity of okra by negatively disturbing the vital physiological processes, enzymatic activity, water status, and ionic contents. Since, tolerant cultivars (OH-713 and OH-139) well maintained these attributes under the effect of salinity so they performed better as compared to sensitive cultivars (Sitara-9101 and Okra-7080). One important thing observed in this study is that salt tolerance potential is highly associated with the ratios of K and Mg. Therefore, the tolerant genotypes with high ratios of these ions in their leaves exhibited the maximum tolerance to salt stress as compared to sensitive ones.

Authors' Contributions

TA, MRS and SS designed the experiment. MI and NS collected the soil and plant samples. MFA, THS and HN analyzed the data. KJ and MN helped in writing and proofreading of the manuscript. All the authors read and approved the final draft before publication.

REFERENCES

Ashraf M and M Tufail, 1995. Variation in salinity tolerance in sunflower (*Helianthus annuus* L.). Journal of Agronomy and Crop Science, 174: 351-362.

Allakhverdiev SI, A Sakamoto, Y Nishiyama, M Inaba and N Murata, 2000. Ionic and osmotic effects of NaCl-induced inactivation of photosystems I and II in *Synechococcus sp*. Plant Physiology, 123: 1047-1056.

Abbas W, M Ashraf and NA Akram, 2010. Alleviation of salt induced adverse effects in egg plant (*Solanum melongena* L.) by glycine betaine and sugar beet extracts. Scientia Horticulturae, 125: 188-195.

Alvarez S, MJ Gomez-Bellot, M Castillo, S Banon and MJ Sanchez-Blanco, 2012. Osmotic and saline effect on growth, water relations, and ion uptake and translocation in *Phlomis purpurea* plants. Environmental and Experimental Botany, 78: 138-145.

Blatt MR, G Thiel and DR Trentham, 1990. Reversible inactivation of K^+ channels of Viciastomatal guard cells following the photolysis of caged 1,4,5-trisphosphate. Nature, 346: 766-769.

Balal RM, MM Khan, MA Shahid, NS Mattson, T Abbas, M Ashfaq, F Garcia-Sanchez, U Ghazanfer, V Gimeno and Z Iqbal, 2012. Comparative studies on the physiobio-chemical, enzymatic and ionic modifications in salt tolerant and salt sensitive Citrus rootstocks under NaCl stress. Journal of the American Society for Horticultural Science, 137: 86-95.

Bakhoum GS, MS Sadak and EAEM Badr, 2020. Mitigation of adverse effects of salinity stress on sunflower plant (*Helianthus annuus* L.) by exogenous application of chitosan. Bulletin of the National Research Centre, 44: 1-11.

Chauhan DVS, 1972. Vegetable production in India (3rd Edn.) Published by Ram Prasad. Sons, Agra, India, pp: 28-30.

Cambrolle J, S Redondo-Gomez, E Mateos-Naranjo, T Luque and ME Figueroa, 2011. Physiological responses to salinity in the yellow-horned poppy, *Glaucium flavum*. Plant Physiology and Biochemistry, 49: 186-194.

Chakraborty K, RK Sairam and RC Bhattacharya, 2012. Differential expression of salt overly sensitive pathway genes determines salinity stress tolerance in Brassica genotypes. Plant Physiology and Biochemistry, 51: 90-101.

Corte-Real N, MS Oliveira, A Jarma-Orozco, D Fernandes, MA dos Santos, L Endres and MF Pompelli, 2020. Comparative analysis of salt-induced changes in the leaves proteome of two contrasting Jatropha curcas genotypes. Brazilian Journal of Development, 6: 39845-39872.

Dobrota C, 2006. Energy dependant plant stress acclimation. *In*: Amils R., C Ellis-Evans and H Hinghofer-Szalkay (eds), Life in extreme environments. Springer, Dordrecht.Dinakar C, D Djilianov and D Bartels, 2021. Photosynthesis in desiccation tolerant plants: Energy metabolism and antioxidative stress defense. Plant Science, 182: 29-41.

Esau K, 1977. Anatomy of seed plants. Wiley, New York, USA.

EL Sabagh A, MS Islam, M Skalicky, M Ali Raza, K Singh, MA Hossain and A Arshad, 2021. Salinity stress in wheat (*Triticum aestivum* L.) in the changing climate: adaptation and management strategies. Frontiers in Agronomy, 3: 661932.

Garcia-Legaz MF, JM Ortiz, A Garcia-Lindon and A Cerda, 1993. Effect of salinity on growth, ion content and CO_2 assimilation rate in lemon varieties on different rootstocks. Physiologia Plantarum, 89: 427-432.

Ghassemi F, AJ Jakeman and HA Nix, 1995. Salinization of land and water resources. University of New South Wales Press, Sydney, Australia.

Grabov A, J Leung, J Giraudat and MR Blatt, 1997. Alteration of anion channel kinetics in wild-type and abi1-1 transgenic *Nicotiana benthamiana* guard cells by abscisic acid. The Plant Journal, 12: 203-213.

Grattan SR and CM Grieve, 1999. Salinity mineral nutrients relations in horticultural crops. Scientia Horticulturae, 78: 127-157.

Gorai M, M Ennajeh, H Khemira and M Neffati, 2010. Combined effect of NaCl-salinity and hypoxia on growth, photosynthesis, water relations and solute accumulation in *Phragmites australis* plants. Flora-Morphology, Distribution, Functional Ecology of Plants, 205: 462-470.

Hillel D, 2000. Salinity management for Sustainable Irrigation. The World Bank, Washington DC. USA.

Hasegawa PM, RA Bressan, JK Zhu and HJ Bohnert, 2000. Plant cellular and molecular responses to high salinity. Annual Review of Plant Physiology and Plant Molecular Biology, 51: 463-499.

Hu Y and U Schmidhalter, 2005. Drought and Salinity: A comparison of their effects on mineral nutrition of plants. Annual review of Plant Biology, 168: 541-549.

Hayat S, SA Hasan, M Yusuf, Q Hayat and A Ahmad, 2010. Effect of 28-homobrassinolide on photosynthesis, fluorescence and antioxidant system in the presence or absence of salinity and temperature in vigna radiate. Annual Review of Plant Biology, 69: 105-112.

Huang Z, X Long, L Wang, J Kang, Z Zhang, R Zed and Z Liu, 2012. Growth, photosynthesis and H^+-ATPase activity in two *Jerusalem artichoke* varieties under NaCl-induced stress. Process Biochemistry, 47: 591-596.

Khan AH, MY Ashraf and AR Azmi, 1990. Effect of NaCl on growth and nitrogen metabolism of sorghum. Acta Physiologiae Plantarum, 12: 233-238.

Kasukabe Y, LX He, K Nada, S Misawa, I Ihara and S Tachibana, 2006. Over expression of spermidine synthase enhances tolerance to multiple environmental stresses and up regulates the expression of various stress-regulated genes in transgenic *Arabidopsis thaliana*. Plant and Cell Physiology, 45: 712-722.

Karlidag H, E Yildirim and M Turan, 2011. Role of 24-epibrassinolide in mitigating the adverse effects of salt stress on stomatal conductance, membrane permeability, and leaf water content, ionic composition in salt stressed strawberry (Fragaria × ananassa). Scientia Horticulturae, 130: 133-140.

Kekere O, TO Ayesa and O Akinbuwa, 2020. Mitigating impact of soil salinization on growth, yield and fruit nutritional quality of *Abelmoschus esculentus* L. (Okra) using *Arbuscular mycorrhiza* fungus. Journal of Applied Sciences and Environmental Management, 24: 619-626.

Lingqiang WL and B Scandalions, 2002. NRA is associated with protein synthesis and plant growth, affected by salinity. Horticulture, Environment and Biotechnology, 32: 88-91.

Li X, N Bu, Y Li, L Ma, S Xin and L Zhang, 2012. Growth, photosynthesis and antioxidant responses of endophyte infected and non-infected rice under lead stress conditions. Journal of Hazardous Materials, 213-214: 55-61.

Moya LJ, A Gomez-Cadenas, E Primo-Millo and M Talon, 2003. Chloride absorption in salt-sensitive *Carrizo citrange* and salt-tolerant *Cleopatra mandarin* citrus rootstocks is linked to water use. Journal of Experimental Botany, 54: 825-833.

Munoz-Mayor A, B Pineda, JO Garcia-Abellan, T Anton, B Garcia-Sogo, P Sanchez-Bel, FB Flores, A Atares, T Angosto, JA Pintor-Toro, V Moreno and MC Bolarin, 2012. Overexpression of dehydrintas14 gene improves the osmotic stress imposed by drought and salinity in tomato. Journal of Plant Physiology, 169: 459-468.

Mahougnon BGG, KK Julien, CGM Armel and BG Christophe, 2021. Salinity resistance strategy of okra (*Abelmoschus esculentus* L. Moench) cultivars produced in Benin Republic. International Journal of Plant Physiology and Biochemistry, 13: 19-29.

Negi J, O Matsuda, T Nagasawa, Y Oba, H Takahashi, M Kawai-Yamada, H Uchimiya, M Hashimoto and K Iba, 2008. CO_2 regulator SLAC1 and its homologues are essential for anion homeostasis in plant cells. Nature, 452: 483-486.

Orsini F, M Alnayef, S Bona, A Maggio and G Gianquinto, 2012. Low stomatal density and reduced transpiration facilitate strawberry adaptation to salinity. Environmental and Experimental Botany, 81: 1-10.

Omoto E, M Taniguchi and H Miyake, 2020. Adaptation responses in C_4 photosynthesis of maize under salinity. Journal of Plant Physiology, 169: 469-477.

Qu C, C Liu, X Gong, C Li, M Hong, L Wang and F. Hong, 2012. Impairment of maize seedling photosynthesis caused by a combination of potassium deficiency and salt stress. Environmental and Experimental Botany, 75: 134-141.

Ramarao CS, VH Patil, BD Dhak and SB Kadrekar, 1983. A simple in vivomethod for the determination of nitrite reductase activity in rice roots. Zeitschrift für Pflanzenphysiologie, 109:81-85.

Rahnama H and H Ebrahimzadeh, 2004. The effect of NaCl on proline accumulation in potato seedlings and calli. Acta Physiologiae Plantarum, 26: 263-270.

Rubio JS, F Garcia-Sanchez, F Rubio and V Martinez, 2009. Yield, blossom-end rot incidence and fruit quality in pepper plants under moderate salinity are affected by K^+ and Ca^{+2}fertilization. Scientia Horticulturae, 119: 79-87.

Reda M, M Migocka and G Klobus, 2011. Effect of short-term salinity on the nitrate reductase activity in cucumber roots. Plant Science, 180: 783-788.

Sym GJ, 1984. Optimization of the in vivo assay conditions for nitrate reductase in Barley (*Hordeum vulgare* L.cv. irgri). Journal of the Science of Food and Agriculture, 35: 725-730.

Surabhi GK, AM Reddy, GJ Kumari and C Sudhakar, 2008. Modulations in key enzymes of nitrogen metabolism in two high yielding genotypes of mulberry (*Morus alba* L.) with differential sensitivity to salt stress. Environmental and Experimental Botany, 64: 171-179.

Shahid MA, MA Pervez, RM Balal, NS Mattson, A Rashid, R Ahmad, CM Ayyub and T Abbas, 2011. Brassinosteroid (24-epibrassinolide) enhances growth and alleviates the deleterious effects induced by salt stress in pea (*Pisum sativum* L.). Australian Journal of Crop Science, 5: 500-510.

Saddiq MS, S Iqbal, MB Hafeez, AM Ibrahim, A Raza, EM Fatima and LF Ciarmiello, 2021. Effect of salinity stress on physiological changes in winter and spring wheat. Agronomy, 11: 1193.

Tarchoun N, W Saadaoui, N Mezghani, OI Pavli, H Falleh and SA Petropoulos, 2021. The effects of salt stress on germination, seedling growth and biochemical responses of tunisian squash (*Cucurbita maxima* Duchesne) germplasm. Plants, 11:800.

Ucarli C, 2020. Effects of salinity on seed germination and early seedling stage. In: Fahad S, S Saud, Y Chen, C Wu and D Wang (Eds.), Abiotic Stress in Plants. IntechOpen.Veuger B, JJ Middelburg, HTS Boschker, J Nieuwenhuize, P Rijswijk, EJ Rochelle-Newall and N Navarro, 2004. Microbial uptake of dissolved organic and inorganic nitrogen in Randers Fjord. Estuarine, Coastal and Shelf Science, 61: 507-515.

Wheatherly PE and C Barrs, 1962. A reexamination of the relative turgidity technique for estimating water deficits in leaves. Australian Journal of Biological Sciences, 15: 413-428.

Wolf B, 1990. A comparative system of leaf analysis and its use for diagnosing nutrient status. Communications in Soil Science and Plant Analysis, 13: 1053-1059.

Xiong L and JK Zhu, 2002. Molecular and genetic aspects of plant responses to osmotic stress. Plant, Cell and Environment, 25: 131-139.

Yeo AR, SJM Capron and TJ Flowers, 1985. The effect of salinity upon photosynthesis in rice (*Oryza sativa* L.) gas exchange by individual leaves relation to their salt content. Journal of Experimental Botany, 36: 1240-1248.

Zribi L, G Fatma, R Fatma, R Salwa, N Hassan and RM Nejib, 2009. Application of chlorophyll fluorescence for the diagnosis of salt stress in tomato *Solanum lycopersicum* (variety Rio Grande)". Scientia Horticulturae, 120: 367-372.

Zekri M, 1991. Effects of NaCl on growth and physiology of sour orange and *Cleopatra mandarin* seedlings. Scientia Horticulturae, 47: 305-315.

Zheng YH, X Li, YG Li, BH Miao, H Xu, M Simmons, and XH Yang, 2020. Contrasting responses of salinity-stressed salt-tolerant and intolerant winter wheat (*Triticum aestivum* L.) cultivars to ozone pollution. Plant Physiology and Biochemistry, 52: 169-178.