

NUTRIENT UPTAKE, PROLINE CONTENT AND ANTIOXIDANT ENZYMES ACTIVITY OF PEPPER (*Capsicum annuum* L.) UNDER HIGHER ELECTRICAL CONDUCTIVITY OF NUTRIENT SOLUTION CREATED BY NITRATE OR CHLORIDE SALTS OF POTASSIUM AND CALCIUM

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ABSTRACT

This study was conducted to evaluate the effects of higher conductivity of nutrient solution created by nitrate or chloride salts of potassium and calcium on growth characteristics of pepper plants (*Capsicum annuum* var *annuum*) during four months of growth period. Two EC5 and EC8 dS/m of Hoagland nutrient solutions were prepared using various salt combinations, namely: KCl + CaCl₂, KNO₃ + CaNO₃, and KNO₃ + CaNO₃ + NaCl. Hoagland nutrient solution with EC 1.8 dS/m served as a control. Higher conductivity treatments had different effects on pepper plant growth. The most significant reduction in growth parameters of plant height, shoot fresh weight, fruit yield and nutrients uptake was in plants treated with KCl + CaCl₂ particularly at EC8. Application of KNO₃ + CaNO₃ particularly at EC5, showed no difference as compared to the control regarding many growth parameters. Application of KNO₃ + CaNO₃ at EC5 resulted in higher shoot fresh weight compared to the control. All salinity treatments, except from KNO₃ + CaNO₃ at EC5, reduced the fruit yield compared to the control. Treatments of KCl + CaCl₂ and KNO₃ + CaNO₃ + NaCl particularly at EC8 of nutrient solution, resulted in higher leaf proline concentration, catalase and peroxidase enzymes activity compared to the control. Other conductivity treatments showed no difference in catalase or peroxidase enzymes activities. Significantly the lowest amount of leaf N, K, Mg and Ca was in KCl + CaCl₂ at EC8. On the other hand, the highest leaf macronutrient concentrations were in KNO₃ + CaNO₃ at EC5 and/or EC8 that showed only higher leaf N and Ca values compared to the control. Leaf micronutrient concentrations were the highest in KNO₃ + CaNO₃ at EC5 that generally showed no difference with control plants. However, application of KCl + CaCl₂ particularly at EC8 and to less extent KNO₃ + CaNO₃ + NaCl at EC8, reduced the leaf micronutrient concentrations. Application of KNO₃ + CaNO₃ at EC5 increased and KCl + CaCl₂ or KNO₃ + CaNO₃ + NaCl at EC8 decreased the leaf Fe concentration compared to control plants.

Key words: *Capsicum annuum*, catalase, environment, peroxidase, plant nutrition, salinity, stress, vegetable

INTRODUCTION

Salinity is a widespread challenge in arid regions that greatly restricts plant growth and production. Drought and salinity are two major environmental stresses that are somehow interrelated; generally have cumulative detrimental effects on plant growth [Souri

and Hatamian 2018, Fageria et al. 2011]. Production of agricultural foods in arid areas including many parts of Iran is largely limited by soil and water salinity [Souri and Hatamian, 2018]. In many plants, salt stress affects every aspect of plant physiology and metabolism

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[Parida and Das 2005, Marschner 2011, Acosta-Motos et al. 2017]. In arid regions, due to limited precipitation, underground (well) water is supplied as a common practice for agricultural food production.

The general character of underground water is higher salinity or electric conductivity (EC) levels. However, in Iran application of underground water in long term intensive cultivation for decades resulted in very high soil EC particularly under greenhouse cultivation with drip irrigation [Ahmadi and Souri 2018]. This higher soil EC is generally beyond threshold levels for many vegetable crops [Souri and Hatamian 2018]. This situation is probably the case for other parts of the world with climatic similarities. In such conditions, water or soil salinity is mainly due to NaCl that other cations and anions like calcium, potassium, magnesium, sulfate and nitrate may also present in relatively high levels [Navaroo et al. 2002, Kaya et al. 2003, Ahmadi and Souri 2018].

During previous decades, a considerable body of studies conducted to evaluate and improve plant salinity tolerance in many important crops [Waters et al. 2013, Piñero et al. 2016, Henschke 2017, Erdinc 2018]. However, most of these studies have applied NaCl to induce salinity, which can be different from the natural occurring salinity that plants are really exposed in the nature. Salinity influences many plant morphological and physiological traits including shoot and root biomass, leaf area and chlorophyll concentration, leaf water relations, and nutrients concentrations and their ratios [Kaya et al. 2002, Lycoskoufis et al. 2005, Komosa and Górnjak 2015, Acosta-Motos et al. 2017, Ahmadi and Souri 2018, Qiu et al. 2018].

Pepper is generally considered a sensitive plant to salinity, more sensitive than those thresholds of

tomato and cucumber. However, within a plant species, the salinity sensitivity (or tolerance) is cultivar dependant and significantly varies with plant growth stage. In many plants seed germination and/or seedling growth are more sensitive stages to salinity. In the present study, higher conductivity of nutrient solution via nitrate or chloride salts of potassium and calcium on growth characteristics of chili pepper (*Capsicum annuum* var. *annuum*) were evaluated that can give us useful information regarding cumulative effects of different ions and compounds as salinity agents.

MATERIALS AND METHODS

This study was conducted in 2017 under greenhouse conditions and at faculty of agricultural Sciences, Tarbiat Modares Uni., Tehran-Iran. The growth of chili pepper plants (*Capsicum annuum* var. *annuum*) were evaluated under application of chloride and nitrate salts of potassium and calcium to create a distinct EC of 5 or 8 dS/m of nutrient solution. Pepper seeds were germinated in sand, and seedlings at 4 leaf stage were used in experiment. Four liter black plastic pots were filled with a mix of perlite and cocopeat (30 : 70 v/v), in which two pepper plants were growing. Hoagland formula was used for preparation of basic nutrient solution in which it contains 15 mM N, 6 mM K, 5mM Ca, 2mM Mg, 1mM P, 46µM B, 9µM Mn, 6µM Fe, 1µM Zn, 0.32µM Cu and 0.11µM Mo. Higher electric conductivity of nutrient solution was performed via combination of different salts. The treatments were electric conductivity (EC) of 5 and 8 dS/m of nutrient solution created by KCl + CaCl₂, KNO₃ + CaNO₃ or KNO₃ + CaNO₃ + NaCl. In each combination treatment, equal amount (mM) was taken for each salt

Table 1. The composition and amounts of salts taken to induce EC of 5 or 8 dS/m of Hoagland nutrient solution

| Treatments | Hoagland | Stock solutions | | | | | |
|---|----------|---------------------------------------|-------------|--|-------------|---|------------|
| | | KCl + CaCl ₂ (each 1 M) | | KNO ₃ + CaNO ₃ (each 1 M) | | KNO ₃ + CaNO ₃ + NaCl (each 1 M) | |
| | | EC5 | EC8 | EC5 | EC8 | EC5 | EC8 |
| Control | × | | | | | | |
| KCl + CaCl ₂ | × | each 17.8mM | each 34.5mM | | | | |
| KNO ₃ + CaNO ₃ | × | | | each 16mM | each 37.5mM | | |
| KNO ₃ + CaNO ₃ + NaCl | × | | | | | each 12mM | each 24 mM |

from their 1 M stock solution. The amounts of applied salts for a given EC were presented in Table 1.

The treatments were arranged in completely randomized design with three replications in which each pot represented one replication contained 2 plants. After transplanting, seedlings were fed with normal nutrient solution for 10 days, thereafter different salinity treatments were applied to pots via nutrient solution. The experiment was extended for 4 months from 10 March until 10 June 2017 under greenhouse conditions of $28 \pm 5^\circ\text{C}$, relative humidity of 75–85% and light intercity of $250\text{--}300 \mu\text{mol m}^{-2} \text{s}^{-1}$.

Chili pepper fruits were harvested several times, and plants were final harvested 20 weeks after sowing. At harvest, the height of two plants per pot was measured using a tape and their average was recorded in the results. The average number of chlorotic leaves of two plants per pot was also recorded. Plants were cut at pot surface and roots were precisely separated and washed via tap water. Shoot and root fresh weight was measured using a precise digital scale. The cumulative fresh weight of fruits in 2–3 harvests was recorded per plant as fruit fresh weight (yield). Leaf nutrient concentrations were determined with different methods. Leaf nitrogen was determined using Kjeldahl method, phosphorus by molybdate-vanadate method and using spectrophotometer, potassium using Flame photometry, calcium, magnesium, iron, zinc, manganese and copper using atomic absorption spectrophotometer.

Leaf proline concentration was determined using ninhydrin method and the absorption of leaf extract samples were measured against different standard proline concentrations of 0, 5, 10, 20 and 40 mg L^{-1} and by spectrophotometer at 520 nm. Determination of catalase activity (CAT) was followed Pereira et al. [2002]. Briefly, the assay solution contained 50 mM phosphate buffer (pH 7.5) and 25 mM H_2O_2 . The process started with adding 0.1 mL of protein enzyme extract to combined reaction. CAT activity was assayed by monitoring a decrease in the absorbance of H_2O_2 within 1 min at 240 nm and 25°C . The amount of enzyme activity was expressed as unit μgPr . For determination of peroxidase activity 4 mM acetate buffer 0.4 M (pH 5), 4 mM H_2O_2 , 3% and 0.2 mL benzidine 2% in methanol 50% were added to each tube while placed in ice, and finally 0.2 mL of protein enzymatic extract was added

to each tube. The absorbance change was read at 530 nm, and the amount of enzyme activity was measured as unit μgPr -1 [Pereira et al. 2002]. Data were analyzed using SPSS Software, and comparison of means was performed using LSD test at 5% level.

RESULTS AND DISCUSSION

The results showed that application of various salts at EC5 or EC8 dS/m of nutrient solution had significant effect on all plant growth parameters. The tallest plants (Tab. 2) were obtained from $\text{KNO}_3 + \text{CaNO}_3$ at EC5; however, it had no difference with control plants. At EC5 only $\text{KCl} + \text{CaCl}_2$ treatment, and at EC8 plants treated with $\text{KCl} + \text{CaCl}_2$ or $\text{KNO}_3 + \text{CaNO}_3 + \text{NaCl}$ (EC8) showed significant shorter plants than control. Regarding plant height, $\text{KNO}_3 + \text{CaNO}_3$ at either EC5 or EC8 and $\text{KNO}_3 + \text{CaNO}_3 + \text{NaCl}$ at EC5 showed no difference with control plants. Application of $\text{KCl} + \text{CaCl}_2$ at EC8 resulted in highest number of chlorotic leaves (Tab. 2), whereas the lowest number of chlorotic leaves was in control plants and in those plants treated with $\text{KNO}_3 + \text{CaNO}_3$ at EC5.

Plant root fresh weight (Tab. 2) was significantly higher in $\text{KNO}_3 + \text{CaNO}_3 + \text{NaCl}$ at EC5 and lower in $\text{KCl} + \text{CaCl}_2$ at EC8 than control plants. Other salts at EC5 or EC8 showed no difference in root fresh weight than control. Application of $\text{KNO}_3 + \text{CaNO}_3$ at EC5 resulted in significantly higher shoot fresh weight (Tab. 2) than control plants. Shoot fresh weight was reduced by application of $\text{KCl} + \text{CaCl}_2$ or $\text{KNO}_3 + \text{CaNO}_3 + \text{NaCl}$ at EC8 of nutrient solution than control.

The cumulative fruit fresh yield of several harvest (Tab. 2) showed that all salinity treatments except $\text{KNO}_3 + \text{CaNO}_3$ at EC5 reduced fruit yield than control. At either EC5 or EC8 the most reduction in fruit yield occurred for plants treated with $\text{KCl} + \text{CaCl}_2$; however, the fruit yield reduction by this treatment at EC8 was higher than EC5.

Salinity generally reduces plant growth parameters and biomass production [Kaya et al. 2002, Parida and Das 2005, Acosta-Motos et al. 2017, Erdinc 2018]. The beneficial effects of $\text{KNO}_3 + \text{CaNO}_3$ at EC5 dS/m of nutrient solution is probably due to nutritional role of K, Ca and NO_3 as they are the major nutrients in plant structure and metabolic functions [Sagi et al. 1997, Kaya et al. 2003, Marschner 2011].

Table 2. Effects of higher ECs of nutrient solution on some vegetative parameters and fruit fresh weight (yield) of chili pepper plants

| | Plant height (cm) | No of chlorised leaves | Root FW (g) | Shoot FW (g) | Fruit FW (g) |
|--|-------------------|------------------------|-------------|--------------|--------------|
| Control | 91 ±5ab | 4 ±1c | 23.8 ±1.1bc | 98.2 ±8bc | 254 ±19a |
| KCl + CaCl ₂ (EC5) | 61 ±8c | 8.3 ±1.4b | 21.3 ±2.1cd | 87.5 ±11cd | 142 ±34c |
| KNO ₃ + CaNO ₃ (EC5) | 97 ±4a | 4 ±1c | 26.0 ±2.6ab | 122.1 ±16a | 225 ±27ab |
| KNO ₃ + CaNO ₃ + NaCl(EC5) | 76 ±6b | 6.7 ±1.6bc | 28.1 ±3.4a | 108 ±17ab | 193 ±30b |
| KCl + CaCl ₂ (EC8) | 46 ±6d | 11.3 ±1.5a | 19.1 ±0.8d | 72.6 ±6d | 69 ±18d |
| KNO ₃ + CaNO ₃ (EC8) | 82 ±3b | 6.7 ±1.2bc | 21.7 ±2.3cd | 84.2 ±12cde | 119 ±25c |
| KNO ₃ + CaNO ₃ + NaCl(EC8) | 57 ±3c | 8 ±2.6b | 21.4 ±1.8cd | 75.7 ±9d | 109 ±28cd |

In each treatment combination, amount of salts were taken in equal molarities
Plants were grown under higher EC treatments for 16 weeks
Comparison of means was performed at 5 % level of LSD test

Table 3. Effects of higher ECs of nutrient solution on leaf macronutrient concentrations of chili pepper plants

| | N (%DW) | K (%DW) | P (%DW) | Mg (%DW) | Ca (%DW) |
|--|--------------|------------|--------------|--------------|------------|
| Control | 2.9 ±0.36cde | 2.4 ±0.3ab | 0.40 ±0.06a | 0.53 ±0.09ab | 3.5 ±0.4bc |
| KCl + CaCl ₂ (EC5) | 2.7 ±0.26de | 2.5 ±0.4ab | 0.35 ±0.07ab | 0.46 ±0.07ab | 3.2 ±0.3bc |
| KNO ₃ + CaNO ₃ (EC5) | 4.3 ±0.55ab | 2.9 ±0.4a | 0.40 ±0.08a | 0.56 ±0.08a | 3.8 ±0.2ab |
| KNO ₃ + CaNO ₃ + NaCl(EC5) | 3.7 ±0.45bc | 2.5 ±0.3ab | 0.36 ±0.05ab | 0.51 ±0.08ab | 3.8 ±0.6ab |
| KCl + CaCl ₂ (EC8) | 2.4 ±0.36e | 2.2 ±0.2b | 0.26 ±0.08b | 0.27 ±0.03c | 3.0 ±0.4c |
| KNO ₃ + CaNO ₃ (EC8) | 4.7 ±1.1a | 3.0 ±0.5a | 0.40 ±0.06a | 0.41 ±0.06b | 4.4 ±0.9a |
| KNO ₃ + CaNO ₃ + NaCl(EC8) | 3.5 ±0.6bc | 2.7 ±0.3ab | 0.36 ±0.05ab | 0.42 ±0.05b | 3.8 ±0.2ab |

In each treatment combination, amount of salts were taken in equal molarities
Plants were grown under higher EC treatments for 16 weeks
Comparison of means was performed at 5 % level of LSD test

Table 4. Effects of higher ECs of nutrient solution on leaf micronutrient concentrations of chili pepper plants

| | Fe (mg kg ⁻¹ DW) | Zn (mg kg ⁻¹ DW) | Mn (mg kg ⁻¹ DW) | Cu (mg kg ⁻¹ DW) |
|--|-----------------------------|-----------------------------|-----------------------------|-----------------------------|
| Control | 69 ±12ab | 43 ±9ab | 78 ±9a | 9.8 ±1.7ab |
| KCl + CaCl ₂ (EC5) | 58 ±6bc | 41 ±6ab | 55 ±5bc | 9.1 ±1.1ab |
| KNO ₃ + CaNO ₃ (EC5) | 76 ±7a | 47 ±7a | 79 ±9a | 10.1 ±2.2a |
| KNO ₃ + CaNO ₃ + NaCl(EC5) | 64 ±6ab | 41 ±4ab | 68 ±11ab | 9.2 ±1.9ab |
| KCl + CaCl ₂ (EC8) | 48 ±9c | 37 ±5b | 50 ±10c | 5.6 ±1.2c |
| KNO ₃ + CaNO ₃ (EC8) | 65 ±8ab | 44 ±4ab | 57 ±8bc | 7 ±1.3bc |
| KNO ₃ + CaNO ₃ + NaCl(EC8) | 53 ±10c | 39 ±6ab | 56 ±8bc | 7.7 ±1.7bc |

In each treatment combination, amount of salts were taken in equal molarities
Plants were grown under higher EC treatments for 16 weeks
Comparison of means was performed at 5% level of LSD test

All components of this salt-making treatment are essential nutrients that their gradual uptake could benefit plant growth. Treatment of plants with KCl + CaCl₂ particularly at EC8 dS/m of nutrient solution resulted in the most reduction of root and shoot fresh weight. Similar effects were also obtained by other researchers with application of NaCl or KCl [Navarro et al. 2002, Borgognone, et al. 2014, Komosa and Górnjak 2015]. However, it has been reported that CaCl₂ has not negative effects on plant growth as KCl [Borghesi et al. 2013, Borgognone, et al. 2014]. Application of salinity (at EC 5.1 dS/m) via NaCl or KCl in nutrient solution resulted in reduced biomass production in artichoke and cardoon plants, whereas application of EC 5.1 dS/m via CaCl₂ recorded the same values as control [Borgognone et al. 2014]. Increasing root fresh weight in KNO₃ + CaNO₃ + NaCl at EC5 indicates stressful conditions for plants, whereas lower root fresh weight in KCl + CaCl₂ at EC8 indicates toxicity on plant roots, probably due to effects of chloride anions. The sensitivity of plants to high concentration of chloride has been also reported [Navarro et al. 2002, Komosa and Górnjak 2015]. Nitrate and chloride are the major mineral anions for osmotic adjustment in plants; however, chloride in high concentration can have toxicities on plant growth and nutrients uptake [Komosa and Górnjak 2015]. On the other hand, it has been shown that nitrogen can ameliorate the adverse effects of salinity on sweet pepper plants [Piñero et al. 2016].

Determination of leaf nutrient concentrations showed that macro- and micro-nutrients of leaves were changed under higher conductivity treatments of nutrient solution. Application of KNO₃ + CaNO₃ at either EC5 or EC8 significantly increased leaf N concentration (Tab. 3) than control plants, and this treatment at EC8 resulted in highest leaf N concentration. Nevertheless, there was no significant reduction in leaf N by salinity treatments. Application of salinity treatments at either EC5 or EC8 showed no significant changes in leaf K concentration than control plants (Tab. 3). However, application of KNO₃ + CaNO₃ at either EC5 or EC8 resulted in significantly higher leaf K than those plants treated with KCl + CaCl₂ at EC8.

Leaf phosphorus concentration (Tab. 3) was reduced only in KCl + CaCl₂ at EC8, whereas KNO₃ + CaNO₃ at either EC5 or EC8 had no difference with

control plants. The highest leaf magnesium concentration (Tab. 3) was in plants treated with KNO₃ + CaNO₃ at EC5 that showed no difference with control plants. On the other hand, application of KCl + CaCl₂ at EC8 significantly reduced leaf Mg concentration than control plants. Application of KNO₃ + CaNO₃ at EC8 increased leaf calcium concentration than control (Tab. 3), whereas other salinity treatments showed no difference with control plants.

The highest leaf iron concentration (Tab. 4) was in plants treated with KNO₃ + CaNO₃ at EC5, and the lowest amount was in plants treated with KCl + CaCl₂ or KNO₃ + CaNO₃ + NaCl at EC8 of nutrient solution that showed significant reduction than control plants. Leaf Zn concentration (Tab. 4) was not changed significantly by higher conductivity treatments than control. However, plants treated with KNO₃ + CaNO₃ at EC5 had higher leaf Zn than those plants treated with KCl + CaCl₂ at EC8 of nutrient solution. Leaf manganese concentration (Tab. 4) was highest in control plants and also in KNO₃ + CaNO₃ treatment at EC5. Meanwhile, KCl + CaCl₂ at EC5 and EC8, and KNO₃ + CaNO₃ + NaCl at EC8 had significantly less leaf Mn than control plants. Leaf copper concentration (Tab. 4) was highest in plants treated with KNO₃ + CaNO₃ at EC5 that showed no difference with control plants. Application of KCl + CaCl₂ at EC8 reduced leaf Cu concentration, whereas other higher conductivity treatments showed no significant difference compared to control plants.

Salinity is known to change the nutrient compositions and ratios in plant tissues resulting in nutrient imbalances [Parida and Das 2005, Marschner 2011, Erdinc 2018, Qiu et al. 2018]. It has been shown that high salinity induced by NaCl can significantly reduce nutrients concentration of leaves particularly cations such as Ca, K, Mg and Zn [Sagi et al. 1997, Kaya et al. 2003, Parida and Das 2005, Komosa and Górnjak 2015]. However, in the present study different salts other than NaCl were used in combination, and their effects were quite different from reported NaCl effects on plant growth and nutrient uptake. In some cases, salinity also increases the concentration of elements such as manganese, iron and zinc in the plant [Parida and Das 2005, Ahmadi and Souri 2018]. The concentration of N, P, K, S, Ca, Mg, B, and Fe in the shoots of dianthus plants increased, and molybdenum and zinc

concentrations decreased with increasing fertilizer concentration of nutrient solution [Kang et al. 2002]. It is well known that nutrients have synergetic and antagonistic interactions, particularly when some nutrients are applied in higher levels [Souri and Hatamian 2018]. However, such negative interactions could be less detrimental than NaCl induced toxicity and imbalances [Komosa and Górnjak 2015, Kaya et al. 2003, Kaya et al. 2002]. Accordingly, it was shown that higher application rate of potassium can significantly improve the growth characteristics and fruit quality of pepper [Mardanluo et al. 2018] and strawberry [Tohidloo et al. 2018] plants.

Leaf proline concentration (Fig. 1) was significantly increased by application of KCl + CaCl₂ and KNO₃ + CaNO₃ + NaCl at either EC5 or EC8 of nutrient solution than control plants. However, application of KNO₃ + CaNO₃ at either EC5 or EC8 showed no difference in leaf proline concentration than control. The highest leaf proline concentration was in those plants

treated with KCl + CaCl₂ at EC8 that showed no difference with KNO₃ + CaNO₃ + NaCl treatment at EC8. Increase in proline concentration due to salinity stress has been also reported in other studies [Kong-Ngern et al. 2012, Borgognone, et al. 2014, Ahmadi and Souri 2018]. Proline is a natural osmolite that accumulate in plant tissues particularly under various stresses conditions [Marschner 2011]. Salinity generally induces proline accumulation in leaves of affected plants to protect them towards neutralization of damages and higher tolerance to salinity [Kaya et al. 2003, Parida and Das 2005, Fageria et al. 2011]. On the other hand, exogenous application of proline has been shown to enhance growth of chilli pepper by improving physiological and biochemical attributes under salt stress [Butt et al. 2016].

Leaf catalase (Fig. 2) and peroxidase (Fig. 3) enzymes activity were significantly increased by application of KCl + CaCl₂ at either EC5 or EC8, and application of KNO₃ + CaNO₃ + NaCl at EC8 than

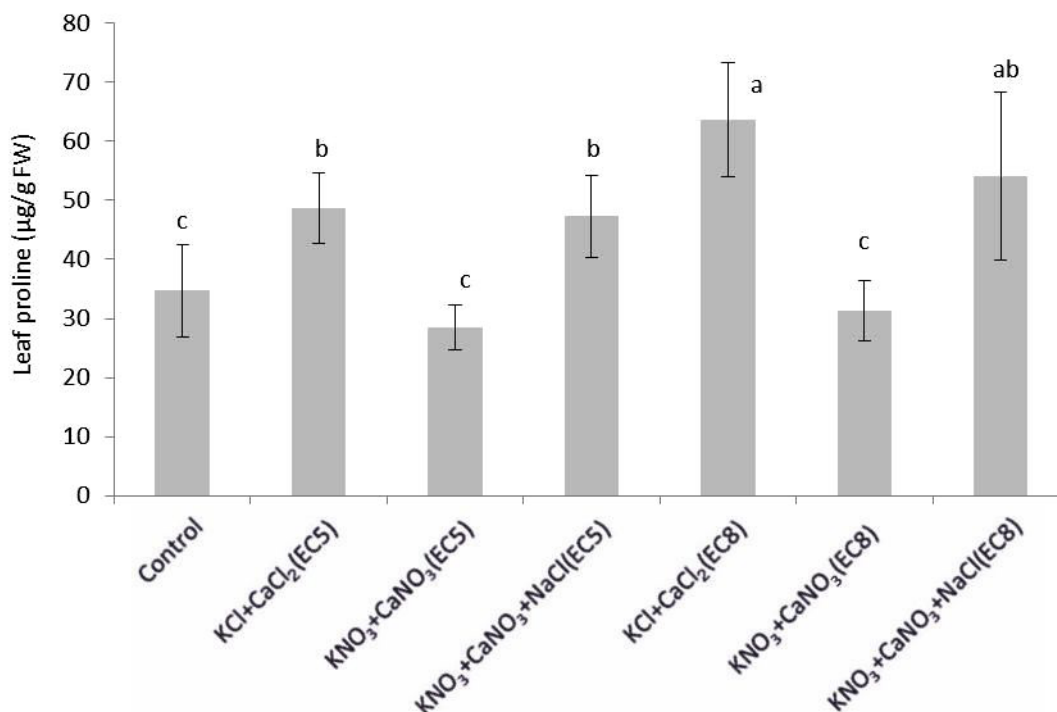


Fig. 1. Leaf proline concentration under application of higher EC of nutrient solution induced by various salts. Comparison of means was performed at 5 % level of LSD test

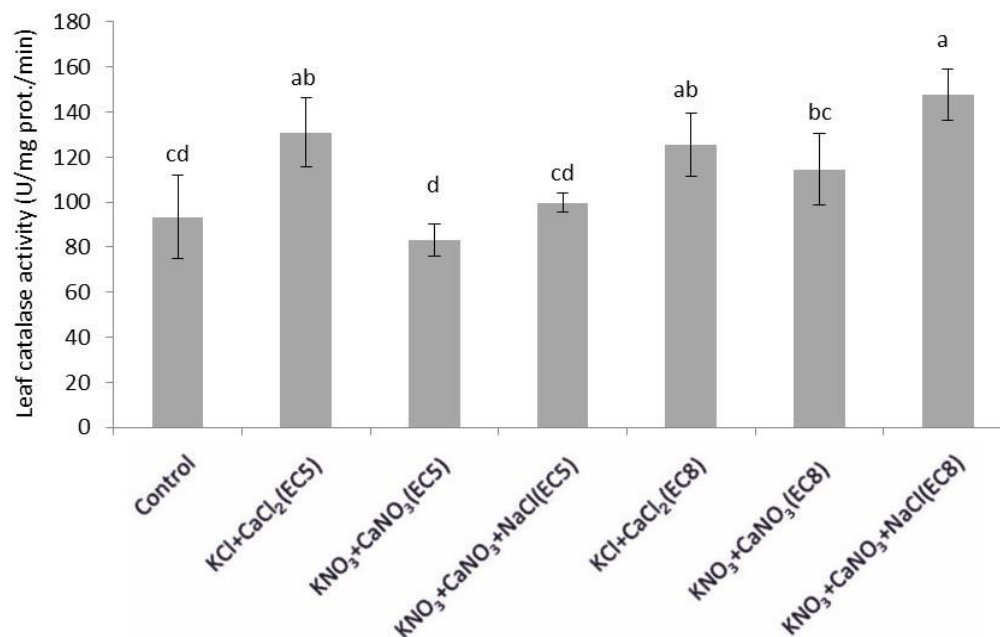


Fig. 2. Leaf catalase enzyme activity of chili pepper plants under application of higher EC of nutrient solution induced by various salts. Comparison of means was performed at 5 % level of LSD test

control plants. Other conductivity treatments showed no difference in catalase or peroxidase enzymes activity than control. Nevertheless, significant higher catalase activity was observed by application of $\text{KNO}_3 + \text{CaNO}_3$ at EC8 rather than EC5 of nutrient solution. Changes in antioxidant enzymes are a common response of plants to salinity [Marschner 2011, Fageria et al. 2011, Acosta-Motos et al. 2017, Erdinc 2018]. Catalase and peroxidase constitute the first defense system against oxidative damages arisen from environmental stresses [Kong-Ngern et al. 2012, Lim et al. 2015]. Salinity induces oxidative stress in plant tissues by peroxidation of membrane lipids and impairing the cellular electron transport within different subcellular compartments. This can lead to the generation of reactive oxygen species (ROS) such as singlet oxygen, superoxide anion, hydrogen peroxide and hydroxyl radicals [Sreenivasulu et al. 2000, Borgognone et al. 2014, Erdinc 2018]. Plants respond to oxidative stress by ROS scavenging through activation of the antioxidant systems, which includes both enzymatic and non-enzymatic defense mechanisms [Borgognone et al. 2014, Serrano et al. 2017].

High salinity conditions have been found to adversely affect several physiological processes and disrupt metabolic functions resulting in hindered plant growth and production. This is mainly due to reduction in cell enlargement and leaf area expansion, leaf chlorophyll concentration and photosynthesis rates, protein biosynthesis, osmotic and water relations [Marschner 2011, Fageria et al. 2011, Acosta-Motos et al. 2017]. However, in many horticultural crops lower level of salinity in hydroponic culture particularly by nutrient salts can improve plant growth and fruit quality [Borghesi et al. 2013, Borgognone, et al. 2014, Noreen et al. 2017, Mardanluo et al. 2018, Tohidloo et al. 2018]. Soil and water salinity can significantly restrict plant growth and vegetation biodiversity [Marschner 2011]. Salinity or higher EC of root medium reduces the absorption and translocation of nutrients in plants that can change the nutrients balances in plant tissues. This could also lead to occurrence of deficiency and toxicity of some nutrients. However, the most prominent effect of salinity is reduction in plant growth rate as presented in many studies [Serrano et al. 2017, Acosta-Motos et al. 2017, Qiu et al. 2018].

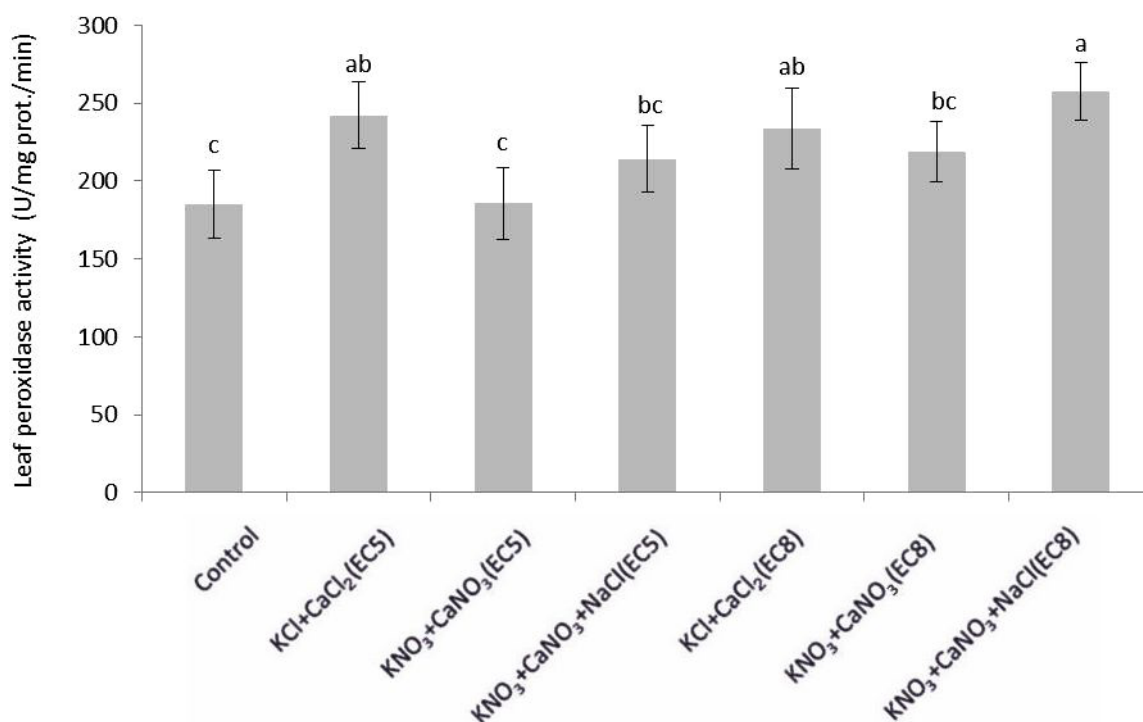


Fig. 3. Leaf peroxidase enzyme activity under application of higher EC of nutrient solution induced by various salts. Comparison of means was performed at 5 % level of LSD test

In the present study it was shown that higher EC of nutrient solution by nitrate salts of potassium and calcium was not detrimental as NaCl induced salinity stress on plants. In NaCl salinity stress, Na and Cl have cumulative adverse effects on plant growth and metabolic processes, whereas K, Ca and NO₃ in KNO₃ + CaNO₃ induced salinity, despite some marginal negative effects, had cumulative positive effects in our study compared to that reported with NaCl effects. Increasing salinity and N concentration in the growth medium can increase organic anions concentration in plants, and the amount of biomass was correlated positively with organic anion concentration in plants exposed to different salinity levels [Sagi et al. 1997]. Plant biomass increased with N concentration in the nutrient solution regardless of the applied salinity level. Higher N in root medium with salinity-stressed plants can result in higher inorganic and organic anions in plant leaves [Sagi et al. 1997]. The ameliorative effects of K, Ca or P application (in form of sulfate or nitrate) on plant growth have been shown that can restore the reduced value of growth parameters in

plants under salinity [Kaya et al. 2002, 2003, Bolat et al. 2006, Golcz et al. 2008, Ramadan and Shalaby 2016, Samson et al. 2016].

It has been shown that increasing nutrient solution strength or fertilization concentration can positively affect growth of some plant species [Barbosa et al. 2000, Kang et al. 2002, Alvaro et al. 2016], whereas it may reduce growth rate of some other plant species [Kang et al. 2002, Amalfitano et al. 2017]. Chrysanthemum plants produce the highest flower numbers per stem, stem length, and fresh and dry weights per plant with 2.5 times increase of potassium concentration of nutrient solution [Barbosa et al. 2000]. Similarly, pretreatment of chrysanthemum cut flower with nitrogen salts of potassium nitrate and calcium nitrate significantly improved flower shelf life and quality [Souri et al. 2018]. Salinity generally increases the total concentration of inorganic anions in plant tissues [Marschner 2011] and application of higher N can change the concentration gradient toward N other than Cl [Sagi et al. 1997]. Application of chloride form of potassium, calcium or sodium significantly reduced

plant growth. High chloride levels of nutrient solution can reduce tomato plant growth [Komosa and Górnica 2015]. However, supplementary calcium nitrate, potassium nitrate or their combination have resulted in generally highest growth parameters, generally similar values to control plants [Kaya et al. 2002, Kaya et al. 2003]. Chloride in high concentration has antagonism effects with N, Ca, S, and Zn, as their concentrations were declined with increasing Cl levels of nutrient solution [Komosa and Górnica 2015].

CONCLUSIONS

The results showed that application of various salts at EC5 or EC8 dS/m of nutrient solution had significant effects on all plant growth parameters. Creating higher EC of nutrient solution via chloride and nitrate salts of potassium and calcium had quite different effect on chili pepper growth. Vegetative growth traits, shoot and root fresh weight, fruit fresh yield and leaf nutrient concentrations were adversely affected by combination of chloride salts of potassium and calcium at either EC5 or particularly at EC8 dS/m nutrient solution. Accordingly, application of KCl + CaCl₂ and KNO₃ + CaNO₃ + NaCl particularly at EC8 of nutrient solution resulted in higher leaf proline concentration, and catalase and peroxidase enzymes activity than control. On the other hand, application of KNO₃ + CaNO₃ particularly at EC5 dS/m showed beneficial effects on many plant growth parameters. These findings indicate that various salts and their combinations may have different effects on plant. In nature, natural occurring salinity is a complex issue that many factors contribute in observed plant responses under such conditions. This is maybe the reason why plants such as tomato that its salinity tolerance threshold is about 2–3 dS/m, can normally grow and produce high yields and quality under some saline soil conditions of 9–13 dS/m (field and greenhouse soils) in some parts in Iran.

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