JOURNAL OF HORTICULTURE AND POSTHARVEST RESEARCH
2022, VOL. 5(4), 363-378

Journal of Horticulture and Postharvest Research

Journal homepage: www.jhpr.birjand.ac.ir

University of Birjand

Salinity tolerance evaluation of twelve selected pomegranate (*Punica granatum*) genotypes to achieve tolerant cultivars and rootstocks

Ali Momenpour*[1], Maryam Dehestani Ardakani[2], Mostafa Shirmardi[2], Jalal Gholamnezhad[2], Fatemeh Ahmadi[2] and Zahra Jamaati[2]

[1]National Salinity Research Center, Agricultural Research, Education and Extension Organization (AREEO), Yazd, Iran
[2]Department of Horticultural Science, College of Agriculture & Natural Resources, Ardakan University, Yazd, Iran

ARTICLE INFO

Original Article

Article history:
Received 21 March 2022
Revised 30 May 2022
Accepted 8 June 2022
Available online 12 November 2022

Keywords:
Morpho Physiological traits
'Chahafzal' and 'Poostsiah' genotypes
Rootstock
Soil salinity

DOI: 10.22077/jhpr.2022.5152.1270
P-ISSN: 2588-4883
E-ISSN: 2588-6169

***Corresponding author:**
[1]National Salinity Research Center, Agricultural Research, Education and Extension Organization (AREEO), Yazd, Iran.
Email: a.momenpour@areeo.ac.ir

ABSTRACT

Purpose: Pomegranate (*Punica granatum* L.) is a very interesting fruit tree for arid and semiarid areas in any part of the world. Like other fruit trees, the selection of tolerant rootstocks and scion is a very good strategy to reduce the adverse effects of salinity on pomegranate. Therefore, this study aimed to evaluate the effect of salinity stress on the growth characteristics of some selected pomegranate genotypes and introduce the most tolerant genotype(s) to salinity for use as a basis in future research. **Research method:** Selected pomegranate genotypes were evaluated using a factorial experiment based on a completely randomized design (CRD) with four replications in 2019-2020. Treatments were included 12 genotypes of Golanr-e-Shahdad (G-Shahdad), Golanr-e-Sarvestan(G-Sarvestan), Golnar-Saveh(G-Saveh), Poostsiah-e-Ardekan(Poostsiah), 'Malas-e-Yazdi'('M-Yazdi'), 'Malas-e-Saveh'('M-Saveh'), 'Shishecap-e-Ferdos' ('Shishecap'), 'Rabab-e-Neiriz' ('Rabab'), 'Vahshi-e-Babolsar'('V-Babolsar'), 'Narak-e-Lasjerd-Semnan'('Narak'), Chahafzal and 'Voshik-e-Torsh-e-Saravan'('Voshik') and the salinity of the irrigation water in five levels (1, 3, 5, 7 and 9 $dS.m^{-1}$). **Findings:** According to the results, the type of genotype and the level of salinity were affected on morphological and physiological traits as well as the concentration of nutrient elements. In all genotypes, the growth indices, relative water content (RWC), chlorophyll index, and total chlorophyll reduced as a result of increasing the salinity level. But the percentage of necrotic leaves, percentage of fallen leaves, ion leakage, concentration of Na^+, the concentration of Cl^- and Na^+/K^+ ratio increased. At salinity level of $7dS.m^{-1}$, necrotic leaves (3.11% &23.98%), fallen leaves (1.05% & 5.70%), ion leakage (5.87% & 22.10%), Na^+(0.31% & 1.29%), concentration of Cl^- (0.13%& 1.10%), concentration of K^+(0.64% & -0.07%) and Na^+/K^+ ratio (0.09 & 2.28 units) increased in Chahafzal and Voshik genotypes, respectively. **Research limitations:** No limitations to report. **Originality/Value:** 'Chahafzal' and 'Poostsiah' genotypes were recognized as the most tolerant to salinity according to the results. In contrast, Voshik and M-Saveh genotypes were more sensitive to salinity. The tolerant genotypes will be used in plans as rootstocks to graft the selected genotypes on them.

INTRODUCTION

Pomegranate (*Punica granatum* L.) is one of the oldest known edible fruits, among the seven kinds of fruit mentioned in the bible (Liu et al., 2018). Pomegranate is native to Iran and grows widely in arid and semi-arid regions of the world (Sarkhosh et al., 2006). Currently, most pomegranate cultivation is in arid and semi-arid regions of the world, where soil salinity and water stress are the main limitations of appropriate yield production. Under salinity stress, plant growth and development are affected negatively by water stress (*i.e.*, by lowering osmotic potential of soil solution and thus reducing water uptake) or by ionic stress (*i.e.*, by nutritional imbalance and/or toxicity), or the combination of the mentioned factors (Ashraf 1994; Marschner 1995; Ashraf & Harris, 2004; Silva-Ortega et al., 2008). Plants can adapt to increasing vacuolar Na^+ concentrations by maintaining their cellular ion homeostasis, accumulating osmotic-adjustment substances e.g., soluble sugars and amino acids; (Flowers & Colmer, 2015), and activating antioxidant systems able to scavenge reactive oxygen species ROS; (Pang & Wang, 2008), via the initiation of an efficient signal transduction network. Salinity is the most challenging factor in the pomegranate-producing regions in central Iran (Naeini et al., 2006a). Pomegranate is relatively tolerant to salinity and can be a good alternative to salinity-sensitive crops in case of increased salinity. However, similar to other plants, the degree of salinity resistance in pomegranate depends on the cultivar. Given the great variety of cultivars available for production, particularly in Iran, there is considerable potential for selecting salinity-resistant cultivars. Different degrees of salinity tolerance have been reported concerning macronutrient uptake, Na^+ and Cl^- distribution, and osmolyte production among the Iranian commercial cultivars (Karimi & Hasanpour, 2014; Khayyat et al., 2014).

The ‘Wonderful’ and ‘Ermioni’ cultivars of pomegranate have shown the ability to grow in marginal saline conditions (Dichala et al., 2021). In a study, the ‘Wonderful’ and ‘Parfianka’ pomegranate cultivars were recognized as tolerant to salinity. Ibrahim (2016) stated that salinity (in the range of 500-700 ppm) did not affect pomegranate growth of ‘Wonderful’ and ‘Manfalouty’ cultivars. The ‘Wonderful’ cultivar showed more tolerance to salinity than ‘Manfalouty’. Okhovatian-Ardakani et al. (2010) examined the salinity tolerance of 10 commercial pomegranate cultivars. The results showed that the best vegetative growth conditions were related to the ‘Post Siyah and ‘Tab -o- Larz’ cultivars at the salinity levels of 4 and 7 $dS.m^{-1}$, respectively.

Pomegranate (*Punica granatum* L.) is a very interesting fruit tree for arid and semiarid areas in any part of the world, because it adapts well to every type of soil and climate found in these regions (Parvizi et al., 2016). There are 760 pomegranate genotypes in a collection of the Agricultural and Natural Resources Research Center in Yazd–Iran. Some of these were collected from regions with water and soil salinity (Naeini et al., 2006b). On the other hand, like other fruit trees, the selection of tolerant rootstocks and scion is a very good strategy to reduce the adverse effects of salinity on pomegranate, particularly in arid areas of the country. Therefore, studying more cultivars and genotypes regarding their tolerance to salinity is necessary to introduce the most tolerant cultivars and genotypes to salinity. Thus, this study aimed to evaluate the effect of salinity stress on the growth characteristics of some selected pomegranate genotypes and introduce the most tolerant genotype(s) to salinity for use as a basis in future research.

MATERIALS AND METHODS

Plant material and salt treatment

To conduct this research, hardwood cuttings with a length of 27±3 cm and a diameter of 10 ± 1 mm were first prepared from the original plants in the collection of pomegranate genetic resources in the Agricultural and Natural Resources Research Center of Yazd Province in early February 2019. The cuttings were then placed in 3-indole butyric acid (IBA) solution at a concentration of 2500 mg L^{-1} for five seconds and cultured in plastic bags containing sand and rooted in the greenhouse. Then, the uniform rooted cuttings with the same size in terms of length and diameter were selected and replanted in 15-kg pots containing soil with loam texture in late April 2019 (Okhovatian-Ardakani et al., 2010), (Table 1). Salinity treatment started after adequate plant growth in late June (Table 2) and continued for three months (13 weeks) (Momenpour et al., 2018; Okhovatian-Ardakani et al., 2010).

Table 1. Physical and chemical properties of soil mixture

Title	Unit	Symbol	Value	Title	Unit	Symbol	Value
Saturation Point	(%)	S.P	38.05	Sand (%)	(%)	Sand	47
Field Capacity (%)	(%)	FC	26.30	Silt (%)	(%)	Silt	35
Permanent Wilting Point	(%)	PWP	13.50	Clay (%)	(%)	Clay	18
Salinity	(dSm^{-1})	EC	6.48*	Texture	-	Texture	Loam
Soil pH	-	pH	7.77	Potassium	mg.kg-1	$K_{avr.}$	227
Nitrogen	(%)	N	0.10	Phosphorus	mg.kg-1	$P_{avr.}$	14.49
Organic Carbon	(%)	O.C	1.01				

* Before transition plants to the pots, the used soil washed three times with fresh water (0.6 $dS.m^{-1}$) then the first soil EC decreased to lower than 1 $dS.m^{-1}$.

Table 2. Growth status of studied genotypes at beginning of the experiment

Cultivar	Number of leaves	Number of shoots	Diameter of main Shoot (mm)	Height of main shoot (mm)
'Shishecap-e-Ferdos' ('Shishecap')	62.30	1.60	2.61	28.5
'Rabab-e-Neiriz' ('Rabab')	64.05	1.80	2.27	27.95
'Malas-e-Saveh' ('M-Saveh')	70.00	1.45	2.53	29.71
'Malas-e-Yazdi' ('M-Yazdi')	74.70	1.60	2.77	30.39
Golnar-e-Zinati-Sarvestan (G-Sarvestan)	47.70	2.00	1.95	22.93
Voshik-e-torsh-e-Saravas (Voshik)	59.15	3.60	2.38	27.44
Golnar-e-Shahdad (G-Shahdad)	66.35	3.35	2.10	20.97
Golnar-e-Saveh (G-Saveh)	36.40	2.80	2.05	16.49
Chah Afzal	74.20	3.20	2.85	33.80
Narak-e-Lasjerd-e-Semnan (Narak)	38.00	3.35	2.87	23.78
Vahshi-e-Babolsar (V-Babolsar)	89.10	2.70	2.58	17.05
Poost- Siah-e-Ardakan (Poostsiah)	46.70	2.00	2.92	22.85

Table 3. Qualitative characteristics of the used water after dilution by tap water by 1:20 ratio

Electrical Conductivity ($dS.m^{-1}$)	pH	Na (mg. L^{-1})	Cl (mg. L^{-1})	Ca (mg. L^{-1})	Mg (mg. L^{-1})	HCO_3^- (mg. L^{-1})
25.10	7.91	211.3	223.11	22.05	29.52	2.77

Table 4. ECe and pH of the used soil mixture in pots after applying different levels of salinity stress

Different levels of water salinity ($dS.m^{-1}$)	Soil pH	Soil Salinity ($dS.m^{-1}$)
1	7.57	1.51
3	7.56	3.77
5	7.60	6.15
7	7.69	9.29
9	7.77	12.59

For salinity treatments, the highly saline water of the Aqda region was used. Table 3 shows the composition of the water used in treatments. Also, salt was added gradually and reached the final concentration within a week to avoid sudden shock and plasmolysis. Accordingly, the plants were first irrigated with treatments of 1, 3, and 5 $dS.m^{-1}$. Then irrigation of the plants was carried out with the treatment of 7 $dS.m^{-1}$ in the second step, followed by the treatment of 9 $dS.m^{-1}$ in the third step to apply the salinity treatments of 7 and 9 $dS.m^{-1}$. Field capacity (FC) of soil in pots was determined before transferring plants to units by a pressure plate (Model F1, make USA). The irrigation schedule was organized according to pots' changes in weight and leaching requirements. Electric conductivity and pH rate were regularly measured in drainage water to maintain the electric conductivity of both input and soil solutions in a stable range. At the end of the experiment, the soil of pots in each level salinity was mixed. Then three samples of each treatment (in total 15 samples) were analyzed (Table 4).

This research was carried out as a factorial experiment based on a completely randomized design (CRD), with two factors; genotypes in 12 levels (including: Golanr-e-Shahdad (G-Shahdad), Golanr-e-Sarvestan (G-Sarvestan), Golnar-Saveh (G-Saveh), Poostsiah-e-Ardekan (Poostsiah), 'Malas-e-Yazdi' ('M-Yazdi'), 'Malas-e-Saveh' ('M-Saveh'), 'Shishecap-e-Ferdos' ('Shishecap'), 'Rabab-e-Neiriz' ('Rabab'), Vahshi-e-Babolsar (V-Babolsar), Narak-e-Lasjerd-Semnan (Narak), Chahafzal, and Voshik-e-Torsh-e-Saravan (Voshik) genotypes and irrigation water salinity in 5 levels (1, 3, 5, 7 and 9 dS/m) and with 4 replications in National Salinity Research Center in 2019. Overall, this research was performed with 240 pots.

Evaluation of plant parameters

Plants height was measured before the application of the salinity treatment to record the increased height, the number of green leaves, and the number of branches. The studied traits were recalculated at the end of the experiment, and the increased values were calculated (Momenpour et al., 2018).

The number of necrotic leaves was counted and divided by the number of total leaves to measure the percentage of necrotic leaves at the end of the experiment. Also, the number of fallen leaves was recorded until the end of the experiment and divided by the number of total leaves to measure the percentage of leaf abscission. The percentage of green leaves of plants was calculated by subtracting the percentage of total leaves from the percentage of fallen leaves, and the percentage of necrotic leaves (Momenpour & Imani, 2018).

After 13 weeks of salinity treatments, for determination of leaf chlorophyll, 0.2 g of the leaf was extracted (in total 240 samples, means 4 replicas for each cultivar and each salinity treatment), with ethanol 80% and chlorophyll *a*, chlorophyll *b* and total chlorophyll content were calculated with the method described by Arnon (1949). The resulting supernatant solution was used to measure chlorophyll. The light absorption was measured at 663 and 645 nm wavelengths for chlorophyll a and b, respectively, using a spectrophotometer (DR2000) (Arnon, 1949). Leaf chlorophyll index was measured on the same leaves using a SPAD (Minolta, 502, made in Japan) after 91 days since treatments began.

Leaf relative water content (RWC) was determined with nine replicas (made by four leaves each) for each treatment and each cultivar, for a total of 630 samples. Fresh weight (Fw) was recorded and then samples were put into distilled water and kept at 4°C for 24 h in the dark. After the emission of extra humidity, samples were weighed again to obtain the Total weight (Tw). Subsequently, samples were kept in the oven at 105°C for 24 hours and Dry weight (Dw) was recorded. Finally, relative water content was calculated via formulae (1) (Yamasaki & Dillenburg, 1999).

$$RWC= [(Fw-Dw) / (Tw-Dw)] \times 100 \quad (1)$$

To measure the relative ion leakage, 0.5 g of the leaves of each cultivar were weighed separately and placed into glass vials, after which 25 ml of distilled water was added to them. The samples were placed in a shaker for 24 h at a temperature of 24 ° C and a speed of 120 rpm. After 24 h, the initial electrical conductivity (Lt) was measured by a conductivity meter (Metrohm 644). The samples were autoclaved for 20 min and then kept at 25°C for 2 h on a shaker with a speed of 120 in/min. Their final electrical conductivity (Lo) was measured and ultimately the percentage of ion leakage was calculated according to the formula (LT/ LO) ×100 (Lutts et al., 1995).

The concentration of Na^+ and K^+ in leaves was measured with four replicas for each treatment and each genotype, for a total of 240 samples. The leaves were separated at the end of the experiment and placed in an oven at a temperature of 75 ° C for 48 h after thorough washing, and then milled to a fine powder to pass through a 30-mesh screen. The amount of 0.5 g of each sample was dry-ached for 6h at 550°C, dissolved in 3 mL of 6 mol L^{-1} HCl and diluted to 50 mL with deionized water. Subsequently, the concentration of Na^+ and K^+ were determined using atomic absorption spectroscopy (Papadakis et al., 2007). To measure chlorine, 0.1 g of the dried leaves in the oven was weighed using a digital scale with an accuracy of 0.001 g and then transferred to a 50 ml Erlenmeyer flask. Then 25 ml of boiling distilled water was added to the samples, after which they were placed on a shaker at 120 rpm for one hour. The extracts were completely filtered in several steps, and their volume was increased by distilled water. In the next step, 4 drops of potassium dichromate were added to 10 ml of the extracts and titrated with 0.05 N silver nitrate solution until a brick red color emerged. The amount of silver nitrate consumed for the samples was recorded, and the percentage of chlorine was calculated using the following formula (Equation 2) (Karimi & Hasanpour, 2014).

$$\text{chlorine percentage} = \frac{\text{silver nitrate used} \times \text{silver nitrate} \times 35.5 \times 100 \times \text{total volume}}{\text{sample weight} \times \text{extract volume} \times 100} \quad (2)$$

This experiment was conducted as a factorial experiment based on a completely randomized design (CRD), with two factors; genotypes at 12 levels and irrigation water salinity at 5 levels, and with four replications for each treatment in National Salinity Research Center, Yazd, Iran during 2019. Finally, data were analyzed using analysis of variance (ANOVA) using SAS software (version 9.1) (SAS Institute, Cary, NC, USA). Means were also compared by Duncan's Multiple Range test (MRT) at 1% level as well as MSTATC (Michigan State University, East Lansing, MI, USA) software (version 2.10).

RESULTS

Morphological parameters

Plant height and number of leaves

As reported in Table 5, salinity treatments negatively affected plant height and the number of leaves. With increasing salinity concentration in irrigation water, the final height and number of leaves in all studied cultivars were decreased. Reduction of the height of the main branch was significant at salinity level of 5 $dS.m^{-1}$for the 'M-Yazdi' cultivar as well as G-Sarvestan and G-Shahdad genotypes, and at salinity level of 7 $dS.m^{-1}$ for the 'Shishecap' cultivar and Chahafzal, Narak, V-Babolsar, and Poostsiah genotypes. However, this reduction was only significant for the 'Rabab' cultivar at the salinity level of 9 $dS.m^{-1}$ compared to the control

plants (Table 5). Although all genotypes showed a significant reduction in the height of the main branch at the salinity level of 9 $dS.m^{-1}$ compared to control plants, the height of the main branch increased by 10.81 cm in the Chahafzal genotype at the salinity level of 9 $dS.m^{-1}$. However, this increase was only 2.10 cm for the Voshik genotype at the same salinity level.

Necrotic, fallen and green leaves

According to the results, the effect of salinity and genotype interaction on the percentage of necrotic, fallen and green leaves was significant ($p < 0.01$) (Table 6). The results showed that with increasing salinity of irrigation water, the percentage of green leaves in all genotypes was decreased. In control plants and also plants treated with salinity level of 3 $dS.m^{-1}$, all leaves plants were green and weren't observed any necrotic leaves (Table 6). At the salinity level of 9 $dS.m^{-1}$, the percentage of necrotic and fallen leaves in the Voshik genotype was 13.87 and 35.47%, respectively. Accordingly, only 50.66% of the leaves were green at the end of the experiment. The highest percentage of green leaves at salinity levels of 7 and 9 $dS.m^{-1}$ (95.94% & 93.77%) was observed in the Chahafzal genotype (Table 6). The percentage of green leaves in this genotype showed a significant reduction compared to control plants only at the salinity level of 9 $dS.m^{-1}$ (Table 6). Also, the highest increase in the number of leaves was obtained in the Chahafzal genotype at the salinity level of 9 $dS.m^{-1}$ (Table 6). Given the physical damage, the Chahafzal genotype had better conditions compared to the other genotypes under study and the rate of leaf abscission was not significant at high salinity levels compared to control plants.

Physiological parameters

Chlorophyll index, total chlorophyll, ion leakage and relative water content

Results on chlorophyll index, total chlorophyll, percentage of ion leakage, and relative water content of leaves treated in different salinity levels are reported in Table 6. The relative water content of the leaves decreased significantly with salinity stress. The relative water content of the leaves decreased from 90.72% in the leaves of the Chahafzal genotype in control plants to 47.07% in the leaves of the Voshik genotype treated with the salinity of 9 $dS.m^{-1}$. Overall, the greatest decrease in relative water content was observed in the 'Voshik' genotype and 'M-Saveh' cultivar, respectively. The Chahafzal and Posstsiah genotypes showed the least reduction in the relative water content of the leaves.

Based on the results, the percentage of relative ion leakage increased in the leaves of all studied genotypes with increasing salt concentration. The increase in the percentage of relative ion leakage showed a significant difference between the studied genotypes. The highest percentage of relative ion leakage was observed in the leaves of the Voshik genotype under the treatment of 9 $dS.m^{-1}$ (61.20%). The percentage of relative ion leakage increased significantly in the leaves of the Voshik genotype and 'M-Saveh' and 'Rabab' cultivars at the salinity levels of 5, 7 and 9 $dS.m^{-1}$ compared to control plants. However, the increase in the relative ion leakage of the leaves was only significant at the salinity levels of 7 and 9 $dS.m^{-1}$ in other studied genotypes compared to control plants (Table 6).

According to Table 7, the total chlorophyll content and chlorophyll index decreased in the leaves of the studied genotypes with increasing salinity levels. Total chlorophyll content and chlorophyll index decreased significantly in the Voshik, G-Saveh, and G-Sarvestan genotypes and 'M-Saveh' cultivar compared to control plants with increasing salinity levels to 5 $dS.m^{-1}$ and more. However, the decrease in total chlorophyll content and chlorophyll index was only significant at salinity levels of 7 and 9 $dS.m^{-1}$ for the 'Rabab' and 'Shishecap' cultivars and Narak, V-Babolsar, and Poostsiah genotypes, and at the salinity level of 9 $dS.m^{-1}$ for the 'M-Yazdi' cultivar as well as Chahafzal and G-Shahdad genotypes compared to control plants.

Table 5. Interaction effect of salinity and genotype on some morphological traits

Genotype	Salinity Level (dS. m^{-1})	Number of leaves	Rate of height increase (cm)	Branch height (cm)	Green leaves (%)
Pr > F*	-	0.0451>	0.0001>	0.0001>	0.0001>
'Shishecap'	1	202.25 c-h	11.25 c-h	42.00 d-g	100.00 a
'Shishecap'	3	203.20 c-g	11.17 c-h	41.30d-h	100.00 a
'Shishecap'	5	195.20 c-h	11.07 d-i	38.85g-j	96.29 b-e
'Shishecap'	7	168.75 d-j	8.79 j-t	36.67 i-k	85.95 jk
'Shishecap'	9	165.00 e-j	7.65 p-w	33.02 m-q	78.72 l
'Rabab'	1	193.50 c-i	10.75d-k	38.82 g-j	100.00 a
'Rabab'	3	192.00 c-i	10.65d-k	38.87 g-j	100.00 a
'Rabab'	5	187.75 c-i	10.03g-m	39.52 f-i	95.52 c-e
'Rabab'	7	169.50 d-j	7.87 n-w	36.55 i-l	90.57 hi
'Rabab'	9	154.50 e-j	6.01 w-z	32.37 n-r	85.95 jk
M-Saveh	1	184.50 c-i	12.42 b-d	43.95 b-d	100.00 a
M-Saveh	3	183.75 c-j	11.40c-g	44.12 b-d	99.66 a
M-Saveh	5	174.50 c-j	9.52 g-p	40.47 e-h	94.41 d-f
M-Saveh	7	146.00 d-j	6.85 s-x	34.67 k-p	85.53 k
M-Saveh	9	116.10 j	4.00 a-	29.52 r-v	57.77 n
'M-Yazdi'	1	208.00 c-g	12.07c-f	43.65 b-e	100.00 a
'M-Yazdi'	3	209.10 c-f	12.13c-e	45.22 bc	100.00 a
'M-Yazdi'	5	207.20 c-g	10.97d-i	42.37 c-f	98.32 a-c
'M-Yazdi'	7	192.50 c-i	9.75g-o	38.63 h-j	91.32 g-i
'M-Yazdi'	9	173.00 d-j	7.85 o-w	35.12 k-o	88.74 ij
G-Sarvestan	1	160.75 d-j	9.95 g-m	35.85 j-n	100.00 a
G-Sarvestan	3	163.00 d-j	9.90 g-m	35.12 k-o	100.00 a
G-Sarvestan	5	155.00 d-j	8.70 k-t	33.02m-q	96.49 b-e
G-Sarvestan	7	133.00 f-j	7.25 q-w	28.70 t-x	86.98 jk
G-Sarvestan	9	115.00 j	4.70y-a/	22.55 z/	79.37 l
'Voshik'	1	188.25 c-i	13.07bc	43.35 b-e	100.00 a
'Voshik'	3	191.20 c-i	12.32b-d	42.37 c-f	100.00 a
'Voshik'	5	179.20 c-j	9.26 h-q	36.32 i-m	91.25 g-i
'Voshik'	7	148.50 d-j	6.45 u-y	31.85 o-t	70.32 m
'Voshik'	9	105.75 j	2.10 b/	26.50 v-y	50.66 o
G-Shahdad	1	185.75 c-i	8.90 j-r	31.67 p-t	100.00 a
G-Shahdad	3	185.25 c-i	9.30 h-p	32.10 o-s	100.00 a
G-Shahdad	5	183.75 c-j	6.87 r-x	27.52 t-x	98.70 ab
G-Shahdad	7	178.25 c-j	4.28 za/	24.05 yz	93.99 e-g
G-Shahdad	9	151.75 d-j	1.86 b/	20.67 za/	80.65 l
G-Saveh	1	168.00 d-j	8.30 l-u	26.20 v-y	100.00 a
G-Saveh	3	168.75 d-j	8.30 l-u	26.25v-y	100.00 a
G-Saveh	5	165.00 d-j	6.02 w-z	22.50 za/	97.50 a-c
G-Saveh	7	154.25 d-j	4.20 za/	19.52b/	92.21 f-h
G-Saveh	9	124.75 h-j	2.17 b/	16.95 c/	79.97 l
Chahafzal	1	219.25 b-d	14.90 a	48.85 a	100.00 a
Chahafzal	3	221.75 b-d	14.92 a	49.95 a	100.00 a
Chahafzal	5	218.50 b-d	14.09 ab	47.96 ab	97.72 a-c
Chahafzal	7	213.50 c-e	12.08 c-f	45.75 a-c	95.84 b-e
Chahafzal	9	199.25 c-h	10.81 d-j	42.28 b-f	93.77 e-g
Narak	1	160.70 d-j	10.17 e-l	36.12 i-n	100.00 a
Narak	3	163.20 d-j	10.07 f-m	36.45 i-m	100.00 a
Narak	5	157.00 d-j	9.09 i-r	33.12 l-q	97.37 a-d
Narak	7	151.50 d-j	7.15 r-w	29.02 q-v	91.85 f-h
Narak	9	139.40 e-j	4.98 x-a/	25.65 w-z	86.19 jk
V-Babolsar	1	286.75 ab	8.32 l-u	26.10w-z	100.00 a
V-Babolsar	3	290.70 a	8.70 k-t	26.47 v-y	100.00 a
V-Babolsar	5	285.00 ab	8.05 m-v	25.30 v-y	97.72 a-c
V-Babolsar	7	251.70 a-c	6.1 v-z	23.20 x-z	89.72 hi
V-Babolsar	9	210.30 c-f	4.26 za/	20.01 a/	81.41 l
Poostsiah	1	154.25 d-j	10.10 f-m	33.97 k-p	100.00 a
Poostsiah	3	156.25 d-j	10.22 e-l	34.72 k-p	100.00 a
Poostsiah	5	152.54 d-j	9.78 g-o	33.52 k-q	96.97 a-d
Poostsiah	7	146.70 d-j	8.83 j-s	30.53 p-t	92.53 f-h
Poostsiah	9	130.50 g-j	6.77 t-x	26.55 v-y	91.12 g-i

Means in each column and for each trait, followed by similar letter(s) are not significantly different at 5% or 1% probability levels, using Duncan's Multiple Range Test.

*= a/ is less than z. Given that variety of data was very wide, Duncan's test grouped data between a to z and less than z such as a/, b/ and c/

Table 6. Interaction effect of salinity stress and genotype on some of the morphological traits and physical damage

Genotype	Salinity Level (dS. m^{-1})	Fallen leaves (%)	Necrotic leaves (%)	Ion leakage	RWC (%)	Total chlorophyll (mg.g)
Pr > F*	-	0.0001>	0.0001>	0.0001>	0.0001>	0.0001>
'Shishecap'	1	0.00 o	0.00 p	19.20 r-t	86.43 a-e	0.71 st
'Shishecap'	3	0.00 o	0.00 p	19.52 q-t	87.62 a-d	0.70 st
'Shishecap'	5	0.76 l-o	2.94 k-p	21.40 p-s	84.98 a-i	0.65 t
'Shishecap'	7	2.10 h-l	11.94 de	29.72 hi	70.00 m	0.57 u
'Shishecap'	9	7.26 c	14.01 cd	44.40 c	55.27 n	0.48 v
'Rabab'	1	0.00 o	0.00 p	18.05 t	87.61 a-d	1.03 g-j
'Rabab'	3	0.00 o	0.00 p	18.60 st	87.32 a-d	1.04 g-i
'Rabab'	5	1.34 j-o	3.14 k-o	22.60 m-p	81.35 d-j	1.01 h-j
'Rabab'	7	3.38 fg	5.60 i-k	26.65 jk	77.84 i-k	0.92 l-n
'Rabab'	9	6.85 cd	7.20 g-j	32.45 g	71.04 lm	0.78 qr
M-Saveh	1	0.00 o	0.00 p	17.57 t	88.21 a-d	1.04 g-i
M-Saveh	3	0.00 o	0.24 op	17.97 t	88.61 a-d	0.99 i-k
M-Saveh	5	1.89 h-m	3.69 k-n	22.87 l-p	78.87 f-k	0.86 n-p
M-Saveh	7	5.17 e	9.29 fg	31.10 gh	69.85 m	0.71 st
M-Saveh	9	8.74 b	33.48 a	50.37 b	50.32 no	0.51 v
'M-Yazdi'	1	0.00 o	0.00 p	18.10 t	88.68 a-d	1.01 h-j
'M-Yazdi'	3	0.00 o	0.00 p	18.15 t	89.43 a-c	1.02 h-j
'M-Yazdi'	5	0.83 l-o	0.84 m-p	19.02 r-t	85.45 a-h	0.97 j-l
'M-Yazdi'	7	1.93 h-m	6.74 g-j	26.00 jk	73.83 k-m	0.87 n-p
'M-Yazdi'	9	2.40 h-k	8.84 f-h	35.57 f	70.20 m	0.75 rs
G-Sarvestan	1	0.00 o	0.00 p	17.15 t	91.30 a	1.06 e-h
G-Sarvestan	3	0.00 o	0.00 p	17.65 t	90.00 ab	1.04 g-i
G-Sarvestan	5	0.93 l-o	2.58 l-p	18.60 st	84.80 a-i	0.97 j-l
G-Sarvestan	7	2.72 g-j	10.30 ef	27.95 ij	72.72 k-m	0.89 m-o
G-Sarvestan	9	5.76 de	14.87 c	41.17d	66.75 m	0.69 st
'Voshik'	1	0.00 o	0.00 p	17.37 t	88.52 a-d	0.97 j-l
'Voshik'	3	0.00 o	0.00 p	18.32 t	88.80 a-d	0.92 l-n
'Voshik'	5	1.71 no	7.04 g-j	24.75 k-n	79.50 e-k	0.84 o-q
'Voshik'	7	5.70 h-k	23.98 b	35.42 f	66.82 m	0.68 t
'Voshik'	9	13.87 c	35.47 a	61.20 a	47.07 o	0.49 v
G-Shahdad	1	0.00 o	0.00 p	18.62 st	87.50 a-d	1.05 f-i
G-Shahdad	3	0.00 o	0.00 p	18.57 st	87.68 a-d	1.06 e-h
G-Shahdad	5	0.35 no	0.93 m-p	18.95 st	84.07 a-g	1.02 h-j
G-Shahdad	7	0.53 m-o	5.47 jk	24.17 k-o	77.25 j-l	0.94 k-m
G-Shahdad	9	4.52 ef	14.82 c	28.02 ij	70.57 lm	0.83 o-q
G-Saveh	1	0.00 o	0.00 p	19.10 r-t	88.19 a-d	1.00 h-k
G-Saveh	3	0.00 o	0.00 p	19.50 q-t	88.75 a-d	0.99 i-k
G-Saveh	5	0.35 no	2.13 m-p	21.20 p-s	84.10 a-j	0.91 l-n
G-Saveh	7	2.41 h-k	5.37 jk	28.05 ij	78.42 h-k	0.82 pq
G-Saveh	9	7.24 c	12.78 c	39.17 de	68.27 m	0.67 t
Chahafzal	1	0.00 o	0.00 p	17.87 t	90.72 a	1.17 a-c
Chahafzal	3	0.00 o	0.00 p	17.90 t	90.56 a	1.19 a
Chahafzal	5	0.00 o	2.28 l-p	18.10 t	88.12 a-d	1.15 a-d
Chahafzal	7	1.05 k-o	3.11 k-o	21.82 o-r	86.04 a-g	1.12 b-e
Chahafzal	9	1.13 k-o	5.10 j-l	25.20 k-m	82.13 c-j	1.03 g-j
Narak	1	0.00 o	0.00 p	19.37 r-t	89.70 a-c	1.12 b-e
Narak	3	0.00 o	0.00 p	19.20 r-t	90.00 ab	1.11 c-f
Narak	5	0.00 o	2.62 l-p	21.40 p-s	86.12 a-f	1.09 d-g
Narak	7	1.73 i-o	6.42 h-j	25.25 k-m	82.50 b-j	1.02 h-j
Narak	9	3.08 g-i	10.74 ef	30.27 g-i	78.73 g-k	0.94 k-m
V-Babolsar	1	0.00 o	0.00 p	19.60 q-t	88.25 a-d	1.04 g-i
V-Babolsar	3	0.00 o	0.00 p	19.40 r-t	86.34 a-e	1.03 g-j
V-Babolsar	5	0.36 no	1.92 m-p	22.20 m-q	85.00 a-i	0.98 i-l
V-Babolsar	7	2.04 h-l	8.24 f-i	29.88 hi	78.78 g-k	0.83 o-q
V-Babolsar	9	3.27 f-h	15.32 c	38.00 e	73.28 k-m	0.65 t
Poostsiah	1	0.00 o	0.00 p	18.10 t	90.53 a	1.17 a-c
Poostsiah	3	0.00 o	0.00 p	18.32 t	90.23 a	1.18 ab
Poostsiah	5	0.67 l-o	2.36 l-p	19.15 r-t	89.50 a-c	1.12 b-e
Poostsiah	7	1.92 h-m	5.55 i-k	22.80 l-p	86.60 a-e	1.09 d-g
Poostsiah	9	2.01 h-l	6.87 g-j	25.35 kl	82.17 c-j	1.02 h-j

Means in each column and for each trait, followed by different letter(s) are significantly different at 5% probability level, using Duncan's Multiple Range Test.

Nutrient content

Results reported in Table 7 assessed that with increasing salinity concentration in irrigation water, the sodium concentration in the leaves of total genotypes increased (Table 7). A comparison of the mean of the data showed that Na^+ increased significantly as a result of increasing salinity concentration. Based on the results, the highest concentration of Na^+ was observed in the leaves of the Voshik genotype (2.72%) at the salinity level of 9 $dS.m^{-1}$. In contrast, the lowest concentration of Na^+ was observed in the leaves of the Chahafzal genotype (0.90%) at the same salinity level. Na^+ concentration increased significantly in the leaves of the Chahafzal and Poostsiah genotypes and 'M-Yazdi' cultivar compared to control plants at the salinity levels of 7 and 9 $dS.m^{-1}$. However, Na^+ concentration increased significantly in the leaves of other studied genotypes at the salinity levels of 3, 5 and 7 $dS.m^{-1}$ compared to control plants (Table 7).

Based on the results, the studied genotypes showed different reactions in response to salinity stress. K^+ concentration in the leaves of the Chahafzal and Poostsiah genotypes and 'Rabab' and 'M-Yazdi' cultivars increased significantly compared to control plants up to the salinity level of 9 $dS.m^{-1}$. However, K^+ concentration in the leaves of the Narak and Shishecap genotypes increased significantly up to the salinity level of 7 $dS.m^{-1}$. Potassium content in the leaves of G-Sarvestan, G-Shahdad, G-Saveh, and V-Babolsar genotypes, was increased in salinity level 5 $dS.m^{-1}$. Then, with increasing salinity, potassium content in leaves, decreased.

The increase of the K^+ content in the leaves of the Voshik and M-Saveh genotypes was not significant compared to control plants. These results show that the Chahafzal and Poostsiah genotypes and 'Rabab' and 'M-Yazdi' cultivars can tolerate the negative and adverse effects of Na^+ more than other genotypes by increasing the amount of K^+ at higher salinity levels (Table 7). These results were consistent with the results of morphological traits. The lowest percentage of necrosis and fallen leaves was observed in the Chahafzal and Poostsiah genotypes, which had the highest accumulation of K^+ in their leaves. Accordingly, 93.77% and 91.12% of the leaves were green at the end of the experiment in these genotypes, respectively (Table 7).

The comparison of the mean of data showed that Cl^- increased in all studied genotypes as a result of increasing salinity, but the increase and accumulation of Cl^- was significantly different in the leaves of the studied genotypes. Based on the results, Cl^- concentration increased significantly in the leaves of the Chahafzal, Poostsiah, and V-Babolsar genotypes at the salinity level of 9 $dS.m^{-1}$ compared to control plants. The increase in Cl^- concentration was significant at the salinity levels of 7 $dS.m^{-1}$ for the Narak genotype and 'M-Yazdi' cultivar and at the salinity levels of 5 $dS.m^{-1}$ for other genotypes compared to control plants (Table 7).

DISCUSSION

Based on the results of this study, with increasing salinity concentration in irrigation water, the final height and number of leaves in all studied cultivars were decreased. Plant height is heavily dependent on the growth environment. Since the growth phenomenon gained vital activities in which condition the plant must have enough water, reduction in the height occurs in case of failure to provide the required water due to the reduction of cell turgor pressure and length of the cells would be negatively affected (Munns, 2002; Munns & Tester, 2008). The osmotic effects of salinity stress can be observed immediately after salt application and are believed to continue for the duration of exposure, resulting in inhibited cell expansion and cell division (Munns 2002; Munns & Tester, 2008).

Table 7. Interaction effect of salinity stress and genotype on changes some physiological traits and percentage concentration of Na^+, K^+ and Cl^-

Cultivar	Salinity Level (dS. m^{-1})	Chlorophyll Index (SPAD value)	K^+ (%)	Na^+ (%)	Na^+/K^+	Cl^- (%)
Pr > F*	-	0.0001>	0.0001>	0.0001>	0.0001>	0.0001>
'Shishecap'	1	19.00 m-t	0.75 s-y	0.12 x-z	0.16 rs	0.50 m-t
'Shishecap'	3	18.75 n-u	0.77 r-x	0.15 w-z	0.19 q-s	0.57 k-p
'Shishecap'	5	16.40 t-y	0.80 q-w	0.50 p	0.62 k-p	0.77 g-j
'Shishecap'	7	16.10 u-z	0.91 j-r	0.90 j	0.98 i-k	0.82 g-i
'Shishecap'	9	13.75 yz	0.73 t-y	1.12 gh	1.53 h	1.80 cd
'Rabab'	1	20.37 h-o	0.70 v-y	0.12 x-z	0.19 q-s	0.41 o-w
'Rabab'	3	20.25 i-o	0.76 r-x	0.16 w-z	0.21 q-s	0.52 m-s
'Rabab'	5	18.52 n-u	0.85 n-v	0.35 st	0.41 m-s	0.73 h-k
'Rabab'	7	16.77 r-x	0.97 i-o	0.79 kl	0.81 j-l	0.87 g-i
'Rabab'	9	14.38 x-z	0.98 h-n	1.14 g	1.16 ij	2.25 a
M-Saveh	1	23.92 a-g	0.72 u-y	0.10 yz	0.13 s	0.44 n-u
M-Saveh	3	24.27 a-f	0.77 r-x	0.12 x-z	0.16 rs	0.58 k-o
M-Saveh	5	19.47 k-r	0.85 n-v	0.69 m-o	0.81 j-l	0.81 g-i
M-Saveh	7	16.50 s-x	0.60 y-a/	1.25 f	2.08 fg	1.21 f
M-Saveh	9	13.47 z	0.45 b/	1.84c	4.08 b	1.95 b
'M-Yazdi'	1	22.77 c-i	0.51 a/	0.09 z	0.17 rs	0.50 m-t
'M-Yazdi'	3	23.65 a-g	0.79 r-w	0.13 w-z	0.17 rs	0.53 m-r
'M-Yazdi'	5	24.10 a-f	0.95iq	0.30 s-w	0.31 n-s	0.59 k-n
'M-Yazdi'	7	20.12 i-p	1.02 g-l	0.70 mn	0.68 k-n	0.89 gh
'M-Yazdi'	9	16.37 t-y	1.05 g-j	1.03 i	0.98 i-k	1.15 f
G-Sarvestan	1	24.00 a-g	0.85 n-v	0.14 w-z	0.17 rs	0.40 p-x
G-Sarvestan	3	23.70 a-g	1.09 f-i	0.19 u-y	0.18 q-s	0.50 m-t
G-Sarvestan	5	21.77 f-l	1.15 d-g	0.61 o	0.53 l-r	0.92 g
G-Sarvestan	7	19.42 k-r	0.99 h-n	1.50 d	1.51 h	1.19 f
G-Sarvestan	9	15.40 v-z	0.68 w-z	2.20 b	3.23 c	2.07 b
'Voshik'	1	22.07 f-k	0.62 z-a/	0.17 v-z	0.27 p-s	0.35 s-x
'Voshik'	3	20.65 h-n	0.67 w-z	0.26 s-v	0.38 m-s	0.55 l-q
'Voshik'	5	17.12 q-w	0.75 s-y	0.63 no	0.84 j-l	0.85 g-i
'Voshik'	7	14.75 w-z	0.55 z-a/	1.46 d	2.65 de	1.45 e
'Voshik'	9	9.60 a/	0.39 c/	2.72 a	6.97 a	1.73 d
G-Shahdad	1	23.15 b-h	1.01 g-m	0.20 u-x	0.20 q-s	0.27 v-z
G-Shahdad	3	23.50 a-g	1.22 c-f	0.29 s-u	0.24p-s	0.42 n-v
G-Shahdad	5	22.40 d-j	1.23 c-f	0.63 no	0.52 l-s	0.64 j-m
G-Shahdad	7	21.25 g-n	1.09 f-i	1.04 hi	0.95 i-k	0.71 i-l
G-Shahdad	9	17.52 p-v	0.96 i-p	1.80 c	1.86 g	1.07 f
G-Saveh	1	22.30 e-j	0.88 l-t	0.20 u-x	0.23 q-s	0.25 w-z
G-Saveh	3	21.82 f-l	1.01 g-m	0.30 s-u	0.29 o-s	0.43 n-w
G-Saveh	5	20.57 h-n	1.05 g-j	0.81 k	0.77 k-m	0.64 j-m
G-Saveh	7	17.50 p-v	0.96 i-p	1.25 f	1.29 hi	1.07 f
G-Saveh	9	14.47 w-z	0.75 s-y	1.77 c	2.35 ef	1.94 bc
Chahafzal	1	25.10 a-d	0.60 y-a/	0.15 w-z	0.25 p-s	0.27 v-z
Chahafzal	3	25.82 ab	0.71 u-y	0.17 v-z	0.24 p-s	0.29 u-z
Chahafzal	5	24.38 a-f	1.13 e-h	0.27 t-w	0.26 p-s	0.33 t-y
Chahafzal	7	22.92 d-j	1.24 c-e	0.47 pq	0.35 n-s	0.40 p-x
Chahafzal	9	19.75 j-q	1.39 ab	0.90 j	0.65 k-o	0.48 m-t
Narak	1	25.28 a-c	0.82 o-w	0.15 w-z	0.18 q-s	0.15 z
Narak	3	25.05 a-e	1.04 g-k	0.22 t-w	0.22 q-s	0.18 yz
Narak	5	23.88 a-g	1.27 b-e	0.30 s-w	0.23 q-s	0.24 x-z
Narak	7	21.95 f-k	1.29 b-d	0.72 lm	0.55 l-r	0.42 n-v
Narak	9	19.08 l-s	1.05 g-j	1.35 e	1.28 hi	0.55 l-q
V-Babolsar	1	22.18 f-k	0.81 p-w	0.21 t-x	0.26 p-s	0.17 yz
V-Babolsar	3	22.10 f-k	0.89 k-s	0.33 r-t	0.37 n-s	0.25 w-z
V-Babolsar	5	21.72 f-m	1.28 b-d	0.61 o	0.48 l-s	0.37 r-x
V-Babolsar	7	17.73 o-v	0.86 m-u	1.33 ef	1.54 h	0.54 m-r
V-Babolsar	9	13.75 yz	0.78 r-w	2.18 b	2.79 d	0.87 g-i
Poostsiah	1	25.57 ab	0.80 q-w	0.17 v-z	0.21 q-s	0.25 w-z
Poostsiah	3	26.15 a	0.91 j-r	0.19 u-y	0.21 q-s	0.28 u-z
Poostsiah	5	25.43 a-c	1.30 bc	0.30 s-w	0.23 q-s	0.38 q-x
Poostsiah	7	22.40 d-j	1.46 a	0.49 p	0.33 n-s	0.48 m-t
Poostsiah	9	19.70 j-q	1.48 a	0.85 jk	0.57 l-q	0.57 k-p

Means in each column and for each trait, followed by different letter(s) are significantly different at 1% probability level, using Duncan's Multiple Range Test.

*= a/ is less than z. Given that variety of data was very wide, Duncan's test grouped data between a to z and less than z such as a/.

Table 8. Rate of Salinity tolerance in the studied genotypes

Genotype	Level of salinity tolerance (dS. m^{-1})
'Shishecap'	3
'Rabab'	5
M-Saveh	3
'M-Yazdi'	5
G-Sarvestan	3
'Voshik'	3
G-Shahdad	7
G-Saveh	3
Chahafzal	7
Narak	7
V-Babolsar	7
Poostsiah	7

According to the results, the percentage of green leaves decreased, and the percentage of necrotic and fallen leaves increased in all studied genotypes. The highest percentage of green leaves was observed in the Chahafzal genotype (95.94% and 93.77%) at the salinity levels of 7 and 9 $dS.m^{-1}$, respectively. Given the amount of physical damage, the Chahafzal genotype had better conditions than the other studied genotypes, and higher levels of salinity did not lead to significant differences in the leaf abscission compared to control plants. These results were consistent with the results of other studies (Momenpour & Imani, 2018; Momenpour et al., 2018). Liu et al. (2018) also showed that salinity up to moderate levels (0.4% NaCl) did not damage the leaves of the Tunisi pomegranate cultivar, indicating the adaptability of this genotype to environmental salinity. The main adverse effect of sodium is the accumulation of its ions in the leaf tissue, leading to the necrosis and aging of the leaves at the tips and margins. These effects continue on the entire leaf area after a while and reduce crop growth in a short time. When plants are exposed to salinity for a longer period, depending on the amount of ion accumulation, specific adverse effects of sodium become apparent in addition to osmotic damage to the plant (Munns, 2002).

The relative water content of the leaves decreased significantly under salinity stress. The relative water content of the leaves decreased from 90.72% in the leaves of the control plants of the Chahafzal genotype to 47.07% in the leaves of the Voshik genotype treated with a salinity of 9 $dS.m^{-1}$. These results are consistent with the results (Shibli et al., 2003; Massai et al., 2004; Liu et al., 2018). Salinity reduces the relative water content and osmotic potential of leaf cell sap during complete turgidity through the gradual accumulation of sodium ions. Osmotic regulation occurs in plants exposed to salinity stress, and as a result, the soluble substances in cells increase due to the high accumulation of sodium and chlorine ions as well as soluble organic matter. This increase in soluble substances in plants tolerant to salinity stress leads to more water uptake compared to the leaves of salinity-sensitive plants, although salinity treatment shows a lower relative water content (Munns et al., 2006).

Previous studies have shown that some pomegranate cultivars are more sensitive to salinity (Dichala et al., 2021; Okhovatian-Ardakani et al., 2010). Many pomegranate species showed a decrease in chlorophyll content in response to salinity stress (Mastrogiannidou et al., 2016; Liu et al., 2108; Dichala et al., 2021). Based on the results, the total chlorophyll content and chlorophyll index decreased in the leaves of the studied genotypes with increasing salinity levels. Decreased chlorophyll levels may be due to increased levels of sodium and chlorine ions. The results were consistent with the results of Ibrahim (2016), Mastrogiannidou et al. (2016), Liu et al. (2018), and Dichala et al. (2021), who reported that the chlorophyll content decreased with increasing salinity levels. Ionic homeostasis in the

environments under salinity stress leads to the increase in sodium and chlorine as toxic ions, their high solubility in water, rapid absorption, and their transfer by transpiration, inhibiting the growth, photosynthesis, and other plant processes. Also, reduction in chlorophyll and photosynthesis, in general, can be attributed to the deficiency of potassium ions in photosynthetic leaf cells (Guo & Tang, 1999). Chlorophyll content decreases under salinity stress due to the activity of the chlorophyllase enzyme. Some growth regulators, such as abscisic acid and ethylene, stimulate this enzyme and increase the concentration of these substances due to stress (Mahajan & Tuteja, 2005; Munns & Tester, 2008).

It is well established that the most important mechanisms that promote cell survival and help with salinity tolerance usually involve controlling Na^+ uptake and distribution in the plant to prevent Na^+ content accumulation in the stem (Tester & Davenport, 2003). In all genotypes under study, leaf Na^+ increased significantly as a result of increasing salinity concentration. Based on the results, the highest concentration of Na^+ was observed in the leaves of the Voshik genotype at the salinity level of 9 $dS.m^{-1}$ (2.72%). In contrast, the lowest sodium concentration was observed in the leaves of the Chahafzal genotype (0.90%). These results were consistent with the results of the morphological traits of plants. Na^+ compartmentalization is usually associated with anatomical adaptations such as succulence and increased cell size or volume (Munns & Tester, 2008). The Voshik genotype, which had the highest accumulation of Na^+ in the leaves, also had the highest percentage of leaf necrosis and abscission, so that only 50.66% of the leaves of this genotype were green at the end of the experiment. Research on various plants under salinity stress has shown that sodium causes osmotic imbalance, damage to the cell membranes, growth reduction, and inhibition of cell division and enlargement (Szczerba et al., 2008; 2009). Other researchers have shown that Na^+ levels increase as a result of increasing salinity levels in pomegranate plants (Naeini et al., 2006b; Okhovatian-Ardakani et al., 2010; Mastrogiannidou et al., 2016; Calzone et al. 2020). K^+ concentration in the leaves of the Chahafzal and Poostsiah genotypes and 'Rabab' and 'M-Yazdi' cultivars was significantly higher than that of plants with increasing salinity up to the level of 9 $dS.m^{-1}$. These results showed that the Chahafzal and Poostsiah genotypes and 'Rabab' and 'M-Yazdi' cultivars are more resistant to the negative and destructive effects of sodium at higher levels of salinity by increasing the amount of K^+. These results were consistent with the results of morphological traits. The Chahafzal and Poostsiah genotypes, which had the highest accumulation of K^+ in the leaves, had the lowest percentage of leaf necrosis and abscission. Accordingly, 93.77% and 91.12% of the leaves were green in these genotypes at the end of the experiment, respectively. In addition to providing the essential nutrient for plant growth, K^+ also plays an essential role in the function of the stomatal apparatus and is required by cells to maintain osmotic balance. Besides, K^+ is an essential factor for many enzymes (Almeida et al., 2017). K^+ has been reported to be effective in maintaining osmotic balance, stomata opening and closing, and reducing the destructive effects of sodium (Szczerba et al., 2008; 2009). Potassium ensures proper CO_2 uptake and respiration after any osmotic changes that may occur by controlling the stomatal apparatus (Araújo et al., 2011). In addition to playing an essential role in vital metabolisms, potassium is so important in salinity stress that its efficient management against sodium in the plant is essential for survival under salinity (Karimi & Hasanpour, 2014). Some plants can protect the cytoplasm of their cells from drastic depletion of potassium and use vacuoles as a reservoir to buffer potassium ions. In this regard, tolerant plants can better maintain their cytosolic potassium levels in the presence of sodium chloride (Karimi & Hasanpour, 2014). A study showed that the K^+ concentration of leaves and roots of the 'Wonderful', 'Ermioni', and 'Grenada' pomegranate cultivars increased in salinity treatments with KCl and K_2SO_4. In contrast, the treatment of plants with NaCl did not show any difference in leaf K^+

concentration compared to the control plants (Dichala et al., 2021), which was consistent with the results of the present study.

CONCLUSION

In general, the results showed that the genotype and salinity level affected the changes in morphological and physiological traits as well as the concentration of nutrition elements. In all studied genotypes, the changes in the measured traits were not significant at the salinity level of 3 $dS.m^{-1}$ compared to the control plants (1 $dS.m^{-1}$), indicating that all studied genotypes could well tolerate this level of salinity. With further salinity increase and at the level of 5 $dS.m^{-1}$, the rate of growth reduction in the Voshik, G-Sarvestan, and G-Saveh genotypes and 'M-Saveh' cultivar was significant compared to control plants. These genotypes could not tolerate this level of salinity while other genotypes could. Voshik genotype and 'M-Saveh' cultivars were identified as the most sensitive to salinity. Overall, the Chahafzal genotype was selected as the most tolerant to salinity stress in this study. This genotype could tolerate salinity up to 7 $dS.m^{-1}$ by maintaining its growth characteristics and increasing K^+ uptake against Na^+. Poostsiah, Narak, M-Yazdi, and G-Shahdad genotypes also showed good salinity tolerance (Table 8). The tolerant genotypes will be used in plans as a base in Ardakan Chah Afzal Station of the National Salinity Research Center (with a longitude of 47° 52', the latitude of 32° 30', and an altitude of 980 meters above sea level; with average water salinity of 7.3+0.5 $dS.m^{-1}$ and average soil salinity at depths of 0-40, 40-80, and 80-120 equal to 7.5, 9.5, and 12 $dS.m^{-1}$ respectively) to graft the selected genotypes on them.

Conflict of interest

Authors declare that they have no conflict of interest in this work.

REFERENCES

Almeida, M. D., Oliveira, M. M., & Saibo. N. J. M. (2017). Regulation of Na+ and K+ homeostasis in plants: towards improved salt stress tolerance in crop plants. Genetics and Molecular Biology, 40, 326–345. https://doi.org/10.1590/1678-4685-gmb-2016-0106

Araújo, W. L., Fernie A. R., & Nunes-Nesi A. (2011). Control of stomatal aperture: a renaissance of the old guard. *Plant Signaling and Behavior*, *6*, 1305–1311. https://doi.org/10.4161/psb.6.9.16425

Arnon, D. I. (1949). Copper enzymes in isolated chloroplast polyphenol oxidase in Beta vulgaris. *Plant Physiology*, *24*, 1- 15.

Ashraf, M. (1994). Organic substances responsible for salt tolerance in *Eruca sativa*. *Biologia. Plantarum*, *36*, 255-259, https://doi.org/10.1007/BF02921095

Ashraf, M., & Harris, P.J.C. (2004). Potential biochemical indicators of salinity tolerance in plants. – *Plant Science*, *166*, 3-16. https://doi.org/10.1016/j.plantsci.2003.10.024

Calzone, A., Cotrozzi, L., Pellegrini, E., Guidi, L., Lorenzini, G., & Nali, C. (2020). Differential response strategies of pomegranate cultivars lead to similar tolerance to increasing salt concentrations. *Scientia Horticulturae*, *271*, 109441. https://doi.org/10.1016/j.scienta.2020.109441

Dichala, O., Therios, I., Papadopoulos, A., Chatzistathis, T., Chatzisavvidis, C., & Antonopoulou, C. (2021). Effects of varying concentrations of different salts on the mineral composition of leaves and roots of three pomegranates (*Punica granatum* L.) cultivars. *Scientia Horticulturae*, *275*, 109718. https://doi.org/10.1016/j.scienta.2020.109718

Emami, A. (1996). *Methods of plant analysis.* Agricultural Research and Education Organization. Soil and Water Institute. 130 Pp.

Flowers, T. J. & Colmer, T. D. (2015). Plant salt tolerance: adaptations in halophytes. *Annals of Botany*, *115*, 327–331. https://doi.org/10.1093/aob/mcu267.

Guo, F. O., & Tang, Z. C. (1999). Reduced Na+ and K^+ permeability of K^+ channel in plasma membrane isolated from roots of salt-tolerant mutant of wheat. *Chinese Academy of Sciences, 41*(9), 217-220.

Ibrahim, H.I.M. 2016. Tolerance of two pomegranates cultivars (*Punica granatum* L.) to salinity stress under hydroponic culture conditions. Journal of Basic *Applied Science Research Flora, 4*, 38-46.

Karimi, H.R. & Z. Hasanpour. 2014. Effects of salinity and water stress on growth and macronutrient concentration of pomegranate (*Punica granatum* L.). *Journal Plant Nutrition, 37*, 1937–1951. https://doi.org/10.1080/01904167.2014.920363.

Khayyat, M., Tehranifar, A., Davarynejad, G.H., & Sayyari-Zahan, M.H. (2014). Vegetative growth, compatible solute accumulation, ion partitioning and chlorophyll fluorescence of 'Malas-e-Saveh' and 'Shishe-Kab' pomegranates in response to salinity stress. *Photosynthetica, 52*(2), 301-312. https://doi.org/10.1007/s11099-014-0034-9.

Liu, C., Ming, Y., Xianbin, H., & Zhaohe, Y. (2018). Effects of salt stress on growth and physiological characteristics of pomegranate (*Punica granatum* L.) cuttings. *Pakistan Journal of Botany, 50* (2), 457-464.

Lutts, S., Kinet, J.M., & Bouharmont, J. (1995). Changes in plant response to NaCl during the development of rice (*Oryza sativa L.*) varieties differing in salinity resistance. *Journal of Experimental Botany, 46*, 1843–1852. https://doi.org/10.1093/jxb/46.12.1843

Mahajan, Sh., & Tuteja, N. (2005). Cold, salinity and drought stresses: An overview. *Archives of Biochemistry and Biophysics, 444*, 139-158. https://doi.org/10.1016/j.abb.2005.10.018

Marschner, H. (1995). Mineral Nutrition of Higher Plants. 2nd Ed. Pp.892. Academic Press Ltd., London 1995.

Massai, R., Remorni, D., & Tattini, M. (2004). Gas exchange, water relations and osmotic adjustment in two scion/rootstock combinations of Prunus under various salinity concentrations. *Journal of Plant Soil Science, 259*, 153-162. https://www.jstor.org/stable/24124369

Mastrogiannidou, E., Chatzissavvidis, C., Antonopoulou, C., Tsabardoukas, V., Giannakoula, A., & Therios, I. (2016). Response of pomegranate cv. wonderful plants to salinity. *Journal of Soil Science and Plant Nutrition, 16*(3), 621-636. http://dx.doi.org/10.4067/S0718-95162016005000032.

Momenpour, A., & Imani, A. (2018). Evaluation of salinity tolerance in fourteen selected pistachio (*Pistacia vera* L.) cultivars. *Advances in Horticultural Science, 32* (2), 249-264. https://doi.org/10.13128/ahs-22261

Momenpour, A., Imani, A., Bakhshi, D., & Akbarpour, E. (2018). Evaluation of salinity tolerance of some selected almond genotypes budded on GF_{677} rootstock. *International Journal of Fruit Science, 18*(4), 410-435. https://doi.org/10.1080/15538362.2018.1468850

Munns, R. (2002). Comparative physiology of salt and water stress. *Plant, Cell & Environment, 25*, 239-250. https://doi.org/10.1046/j.0016-8025.2001.00808.x.

Munns, R., & Tester, M. (2008). Mechanisms of salinity tolerance. *Annual Review of Plant Biology, 59*, 651–681. https://doi.org/10.1146/annurev.arplant.59.032607.092911.

Munns, R., James, R.A., & Lauchli, A. (2006). Approaches to increasing the salt tolerance of wheat and other cereals. *Journal of Experimental Botany, 57*, 1025-1043. https://doi.org/10.1093/jxb/erj100

Naeini, M. R., Khoshgoftarmanesh, A. H., & Fallahi, E. (2006a). Partitioning of chlorine, sodium, and potassium and shoot growth of three pomegranate cultivars under different levels of salinity. *Journal Plant Nutrition, 29*, 1835-1843. https://doi.org/10.1080/01904160600899352.

Naeini, M. R., Khoshgoftarmanesh, A. H., Lessani, H., & Fallahi, E. (2006b). Effects of sodium chloride-induced salinity on mineral nutrients and soluble sugars in three commercial cultivars of pomegranate. *Journal Plant Nutrition, 27*, 1319-1326. https://doi.org/10.1081/PLN-200025832

Okhovatian-Ardakani, A. R., Mehrabanian, M., Dehghani, F., & Akbarzadeh, A. (2010). Salt tolerance evaluation and relative comparison in cuttings of different pomegranate cultivars. *Plant Soil and Environment, 56*(4), 176–185. https://doi.org/10.17221/158/2009-PSE

Pang, C. H., & Wang, B. S. (2008). *Oxidative stress and salt tolerance in llants.* In: In: Lüttge, U., Beyschlag, W., Murata, J. (Eds.), Progress in Botany, vol. 69. Springer, Berlin, pp. 231–245. https://doi.org/10.1007/978-3-540-72954-9_9.

Papadakis, I. E., Veneti, G., Chatzissavvidis, C., Sptiropoulos, T. E., Dimassi, N., & Therios, I. (2007). Growth, mineral composition, leaf chlorophyll and water relationships of two cherry varieties under NaCl-induced salinity stress. *Soil Science and Plant Nutrition, 53*, 252-258. https://doi.org/10.1111/j.1747-0765.2007.00130.x

Parvizi, H., Sepaskhah, A. R., & Ahmadi, S. H. (2016). Physiological and growth responses of pomegranate tree (*Punica granatum* (L.) cv. Rabab) under partial root-zone drying and deficit irrigation regimes. *Agricultural Water Management, 163*, 146–158. https://doi.org/10.1016/j.agwat.2015.09.019

Sarkhosh, A., Zamani, Z., , Fatahi, R., & . Ebadi, A. (2006). RAPD markers reveal polymorphism among some Iranian pomegranate (*Punica granatum* L.) genotypes. *Journal of Horticultural Science*, 111, 24–29. https://doi.org/10.1016/j.scienta.2006.07.033

Shibli, R. A., Shatnawi, M. A., & Swaidat, I. Q. (2003). Growth, osmotic adjustment and nutrient acquisition of bitter almond under induced sodium chloride salinity in vitro. *Communication in Soil Science and Plant Analysis, 34*, 1969-1979. https://doi.org/10.1081/CSS-120023231

Silva-Ortega, C.O., Ochoa-Alfaro, A.E., & Reyes-Aguerro, J.A. (2008). Salt stress increases the expression of *p5cs* gene and induces proline accumulation in cactus pear. *Plant Physiology* and *Biochemistry*, 46, 82-92. https://doi.org/10.1081/CSS-120023231

Szczerba, M. W., Britto, D. T., & Kronzucker, H. J. (2009). K^+ transport in plants: physiology and molecular biology. *Journal of Plant Physiology, 166*, 447-466. DOI: 10.1016/j.jplph.2008.12.009

Szczerba, M. W., Britto, D. T., Balkos, K. D., & Kronzucker, H. J. (2008). NH_4^+ stimulated and -inhibited components of K+ transport in rice (*Oryza sativa* L.). *Journal of Experimental Botany, 59*, 3415–3423. https://doi.org/10.1093/jxb/ern190

Tester, M., & Davenport, R. (2003). Na+ tolerance and Na+ transport in higher plants. *Annals of Botany, 91*, 503–527. https://doi.org/10.1093/aob/mcg058.

Yamasaki, S., & Dillenburg, L. C. (1999). Measurements of leaf relative water content in *Araucaria angustifolia*. *Revista Brasileira de Fisiologia Vegetal, 11*, 69-75.