

## **Photosynthesis Properties and Ion homeostasis of Different Pistachio Cultivar Seedlings in Response to Salinity Stress**

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### **Abstract**

Understanding mechanisms of salt tolerance, physiological responses to salt stress, and screening genotypes for breeding programs are important scientific issues remained to be investigated in pistachio. Therefore, current study was carried out to investigate response of different pistachio cultivars (G1, G2, Kaleghochi and UCB1) to salinity treatments (0.6 as control, 10, 20 dS m<sup>-1</sup> using saline underground water) as a factorial experiment based on randomized complete block design with three replications in greenhouse of Iranian Pistachio Research Institute (Rafsanjan) in 2014-2015. Some physiological and nutrition properties of the pistachio cultivars measured in this study. Results showed decreased stomatal conductance, photosynthesis rate, chlorophyll content, and Fv/Fm in response to salinity treatments. The main cause of these changes was related to the altered ion contents along with the competition among ions for being absorbed by plant. Despite of sufficient amount of potassium in the soil, high concentrations of sodium and other associated elements such as calcium and magnesium decreased the ability of pistachio plants to absorb adequate amount of vital ions such as potassium. As a result of sodium accumulation and deficiency of potassium, K<sup>+</sup>/Na<sup>+</sup> ratio was decreased in pistachio leaves. Consequently, toxicity of sodium ions in the plant cells reduced stomatal conductance and the rate of photosynthesis. Comparison between cultivars showed that for the most of the traits the difference between control and moderate salinity (10 ds m<sup>-1</sup>) in all cultivars was not significant. However, G2 cultivar showed higher ability to accumulate potassium and absorbed lower concentration of sodium, calcium, and magnesium under severe salinity treatment (20 ds m<sup>-1</sup>). These result suggested that G2 could be considered as a potential tolerant cultivar for cultivation in saline area.

**Keywords:** Salt tolerance, K<sup>+</sup>/Na<sup>+</sup> ratio, stomatal conductance, potassium, magnesium.

### **Introduction**

In arid and semi-arid areas, salinity (soil or water salinity) is considered as one of the most important environmental challenges and abiotic stresses for agriculture. In these areas, normally, excess evapotranspiration compared to the lower precipitation causes the accumulation of salts in the soil (Deinlein et al., 2014). Under salinity condition, plants

are faced to serious problems regarding achieving proper content of water resulting from alternation in mineral contents of the soil solution which reduces the osmotic potential of the soil and make challenging and difficult situation regarding water absorption by plant (Karimi et al., 2009). Under salinity conditions, reduced growth and fixation of carbon dioxide are associated with decreased stomatal conductance, electron transfer rate, leaf water potential,

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reduced photosynthetic efficiency, and increased concentration of sodium ions and chlorine in leaves (García-Sánchez et al., 2003). Regarding the effect of salinity on plant properties a study was carried out on almond which showed that salinity increased chlorophyll fluorescence which resulted from disruption of chloroplast thylakoid membrane by salinity stress (Oukarroum et al., 2007). High salt concentration also causes disruption in many physiological processes in the plant (Karimi et al., 2009). Documented evidences showed that salinity changes the ratio between chlorophyll and carotenoids (Attarzadeh et al., 2016; Zadsalehimasouleh et al., 2014). Chlorophylls as photosynthetic pigments are the most important factors determining photosynthetic capacity of the plants which directly affects the photosynthetic rate and final plant's production and biomass (Najafian et al., 2008; Paknejad et al., 2007). According to study of Galvan-Ampudia et al. (2013), salinity stress also reduces leaf growth through the prevention of cell growth, leading to reduced water absorption from the soil and reduced plant evapotranspiration. García-Sánchez et al. (2003) in a field research found that by increasing the salinity, concentration of the sodium in leaves of lemon increased, while potassium concentration was not influenced, so that the ratio of potassium to sodium decreased. Different scientists have noted that the imbalance between potassium and sodium may lead to biochemical disturbances and reduce the performance or quality of the fruit (Attarzadeh et al., 2016; Bernstein, 1975; Deinlein et al., 2014). Sodium can also directly disturb the nutrients balance and physical structure of soil. Excess content of sodium in the soil can induce calcium, potassium and magnesium deficiencies. On the other hand, increased ratio of sodium to calcium significantly reduces water infiltration in soil (Bernstein, 1975).

Pistachio is one of the most important exporting crops of Iran that is commonly cultivated and grown in saline soils.

Accordingly, distinguishing and using salt tolerant cultivars is an acceptable method to cope with salinity and achieve proper yield. It is recently determined that the soils which have been using for pistachio cultivation in Kerman province of Iran, as one of the greatest and significant production center of pistachio in Iran, are increasingly moved toward higher salinity condition. Therefore, investigating effective methods to provide practical approaches to minimize the harmful effects of this stress is necessary (Sajedi Nia, 2008). Alipur et al. (2012) examined 32 cultivars of female pistachio in the collection of Pistachio Research Institute of Iran and 27 male genotypes from the areas of Kerman, Rafsanjan, Sirjan and Shahrabak (all located in Iran) and conducted artificial crosses for 32 male cultivars with one male genotype and also 27 male genotypes with one hazelnut cultivar. They found that female cultivars of Ebrahimi, Serizi, Jandaghi, Pustkaghazi, Fandoghi 48, Badami-Zarand, Saifuddini and Sarakhs male genotypes of M16, M21, M23, M25, M26 and M27 were relatively salt-tolerant cultivars due to higher number of survived seedlings, seedling length, number of leaves, root dry weight, and lower amount of sodium in stem when compared to the other cultivars. Although some researchers have conducted studies to introduce salt tolerant cultivars (Alipur et al., 2012; Bastam et al., 2013; Mozaffari et al., 2015; Sajedi Nia, 2008), there is still little information considering the physiological mechanism of salt tolerance in pistachio plant. Therefore, this study was conducted to investigate the physiological properties and ion status of four pistachio cultivars in response to saline conditions.

### Materials and methods

To study the physiological aspects of salt tolerance in two known cultivars along with two genotypes of pistachio, a greenhouse experiment was conducted at Pistachio Research Institute of Rafsanjan in 2015. The experiment was arranged as a factorial

experiment based on Completely Randomized Block Design (CRBD or RB) with three replications. Two main factors that were investigated in this research were: salinity (in three levels: 0.6 (as a control), 10 and 20 dS m<sup>-1</sup> using saline water wells in the area) and pistachio cultivars (two genotypes of G1, G2, along with Kallehghoochi and American Ucb1 pistachio as two known cultivars). Both genotypes of G1 and G2 were formerly cultivated and grown in Rafsanjan region for about 38 years, which are grown in orchards under the soil salinity of 45 dS m<sup>-1</sup>. After chemical analysis of soil (Table 1), 5 kg soil was poured into the pots with volume of 5 liters. Then, germinated seeds of pistachio were planted in pistachio pots and after the initial growing; three seedlings were kept in each pot. Pots were normally irrigated with no-saline water until three-leaf stage considering 50% soil moisture discharge. After this stage, application of salinity treatments was started. For irrigation, 50% soil moisture discharge of the soil in the pots were taken as the index for next irrigation regime where it was determined using moisture suction curve and the available water in the soil (Tavallali et al., 2008). The amount of required water for irrigation was determined as usual of the region. In order to provide the proper nutrients content, initial analysis of the soil was carried out and accordingly chemical fertilizer as P-N-K at the rates of 20-20-20 percent was used and 50 g applied for every pot. Salinity treatments were last for four months until the final growth stage of the seedling. To avoid higher accumulation of salt in the pots as what was designed for the experiment, the amount of salinity in the soil as a result of irrigation with saline water was

regularly measured and the pots were leached when it was required. Subsequently, the amount of salinity in the soil solution was not reached to 1.5 times higher than irrigation water. At the end of experiment when the seedlings reached their full growth stage (about four months after onset of experiment), properties such as the concentration of elements such as calcium, magnesium, sodium and potassium of the leaves were measured. The two top leaves of each seedling for all three ones in each pots were taken as leaf samples where the number of final leaves for each pots reached the six samples. After sampling the leaves, the samples were transferred to laboratory of Pistachio Research Institute and the elements content were determined using Inductively Coupled Plasma Spectrometry (Perkin Elmer model optime 7000 Dv made in America). Additionally, some physiological properties of seedlings including photosynthesis rate, stomatal conductance, transpiration, and leaf temperature were measured by ADC Bioscientific LCA4 device. Chlorophyll index (spad index) along with the ratio of Fv/Fm (maximum quantum yield of photosystem II) were measured in the last and highest leaf of each seedling before sampling for element contents at the afternoon time using OptiScience Inc device . For measuring the Fv/Fm, leaves were exposed firstly to darkness for half an hour with special clips.

After collecting the data, analysis of variance (ANOVA) and mean comparison were done according to the Duncan's Multiple Range Test (DMRT) at 5% level and Pearson Correlation Coefficient using core package and Agricolea package of R 3.4.2 software.

**Table 1. Physical and chemical properties of the soil used in the experiment**

ECe*	pH	SAR	K <sub>(AVA.)</sub>	P <sub>(AVA.)</sub>	Na <sub>s</sub>	Ca <sub>s</sub>
4	7.6	6.8	350	20	19.5	8.4
Mg <sub>s</sub>	Cl	T.N.V	Clay	Silt	Sand	Texture
11	36	14	4.4	34.6	61	Sandy loam

\* ECe: electric conductance (dS/m), pH: acidity of flower saturated, SAR: sodium absorption ratio, K<sub>(AVA.)</sub>: absorbable potassium (mg per kg of soil), P<sub>(AVA.)</sub>: absorbable phosphorus (mg per kg of soil), Cl, Mg<sub>s</sub>, Ca<sub>s</sub>, Na<sub>s</sub>: respectively, chloride, magnesium, calcium, and sodium in soil solution (meq per liter), T.N.V: percent of neutralizing materials (calcium carbonate equivalent), Clay: percentage of clay, silt: silt percentage, sand: sand percentage, texture: soil texture

## Results

The results of analysis of variance (ANOVA) showed that the effect of salinity and interaction of salinity and cultivar were significant for stomatal conductance (Table 2). Result of mean comparison for the interaction of salinity and cultivar is presented in Table 3. This result indicated that with the increasing the level of salinity up to 20 ds m<sup>-1</sup>, stomatal conductance reduced in all cultivars. Although the stomatal conductance decreased by increasing the salinity, this reduction was depended on the cultivar, where the rate of decrease in stomatal conductance under the control condition to 20 ds m<sup>-1</sup> was 75% in UCB1 cultivar but this decrease was not statistically significant for G2 cultivar (Table 3). The highest stomatal conductance was observed in UCB1 cultivar under the control condition, while it showed no significant

difference with Kallehghoochi under both control and 10 ds m<sup>-1</sup> and also with UCB1 under 10 ds m<sup>-1</sup>. The lowest stomata conductance was observed in control and 10 ds m<sup>-1</sup> salinity levels in G2 cultivar.

For photosynthesis rate, significant difference was found among pistachio cultivars and also for the interaction between cultivar and salinity (Table 2). The highest and lowest rates of photosynthesis were obtained in cultivars UCB1 and G1, respectively. Interaction effect of salinity and cultivar showed that with an increase in salinity from 0.6 to 20 ds m<sup>-1</sup>, photosynthesis rate of UCB1 cultivar reduced by approximately 28 percent, while it did not change significantly in Kallehghoochi cultivar (Table 3). The overall difference between the control and the highest salinity level (20 ds m<sup>-1</sup>) was significant for Kallehghoochi and UCB1, but it was not significant for G1 and G2 cultivars.

**Table 2. Analysis of variance (ANOVA) for measured traits in the experiment**

Source of changes	degree of Freedom	Stomatal conductance	Photosynthesis rate	Fv/Fm	Chlorophyll index	Leaf potassium	Leaf sodium
Replication	2	0.00005	71/25	0.0098	544.27	0.23	0.018
Salinity	2	0.00006*	143.16ns	0.009**	205.34*	0.04ns	3.80**
Cultivar	3	0.00003ns	1669.7**	0.007**	1133.05**	0.4**	0.25*
Salinity*Cultivar	6	0.00004*	1014.95**	0.001ns	257.88ns	0.11ns	0.37**
Error	22	0.00001	90.37	0.001	215.99	0.057	0.065
Coefficient of variation		22.32	29.09	4.56	24.51	13.52	21.51
Source of changes	degree of Freedom	K <sup>+</sup> /Na <sup>+</sup> ratio	Leaf calcium	Leaf magnesium	Leaf temperature	Transpiration	
Replication	2	0.34	0.21	0.004	4.62	0.0015	
Salinity	2	18.98**	0.53*	0.38**	0.19ns	0.02ns	
Cultivar	3	1.51**	1.04**	0.27**	0.05ns	0.024ns	
Salinity*Cultivar	6	1.90**	0.33*	0.22*	0.22ns	0.038ns	
Error	22	0.28	0.128	0.45	0.26	0.03	
Coefficient of variation		22.43	22.24	24.94	1.62	21.35	

ns, \*, \*\* are representing non-significance, and significance at 5% and 1%, respectively.

**Table 3. Mean comparison for properties in which the interaction between salinity and cultivar were significant.**

Treatment	Stomatal conductance (mmol H <sub>2</sub> O m <sup>-2</sup> s <sup>-1</sup> )	Photosynthesis rate (μmol CO <sub>2</sub> m <sup>-2</sup> s <sup>-1</sup> )	Leaf sodium	K <sup>+</sup> /Na <sup>+</sup> ratio	Leaf calcium	Leaf magnesium
Kallehghoochi * 0.6 dS/m	0.015a-c	9.4de	0.52d	3.70a	.138cd	0.67d-f
Kallehghoochi * 10 dS/m	0.016ab	12.2de	1.51ab	1.27d	1.54b-d	0.69d-f
Kallehghoochi * 20 dS/m	0.013bc	9.2e	1.4b	1.3dc	1.32cd	0.62d-f
G1 * 0.6 dS/m	0.013bc	7.0e	0.55d	3.70a	1.39cd	0.84c-f
G1 * 10 dS/m	0.011bc	7.7e	1.09bc	1.53c-d	2.07b	0.84c-f
G1 * 20 dS/m	0.010cd	14.7cd	1.96a	1.11d	1.68b-d	1.49a
UCB1 * 0.6 dS/m	0.020a	24.8a	0.61cd	2.14bc	1.08d	1.21ab
UCB1 * 10 dS/m	0.015a-c	12.9c-e	1.52ab	1d	1.17cd	0.86b-e
UCB1 * 20 dS/m	0.005d	17.6bc	1.43b	1.05d	1.56b-d	0.98b-d
G2 * 0.6 dS/m	0.01cd	9.3de	0.47d	4.12a	1.71bc	0.5f
G2 * 10 dS/m	0.01cd	20.8ab	1.2b	1.75b-d	1.71bc	0.6ef
G2 * 20 dS/m	0.011bc	11.4de	1.23b	1.75b	69.2a	1.14a-c

Means having at least one shared letter have no significant difference according to Duncan's Multiple Range Test (DMRT 5%)

Individual effect of salinity and cultivar on Fv/Fm was significant but their interactions showed no significant effect in this regard (Table 2). Since the interaction of the cultivar and salinity was not significant, their main effects could be directly analyzed by mean comparison. The result of mean comparison for salinity and cultivar levels for Fv/Fm is presented in Fig.

1a and Fig. 2a, respectively. Increased salinity from control to 20 dS m<sup>-1</sup> led to a decrease by rate of 7% in Fv/Fm (Fig. 1a). No significant difference was observed between control and 10 ds m<sup>-1</sup> salinity level. Among cultivars, the cultivar UCB1 showed the highest Fv/Fm, while the rest of the cultivars (Kalleghoochi, G1, and G2) showed no significant difference (Fig. 2a).

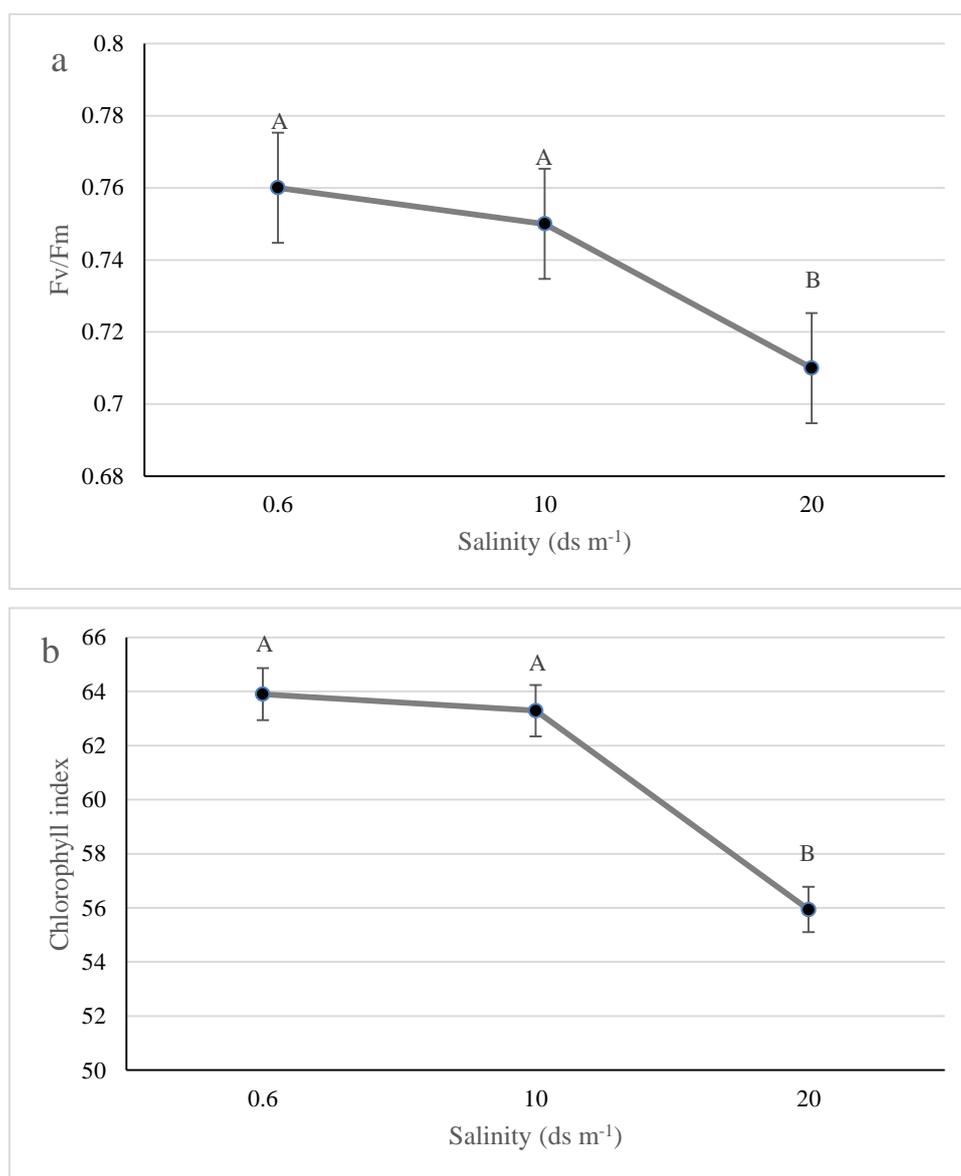
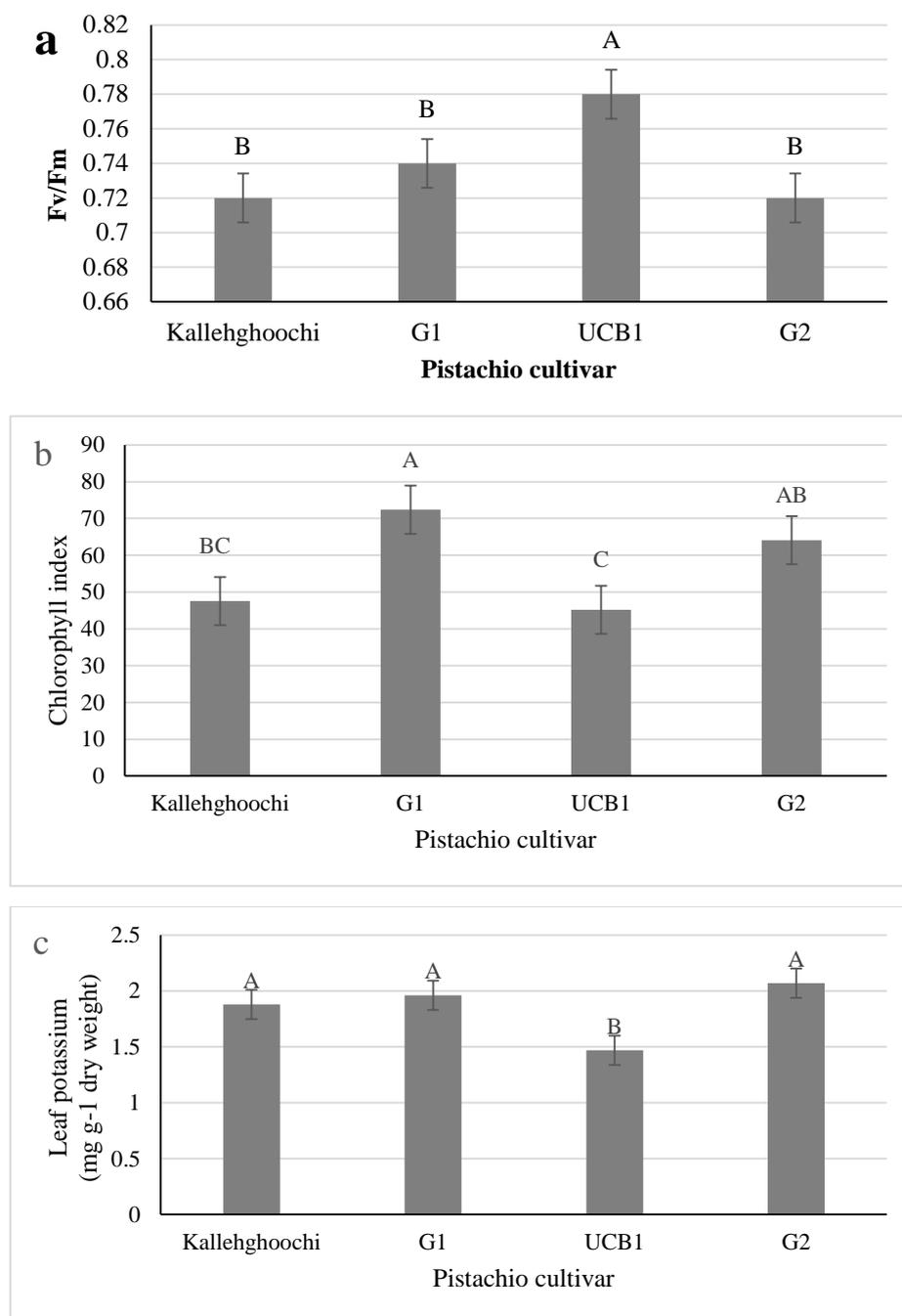


Fig. 1. Mean comparison of salinity levels in all pistachio cultivars for Fv/Fm (a) and leaf chlorophyll index (b) according to Duncan's Multiple Range Test (DMRT 5%)



**Fig. 2.** Mean comparison of pistachio cultivars under different salinity levels for Fv/Fm (a), chlorophyll index (b), and leaf potassium content (c) according to Duncan's Multiple Range Test (DMRT 5%).

The main effect of salinity and cultivar on chlorophyll index in ANOVA table (Table 2) was significant at 5% and 1% level, respectively, but their interactions showed no statistically significant effect. Chlorophyll index reduced by approximately 20% in highest level of salinity (20 ds m<sup>-1</sup>) in comparison with

control (Fig. 1b). The difference between control and 10 ds m<sup>-1</sup> salinity level was not statistically significant. G cultivar showed the highest chlorophyll index but its difference with G2 was not significant. The lowest mean value for this index was observed in UCB1 cultivar.

The effect of cultivar on potassium

content of pistachio leaves was significant while the salinity and the interaction effects were not significant (Table 2). The result of mean comparison for cultivars showed that G2 cultivar had the highest potassium content where its difference with G1 and Kallehghoochi was not significant (Fig. 2c). The lowest content of potassium was obtained in UCB1 cultivar.

Analysis of variance showed that foliar sodium concentration was affected by salinity, cultivar and their interactions (Table 2). By increasing the salinity from 0.6 to 10 and 20 dS m<sup>-1</sup>, sodium concentration in leaves increased up to 2.4 and 2.8 times, respectively. Although, salinity significantly increased the content of sodium in all studied cultivars, but the rate of increase was varied for different cultivars. The highest increase rate in sodium content in response to salinity was recorded for G1 cultivar (about three times), while the lowest rate achieved by G2 cultivar by almost 1.4 times increase (Table 3). The highest sodium content among all cultivars was observed in G under highest salinity level (20 ds m<sup>-1</sup>), while the lowest sodium content was achieved in G2 under control condition.

The main effects of salinity and cultivar in addition to their interactions were significant regarding the K<sup>+</sup>/Na<sup>+</sup> ratio (Table 2). The ratio of K<sup>+</sup>/Na<sup>+</sup> was decreased in response to increase in the salinity levels, but the rate of reduction was varied among studied pistachio cultivars (Table 3). By increasing salinity from 0.6 to 20 dS m<sup>-1</sup>, the ratio of K<sup>+</sup>/Na<sup>+</sup> in the Kallehghoochi cultivar reduced by 65%, while this reduction in other three cultivars of G1, UCB1, and G2 was 70, 50, and 57 percent, respectively (Table 3). In highest salinity level (20 dS m<sup>-1</sup>) the ratio of K<sup>+</sup>/Na<sup>+</sup> in G2 cultivar was highest compared to its ratio in other cultivars, where the rate of increase in G2 cultivar in comparison with Kallehghoochi, UCB1, and G1 under the same salinity level was 25, 36 and 40 percent, respectively. UCB1

under highest salinity level and G2 under control condition showed the lowest and the highest K<sup>+</sup>/Na<sup>+</sup> ratio, respectively (Table 3).

The result of analysis of variance showed significant effects of salinity, cultivar, and their interactions for calcium and magnesium contents in pistachio leaves (Table 2). The results of mean comparison showed that by increasing the salinity from 0.6 to 20 dS m<sup>-1</sup>, leaves calcium content was significantly increased from 1.39 to 1.81 (Table 3). Calcium content varies among different cultivars; accordingly, cultivars G2 and UCB1 had the highest and lowest content of calcium in their leaves, respectively. Increase salinity from 0.6 to 20 dS m<sup>-1</sup> led to a 4.3% decrease in calcium content of leaves in the Kallehghoochi cultivar, but it was increased by 20.8, 44, and 57.3% in G1, UCB1, and G2 cultivars, respectively (Table 3). At all levels of salinity, cultivars G2 and UCB1 in control condition had the minimum calcium content in their leaves. In all cultivars, higher salinity levels caused an increase in magnesium content in the pistachio leaves. The highest content of magnesium in all cultivars was observed under 20 dS m<sup>-1</sup> salinity level (Table 3). Cultivar G1 under the highest salinity level (20 dS m<sup>-1</sup>) had the highest and cultivar G2 under the salinity of 0.6 dS m<sup>-1</sup> had the lowest foliar magnesium content.

According to result of analysis of variance for leaf temperature and transpiration, all of the main effects neither for salinity and cultivar nor for their interaction were significant (Table 2).

## Discussion

The result for stomata conductance showed the negative effects of salinity on this parameter for all studied cultivars, but with different rate in each cultivar. In accordance, the negative effects of salinity on stomatal conductance have been previously reported (Hossain et al., 2015; Saed-Moocheshi et al., 2014). Stomatal

conductance is directly affected by the stomata closure. Under salinity stress, due to the imposed water shortage closing of the stomata occurs to maintain water in the cells. Therefore decreased stomata conductance because of stomatal closure is not surprising (Pirasteh-Anosheh et al., 2016). Mozaffari et al. (2015) showed that with increase salinity, water potential in soil decreases and the rapid stomatal closure can be due to low water potential in both soil and finally in plant cells. They also disused that less water absorption by the plant's roots reduces the degrading effects of sodium toxicity in plant (Mozaffari et al., 2015). Jalili Marandi et al. (2012) reported that under salinity conditions, stomatal resistance increases and stomatal conductance decreases. They also expressed that the absorption and transport of sodium to leaves causes stomata closure in sensitive plants to salinity. Under salinity condition, the content of sodium in plant is increased which results in triggering accumulation of abscisic acid leading to decrease in stomatal conductance (Bernstein, 1975). In line with the decrease in stomatal conductance, photosynthetic rate and finally the plant growth is decreased (Yuan and Lin, 2008). Accordingly, the rate of photosynthesis in all pistachio cultivars have decreased in response to salinity condition. Decrease in the content of photosynthesis is also associated with the content and the ability of chlorophylls in plant leaves. The chlorophyll index as an index representing the chlorophyll content decreased under salinity stress. Salinity stress in most of plants reduced the amount of photosynthesis and chlorophyll content, but the amounts of reduction varied in different plants (Ranjbar et al., 2001). Previous reports suggest reduced photosynthesis up to about two-thirds due to increased stomatal resistance. Increased stomatal resistance in plants under salinity stress is created to maintain a positive balance of water inside the plant (Ranjbar

et al., 2001; Sevengor et al., 2011). In the current study, in most of the cases related to stomatal conductance, photosynthesis rate, and chlorophyll content, the different between control and moderate salinity level ( $10 \text{ ds m}^{-1}$ ) were not significant indicating the partial resistance of the used cultivars to salinity. Under the highest level of salinity highest stomata conductance, photosynthesis rate, and chlorophyll index were obtained in Kalleghoochi, UCB1, and G1, respectively. Mozaffari et al. (2015) showed that the rate of photosynthesis in pistachio (Gazvini cultivar) was 12% higher than that in Badami cultivar, which is in accordance to the current result for varied response of the cultivars. Mozaffari et al. (2015) have also reported that salinity stress reduces chlorophyll fluorescence. In all used cultivars of the current study, the Fv/Fm was similarly decreased under salinity stress by different rate in different cultivars of pistachio. UCB1 cultivar showed highest mean value for this trait under all salinity levels. Reduction in chlorophyll fluorescence parameters under salinity stress could be due to the destruction of chloroplasts, especially thylakoid membrane. The cause of the reduction in the ratio of Fv/Fm is increased value of  $F_0$  as a result of salinity stress (Paknejad et al., 2007). Sajedi Nia (2008) observed that there is difference among cultivars in terms of chlorophyll fluorescence, and in five stages of measurement, the Rezaei cultivar had the highest value of this index. Decreased content of chlorophyll along with lower stomatal conductance is leading to reduction in absorption of carbon dioxide which affects the chlorophyll fluorescence properties. In this regard, under high salt concentration, due to the effect of salinity on proteins, the bond between chlorophyll and chloroplast proteins is loosed and chlorophylls are destroyed (Najafian et al., 2008).

In saline soils, the content and the standard proportion of nutritional elements

for plants is altered which normally leads to increase in the content of unnecessary ions in comparison with the necessary ones. The contents of potassium and sodium in pistachio cultivars used in the current study have inversely altered in response to salinity condition. Potassium decreased with increase in salinity severity, while the content of unwanted sodium increased. As a result,  $K^+/Na^+$  ratio have also decreased under salinity stress. Higher concentration of sodium in both soil and plant leads to lower water potential in line with lower potential of potassium and other necessary elements. In this situation plant has to absorb the water and necessary nutrients in opposite direction of their potential and in the presence of unnecessary and unwanted elements such as sodium leading to consume higher energy which results in altered physiological and growth responses. Adish et al. (2010) discussed that species and genotypes that have acquired higher ability to absorb nutrients in saline conditions would show greater tolerance to salinity. The result of mean comparison for cultivars in the current study has shown that G2 cultivar had the highest potassium content among all cultivars. Also, the highest increase rate in sodium content in response to salinity was recorded for G1 cultivar (about three times), while the lowest rate achieved by G2 cultivar by almost 1.4 times increase. The high percentage of potassium while low content of sodium in G2 cultivar compared to other cultivars indicates the ability of this cultivar to absorb potassium as a necessary element under saline conditions. G2 cultivar by acquiring lower sodium content while higher potassium and  $K^+/Na^+$  ratio under severe salinity level could be suggested for cultivation in saline area, especially in Rafsanjan district. In a study on olive cultivars under salinity conditions, it was reported that different cultivars have significantly different capacity to absorb calcium. In addition, the researchers reported that calcium content

cannot be considered as an indicator for salinity tolerance (Rezaei et al., 2006). Potassium ions in the cell cytosol play an important role in activity of many enzymes of the photosynthesis and it plays as osmotic regulator during the cell development and stomatal activities (Falah et al., 2015). Also, because of the ability of potassium in assisting detoxification of reactive oxygen species (ROS), higher potassium under salinity condition might prevent decomposition of chlorophyll and increase the content of chlorophyll leading to higher tolerance of plants to salinity stress. Potassium also is growth triggering element in the plant and its higher content under saline condition would also increase the leaf surface, causing higher light absorption along with higher chlorophyll content and photosynthesis rate (Najafian et al., 2008). Consequently, cultivars with ability to absorb the higher potassium would have higher tolerance to salinity which G2 in this study represent this capability. Contrarily, it has been claimed that low accumulation of sodium in plant tissues is highly associated with salt tolerance (Laohavisit et al., 2013; Sajedi Nia, 2008). In accordance with our results, Tavallali et al. (2009) reported that by increase in the salinity level, foliar sodium content will be also increased. In accordance with the obtained result in the current study, lower ratio of  $K^+/Na^+$  have also reported by other authors such as Khan et al., (2009) in which the absorption of sodium ions increased while the potassium absorption decreased due to the phenomenon of competition in pepper cultivars.

In addition to sodium and potassium contents, the availability portion of other elements is altered by salinity stress. Increasing the salinity level in this study led to increase in the content of calcium and magnesium of pistachio cultivars. In cell membrane of the root tissues, there are some spaces to bind calcium ions under the salinity conditions. accordingly, the

magnesium can also occupy these spaces under salinity condition leading to higher concentration of magnesium in the leaves (Jalili Marandi et al., 2012; Tavallali et al., 2009). Meanwhile, since the content of calcium and magnesium is increased under salinity condition which might lead to higher competition for potassium and some other proper elements such as iron, lower content of these two elements would be proper in plants. In the current study the content of both calcium and magnesium under salinity treatments was higher in G cultivar compared to their concentrations in the other cultivars. Hokmabadi and Sharafati (2015) reported that concentrations of calcium, phosphorus and magnesium in the leaf of cultivar Akbari are more than those in other cultivars.

### Conclusion

The results of present study indicated that stomatal conductance, photosynthesis rate, chlorophyll content, and Fv/Fm decreased in response to salinity treatments. The main cause of these changes in physiological properties of the pistachio cultivars was pointed out to be the altered content of ions and their competitions for absorption by plant. Despite the probable sufficient amount of potassium in the soil, high concentrations of sodium and other associated elements such as calcium and magnesium prevented the pistachio plants to absorb required content of vital ions such as potassium. As a result, with increased sodium accumulation and lower concentration of potassium,  $K^+/Na^+$  ratio in leaves decreased. Consequently, toxicity of sodium ions in the plant cells reduces stomatal conductance and the rate of photosynthesis along with other physiological properties. Comparison between cultivars showed that for the most of the traits the difference between control and moderate salinity ( $10 \text{ ds m}^{-1}$ ) in all cultivars was not significant. However, G2 cultivar showed higher ability to accumulate potassium and absorb lower

concentrations of sodium, calcium and magnesium under sever salinity treatment ( $20 \text{ ds m}^{-1}$ ). These result suggested that G2 could be considered as a potential tolerant cultivar for cultivation in saline area such as Rafsanjan.

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