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The Evaluation of Inter-Specific Hybrid of *P. atlantica* × *P. vera* cv. 'Badami Zarand' as a Pistachio Rootstock to Salinity Stress

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Abstract

In order to evaluate the inter-specific hybrid of *P. atlantica* × *P. vera* cv. 'Badami Zarand' to salinity stress, an experiment was conducted as a factorial based in a completely randomized design with 0, 60, 120 mM salinity levels of $NaCl_2$, $CaCl_2$ and $MgCl_2$ (3:2:1) on two rootstocks 'Qhazvini' and 'Badami Zarand' and an inter- specific hybrid of *P. atlantica* × *P. vera* cv. 'Badami Zarand'. Mineral concentrations of sodium (Na), chloride (Cl), magnesium (Mg), calcium (Ca), copper (Cu) and iron (Fe) in shoot increased at 120 mM salinity level; however, the phosphorus (P) concentration in root decreased. Rootstocks had significant differences in the concentrations of elements of shoot. The highest concentrations of Na and Cl were observed in 'Badami Zarand' and 'Qhazvini'rootstocks, and the lowest was observed in hybrid rootstock. The highest concentration of Mg was observed in hybrid rootstock, and the lowest was observed in 'Badami Zarand'. Due to restriction in the absorption and transport of chloride and sodium to the shoot, hybrid rootstock could tolerate more salinity than 'Badami Zarand' and 'Qhazvini' rootstocks.

Keywords: Inter specific hybrid, NaCl, Pistachio rootstock, Salinity.

Introduction

Pistachio (*Pistacia vera* L.) is one of the most important commercial crops in Iran (Karimi and Sadeghi-Seresht, 2015). The majority of pistachio orchards is located in areas with saline soil and is irrigated with low quality and saline water (Karimi *et al*., 2015). Although pistachio trees are classified as salt tolerant, previous research has demonstrated that growth rates of pistachio trees decrease with increasing sodium chloride (NaCl) concentration in soil. In addition, there is a positive correlation between sodium (Na) as well as chloride (Cl) concentrations in plant tissue and soil (Picchioni *et al*., 1990; Sepaskhah and Maftoun, 1982). More than 800 million hectares of lands around the world have been affected by salinity, which is equivalent to 6% of total area (Naeini *et al*., 2006). Salinity stress damage plants by osmotic and ion toxicity mechanisms. Mechanism of ionic toxicity related to ion absorption and changes in physiological processes are due to toxicity, shortages or changes in the balance of minerals (Nieves *et al*. 1990).

Picchioni *et al*., (1990) studied the effect of salinity on pistachio rootstocks and reported that with increasing salinity, the root to shoot ratio was increased in all pistachio rootstock, especially in terebinthus rootstock. In other research, Hokmabadi *et al*., (2005) studied theresponse of three pistachio rootstocks ('Badami Zarand', 'Qhazvini' and 'Sarakhs') to salinity stress and reported that shoot dry weight was decreased with

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increasing salinity in all rootstocks, although the most of the shoot dry weight was observed in the ‘Qhazvini rootstock. Doring and Ludders (1986) reported that the Na and Cl concentration of pomegranate leaves increased with increasing NaCl concentrations in culture solution and the highest Na and chloride was observed in root and leaf, respectively.

Water stress is one of the most common environmental stresses that could affect stomatal conductance, photosynthetic rate, morphology of leaves and roots and nutrient balance in plants (Behboudian *et al*., 1986).

It is reported by Karimi *et al*., (2012) that the ‘Badami Zarand’ rootstock is the most common pistachio in Iran and that it is less sensitive to *Phytophtora* and more resistant to salinity compared to ‘Sarakhs’ and *P. mutica*. *P. atlantica* is another rootstock that is utilized in some countries (Karimi *et al*., 2011). It has a resistance to salinity and a high efficiency for zinc absorption compared with other rootstocks (Sepaskhah and Maftoun, 1982). Hence, seedlings produced by crossing ‘Badami Zarand’ and *P. atlantica* may be able to tolerant more salinity than ‘Qhazvini’ rootstock. The objectives of this study was to evaluate the inter-specific hybrid of *P. atlantica* and *P. vera* cv. ‘Badami Zarand’ to salinity stress and compare it with the ‘Qhazvini’ and ‘Badami Zarand’rootstocks.

Materials and Methods

The experiment was conducted in the Research Greenhouse of Horticultural Department, Faculty of Agriculture, Vali-e-Asr University of Rafsanjan in the year 2011. The seeds of ‘Qhazvini’ and ‘Badami Zarand’ cultivars were obtained from the Iranian Pistachio Research Institute (IPRI). Hybrid seeds were produced by crossing a selected *P. atlantica* female tree with a selected *P. vera* ‘Badami Zarand’ male tree during a breeding program in IPRI. Pollen from the stamens of the male tree was collected and introduced to the flowers of the female tree. In the first September, nuts were collected from the tree and were dried in temperature room.

The seeds were planted on plastic pots containing 4200 g of soil. A sandy clay soil with a pH of 7.5 and electrical conductivity of 0.85 $dS.m^{-1}$ was used for this experiment. The experiment was conducted as a factorial based on completely randomized design with 0, 60, 120 mM of salinity levels of $NaCl_2$, $CaCl_2$ and $MgCl_2$ (3:2:1) with four replications on two rootstocks, ‘Qhazvini’ and ‘Badami Zarand,’ and an inter- specific hybrid of *P. atlantica* × *P. vera* cv. ‘Badami Zarand’. Rootstocks were exposed to salinity treatments for 45 days. The temperature of the greenhouse fluctuated between 16 and 35ºC during the experiment. At the end of the experiment, shoots and roots were dried and some nutrient concentrations of shoot and root were measured. Samples of shoot and root were ashed in a muffle oven at 550 ± 25°C. The resulting white ash was then dissolved in 10 mL of 2N hydrochloric acid (HCl) and adjusted to volume of 100 mL for determination of Na, P, Mg, K, Ca, Fe, Zn, Mn, Cu concentration. The Cl concentration was measured according to the method of Chapman (1961). The phosphorous concentration was measured following the method of Olsen *et al*. (1954). Ca, Mg, Fe, Cu, Zn and Mn concentrations were measured using an atomic absorption spectrometry (Model GBC, AVANTA, Australia) and K and Na were measured using a flame photometer (Model PFP7, JENVY, England). The absorption (AR) rate and translocation (TR) ratio calculated according to the following equation:

$$\text{AR (mg/ pot)} = (\text{dry weight of root} \times \text{concentration}) + (\text{dry weight of shoot} \times \text{concentration})$$

$$\text{TR} = \text{AR (shoot)} / \text{AR (root)}$$

Data analysis

Analysis of variance (ANOVA) was performed using the SAS software (SAS Inc., Cary NC). If ANOVA determined that the effects of the treatments were significant ($P<0.05$, $P<0.01$), then the means were compared by Duncan's Multiple Range Test. The graphs were plotted using EXCEL program (Microsoft Crop., Pullman WA).

Results

Shoot and root weight

The shoot fresh weight was significantly affected by the rootstock and salinity ($P < 0.05$) (Table 1). The weight decreased with increasing salinity. The lowest shoot fresh weight was obtained at 60 mM. However, no significant difference was found between the 60 and 120 mM salinity levels. The highest shoot fresh weight was obtained in hybrid rootstock and the lowest was

Table 1. Analysis of variance for Cl absorpsion, Cl translocation, shoot and root nutrient elements and fresh and dry weight of pistachio rootstocks.

Treatment	df	Cl absorpsion mg/pot	Cl translocation (ratio)	Root		Shoot	
				Na	Cl	Ca	K
Salinity	2	*0.645	0.131 **	1.524 **	1.2007 *	0.0892**	0.0015 ns
Rootstock	2	0.6072**	0.203 **	0.271**	3.902**	0.0677**	0.0005 ns
Salinity × Rootstock	4	0.0421 ns	0.0204 ns	0.092 ns	0.287 ns	0.0362*	0.009 ns
Error	18	0.116	0.022	0.053	0.253	0.0126	0.007 ns
Cv		23.8	29.43	26.85	27.28	24.68	12.90

Treatment	df	Shoot					
		Na	Cl	P	Mg	Fe	Cu
Salinity	2	1.524 **	0.335 **	0.00039*	0.0044*	33.57 **	3.903 **
Rootstock	2	0.271**	0.122 *	0.0002 ns	0.0168**	3.65 ns	0.784 ns
Salinity × Rootstock	4	0.092 ns	0.042 ns	0.00018 ns	0.003*	5.65 ns	1.764 **
Error	18	0.053	0.03	0.00016	0.0008	3.679	0.3870
Cv		26.85	24.03	20.85	13.96	16.61	16.70

Treatment	df	Root dry Weight	Root fresh Weight	Shoot dry Weight	Shoot fresh weight
Salinity	2	0.026 ns	0.518 **	0.013ns	0.108 *
Rootstock	2	0.168**	0.507**	0.013*	0.132 *
Salinity × Rootstock	4	0.792**	0.460**	0.009 ns	0.002 ns
Cv	18	20.93	16.45	9.34	11.14

* and* significant at 1 and 5% , respectively; ns: no significant

Table 2. Effects of irrigation water salinity on Cl translocation, Cl absorption and mineral concentration of root and shoot.

Salinity levels	Root		Cl translocation (ratio)	Cl absorption (mg/pot)	
	Na^+ (%)	Cl^- (%)			
0	0.734 b	1.429 b	0.272b	1.44b	
60 mM	0.967ab	2.12 a	0.347ab	2.33a	
120 mM	1.161a	1.982a	0.473a	2.85a	
			Shoot		
Salinity levels	Mg (%)	P (%)	Fe (mg/kgdw)	Cu (mg/kgdw)	Cl (%)
0	0.039b	0.0597a	90.69 b	4.46a	0.2761b
60 mM	0.047ab	0.0543ab	142.14a	3.36b	0.6311a
120 mM	0.0565a	0.0465b	185.23a	3.2b	0.8481a

Means with a common letter in each column are not significantly different (Duncan test, $P = 0.05$).

Root fresh weight also was affected by all treatments (*P < 0.01*) (Table1). The highest root fresh weight was observed in 'Qhazvini' and hybrid rootstock. The lowest was found in 'Badami Zarand'. The results of interaction of salinity and rootstock on root fresh weight and dry weight showed that in 'Badami Zarand,' root fresh weight and dry weight were decreased at 120 mM salinity (Fig. 1). The highest root dry weight was observed in hybrid at 120 mM salinity (Fig. 2).

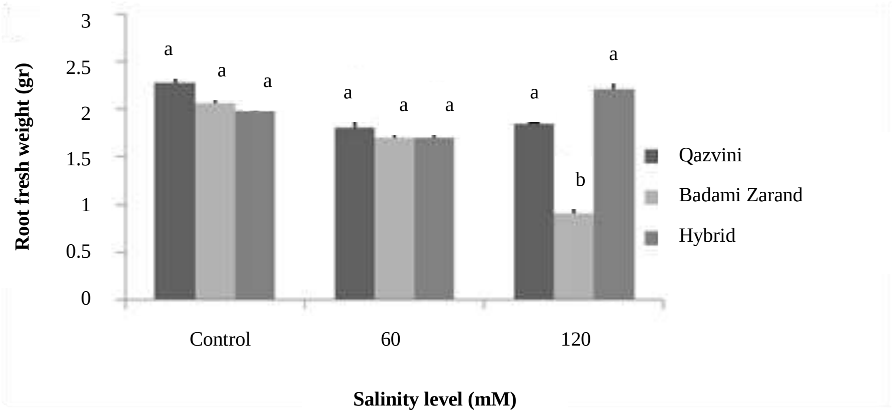

Fig 1. Interaction of salinity and rootstock on root fresh weight.

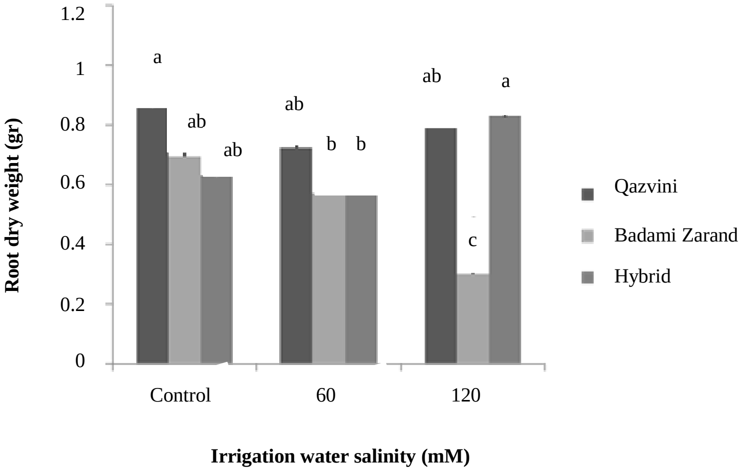

Fig 2. Interaction of salinity and rootstock on root dry weight.

Na and Ca concentration of shoot

The sodium concentration of shoot was significantly affected by salinity and rootstock (Table 1). Shoot Na concentration increased with increasing salinity. The highest shoot Na concentration was observed at 120 mM salinity level, and the lowest was observed in the control (0 mM) (Table 2). The data showed that there was a significant difference between the rootstocks. The highest shoot Na concentration was obtained with ‘Badami Zarand’ rootstock, and the lowest was found in hybrid rootstock (Table 3). Ca concentration of shoot increased significantly in ‘Badami Zarand’ rootstock, whereas it was not affected by salinity in the hybrid rootstock (Fig. 3).

Table 3. The effect of rootstock on Na and Cl concentration of shoot and root and Cl translocation ratio.

Tissue	Element	Rootstock		
		Hybrid	‘Qazvini’	‘Badami-Rize-Zarand’
Shoot	Na (%)	0.98 a	0.85 ab	0.65 b
Shoot	Cl (mg/kg dw)	0.40 b	0.63 ab	0.75 a
Root		2.50 a	1.20 c	1.80 b
Translocation ratio		0.17 b	0.5 a	0.48 a

Means with a common letter in each column are not significantly different (Duncan test, *P* = 0.05).

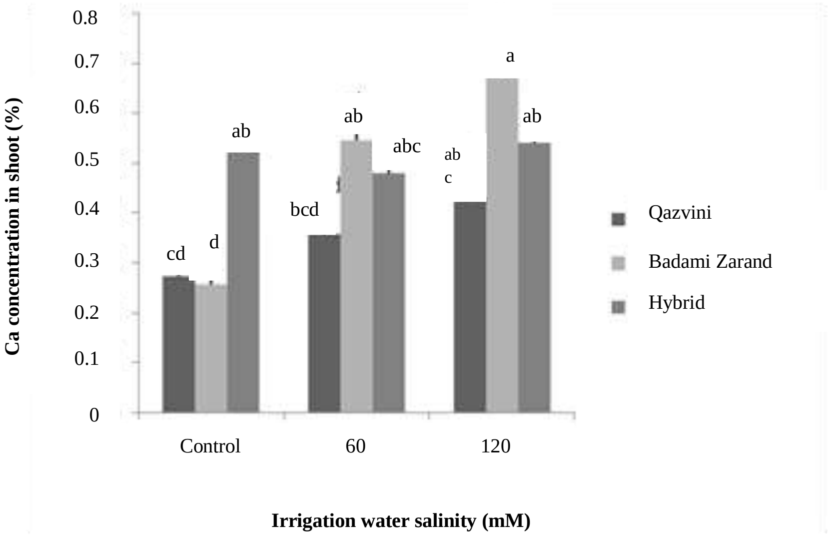

Fig 3. Interaction of salinity and rootstock on Ca concentration of shoot.

Mg and K concentration of shoot

The magnesium concentration of shoot was significantly affected by all treatments (salinity and rootstock) (Table 1).The results of the interaction of salinity and rootstock on shoot Mg concentrations showed that in 'Badami Zarand' rootstock, the shoot Mg concentration increased with increasing salinity, whereas in the hybrid and 'Qhazvini' rootstocks, there were no significant difference between levels of salinity (Fig. 4). Shoot K concentration was unaffected by treatments (salinity and rootstock) (Table 1).

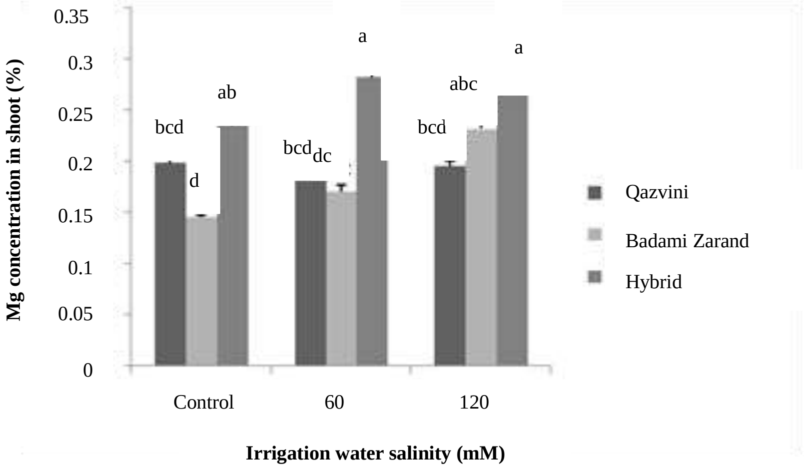

Fig 4. Interaction of salinity and rootstock on Mg concentration of shoot.

P and Cl concentration of shoot

The potassium concentration of shoot was only affected by salinity and rootstock. The interaction salinity and rootstock was not significant. Shoot P concentration significantly decreased at 120 mM, although there was not a significant difference between 60 mM and control (Table 2). Shoot Cl concentration was significantly affected by salinity and rootstock (Table1). Salinity significantly increased Cl concentrations of shoot. However, there was not a significant difference between 60 and 120 mM of salinity levels (Table 2). Rootstocks had significant differences related to shoot Cl concentrations. The highest shoot Cl concentration was obtained in 'Badami Zarand' rootstock, and the lowest was found in hybrid rootstock (Table 3).

Fe and Cu concentration of shoot

Iron concentration of shoot was only affected by salinity. The interaction between salinity and rootstock was not significant. Salinity significantly increased Fe concentrations of shoot, although there were no significant differences between 60 and 120 mM of salinity levels (Table 2). The results of variance analysis showed that Cu concentration of shoot was significantly affected by salinity and interaction between salinity and rootstock (*$P<0.01$*) (Table 1). The results of interaction of salinity and rootstock on shoot Cu concentration showed that in hybrid and 'Badami Zarand' rootstocks, shoot Cu concentration decreased with increasing salinity, whereas in the 'Qhazvini' rootstock, there was no significant differences between 60 and 120 mM of salinity levels (Fig. 5).

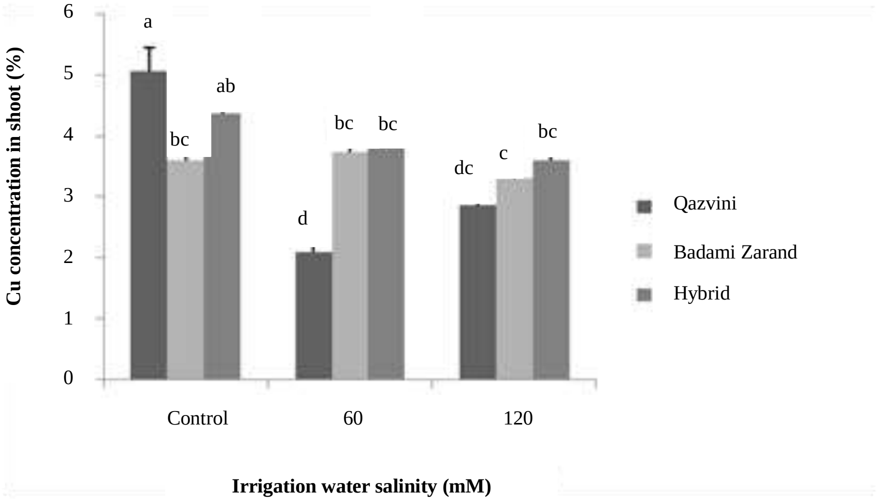

Fig 5. Interaction of salinity and rootstock on Cu concentration of shoot.

Na and Cl concentration of root

Salinity significantly increased Na concentrations of shoot. There was not a significant difference between 60 and 120 mM of salinity levels (Table 2). The results of analysis of variance data related to root Cl concentration showed that root Cl concentration was significantly affected by salinity and rootstock (Table 1). There were no significant differences between 60 and 120 mM of salinity levels (Table 3). Three rootstocks showed significant differences in root Cl concentration in that the highest value was observed in hybrid rootstock and the lowest was found in Qhazvini rootstock (Table 2).

Cl absorbtion and translocation

The results of analysis of variance data related to root Cl absorption and translocation showed that Cl absorption and translocation were affected by salinity and rootstock (Table 1).

Cl absorption and translocation significantly increased at 120 mM salinity level. However, there was not a significant difference between 60 mM and control (Table 2). The rootstocks were significantly different in regards to the Cl translocation ratio. The highest of Cl translocation was obtained with the hybrid rootstock and the lowest was found in 'Qhazvini.' There were no significant differences between 'Badami Zarand' and 'Qhazvini' rootstocks.

Discussion

This study showed the salinity treatment led to a reduction of shoot and root weight in three pistachio rootstocks, which is in agreement with Saadatmand *et al*. (2007), Picchioni *et al*., (1990), Walker *et al*. (1987,1988), Behboudian *et al*. (1986) and Sepaskhah and Maftoun (1982). The comparison of fresh weight with dry weight of shoot showed that salinity affected fresh weight more than dry weight. This suggested that leaf fresh weight was more affected by water than specific-ion-toxicity. Salinity also increased Na and Ca concentration of shoot in 'Badami Zarand' rootstock. This results confirmed previous reports of Walker *et al*. (1987, 1988), Tajabadipure *et al*. (2005), Saadatmand *et al*. (2007) and Karimi *et al*. (2011) in that increasing salinity shoot resulted in an increase of Na and Ca concentration in pistachio. The results of this study confirmed previous reports (Banakar and Ranjbar, 2010; Karimi *et al*., 2011) that genotypes of pistachio that have low Na concentration in shoot are resistant to salinity stress.

The results also showed that K concentration of shoot was unaffected by salinity and rootstock. Previous studies reported that salinity decreased K concentration in pistachio seedling (Picchioni *et al*., 1990) and pomegranate cutting (Naeini *et al*., 2006). This differenence was likely related to the kind of salts used. Saadatmand *et al*. (2007) studied the effects of NaCl salinity on pistachio seedling and reported that salinity decreased K concentration in shoot.

Salinity significantly increased Cl concentrations of shoot. The highest shoot Cl concentration was obtained in 'Badami Zarand' rootstock, and the lowest was found in hybrid rootstock. This result is in agreement with Ferguson *et al*. (2002), who reported that Cl concentration in shoot of 'Kerman' grafted on *P. atlantica* and UCB_1 rootstocks was lower than *P. integrima* rootstock. Ferguson *et al*. (2002) also reported that with increasing of NaCl level in medium, the accumulation of Na, K, and Cl in shoot and root of the pistachio rootstocks were increased.

This study showed that in hybrid rootstock, Cl accumulated in root led to more resistance to salinity than to Badami Zarand'and 'Qhazvini' rootstocks. It is reported that the root system in *P. atlantica* rootstock retained sodium and chlorine and transferred less to shoot. Lessani and Marschner (1978) believed that this could be due to lack of mechanisms to maintain ionic balance within the plant. Therefore, salinity disturbs ionic balance within the plant, resulting in an accumulation of chlorine in plant tissue. Our result is in accordance with Grieve and Walker (1983) and Sepaskhah and Maftoun (1982), who reported that the sensitivity of citrus and pistachio varieties were associated with the accumulation of excessive amount of Cl and sometimes Na in the leaves.

Conclusions

The results of our study showed that the hybrid rootstock had good growth vigor and an exclusion mechanism that restricted the uptake or transport of Na or Cl. The results of absorption and translocation Cl and Na showed that the hybrid rootstock may maintain sodium and chloride in the parenchyma cells of the root xylem. This will prevent the translocation of sodium into the shoots and could be considered as an avoidance mechanism of salinity resistance.

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References

Ashraf M, Mcneilly T (1990) Responses of four Brassica species to sodium chloride.

Environmental and Experimental Botany. 30, 475-487.

Banakar MH, Ranjbar GH (2010) Evaluation of salt tolerance of pistachio cultivars at seedling stage. American-Eurasian Journal of Agricultural and Environmental Sciences. 9, 115-120

Behboudian MH, Walker RR, TorokfalvyE (1986) Effect of water stress and salinity on photosynthesis of pistachio. Scientia Horticulture. 29, 251-261.

Chapman HD, Prah DF (1961) Methods of analysis for soil, plants and water. University of California Division Agricultural Science. Pp, 60-62.

Dahiya SS, Singh M (1976) Effect of salinity, alkalinity and iron application on availability of iron, manganese, phosphorus and sodium in pea (*Pisum sativum)* crop. Plant Soil. 44, 697-702.

Demiral MA (2005) Comparative response of two olive (*Olea europaea*) cultivars to salinity. Turkish Journal of Agriculture. 25, 267-274

Doring J, Ludders P (1986) Effect of different salt treatment of *Punica granatum* L. at different root treatment. Artenbauwissenschaft. 52, 92-96.

Duran-Zuazo VH, Martinez-Raya H, Aguilar-Ruiz J (2003) Salt tolerance of mango rootstock (*Magnifera indica* L. cv. Osteen). Spanish Journal of Agricultural Research. 1, 67-78.

Fergusen L, Poss JA, Grattan SR, Grieve GM, Wang D, Wilson C, Donovan-Chao CT (2002) Pistachio rootstocks influence scion growth ion relations under salinity and boron stress. Journal of the American Society for Horticultural Science. 127, 194-199.

Garcia-Sanchez F, Jifon JL, Carrajal M, Syvertsen JP (2002) Gas exchange, chlorophyll and nutrient contention relation to Na^+ and Cl^- accumulation in Sunburst mandarin grafted on differnent rootstocks. Plant Science. 162, 705-712.

Grattan SR, Grieve CM (1999) Salinity-mineral nutrient relations in horticultural crops. Scientia. Horticulturae. 78, 127-157.

Hasanpour Z, Karimi HR, Mirdehghan SH (2015) Effects of salinity and water stress on echophysiological parameters and micronutrients concentration of pomegranate (*Punica granatum* L.). Journal of Plant Nutrition. 38, 1-13.

Hokmabadi H, Arzani K, Grierson PF (2005) Growth, chemical composition, and carbon isotope discrimination of pistachio (*Pistacia vera* L.) rootstock seedlings in response to salinity. Australian Journal of Agricultural Research. 56, 135-144.

Karimi H (2012) Evaluation of the behavior of native Iranian pistachio species as rootstocks. Journals of Nuts. 3, 41-46.

Karimi HR, Ebadi A, Zamani Z, Fatahi R (2011) Effect of water salinity on growth indices physiological parameters in some pistachio rootstocks. Journal of Plant Nutrition. 34, 935-944.

Karimi HR, Hasanpour Z (2014) Effects of salinity and water stress on growth and macronutrients concentration of pomegranate (*Punica granatum* L.). Journal of Plant Nutrition. 37, 1-15.

Karimi HR, Sadeghi-Seresht E (2015) Investigation of genetic nature and diversity of Banebaghi as genotype of *Pistacia* genus by using morphological characteristics and RAPD marker. Plant Cell Biotechnology and Molecular Biology. In press.

Karimi HR, Nasrolahpour-Moghadam S, Mohammadi Mirik AA (2015) Variation and correlation between seeds and their seedlings traits in pistachio (*Pistacia vera* L.) in separation sex. Plant Cell Biotechnology and Molecular Biology. In press.

Lessani H, Marschner H (1978) Relation between salt tolerance and long-distance transport of sodium and cloride in various crop species. Australian Journal of Plant Physiology. 5, 27-37.

Matsumoto K., Tamura F, Chun J, Tanabe K (2006) Native mediterranean *Pyrus* rootstock, *P. amygdaliformis* and *P. elaeagrifolia*, present higher tolerance to salinity stress compared with asian natives. Japanese Society for Horticultural Science. 75, 450-457.

Munns R (2002) Comparative physiology of salt and water stress. Plant Cell Environmental. 25, 239-250.

Naeini MR, Khoshgoftarmanesh AH, Fallahi E (2006) Partitioning of chlorine, sodium, and potassium and shoot growth of three pomegranate cultivars under different levels of salinity. Journal of Plant Nutrition. 29, 1835-1843.

Nieves M, Martinez V, Cerda A., Guillén MG (1990) Yield and mineral composition of ‘‘Verna’’ lemon trees as affected by salinity and rootstock combination. Journal of Horticultural Science. 65, 59-366

Olsen SR, Cole CV, Watanbe FS, Dean LA (1954) Estimation of available phosphorous in soil by extraction with sodium bicarbonate. USDA Circ. 939, U.S. Government. Printing. Office, Washington, DC., USA. Pp. 1-19.

Parsa AA, Karimian N (1975) Effect of sodium chloride on seedling growth of two major varieties of Iranian pistachio (*P. vera* L.). Journal of Horticultural Science. 50, 41-46.

Picchioni GA, Miyamoto S, Storey JB (1990) Salt effects on growth and ion uptake of pistachio rootstock seedlings. Journal of the American Society for Horticultural Science. 115, 647–653.

Saadatmand AR, Banihashemi Z, Maftoun M, Sepaskhah AR (2007) Interactive effects of soil salinity and water stress on growth and chemical compositions of pistachio nut trees. Journal of Plant Nutrition. 30, 2037–2050.

Sepaskhah AR, Maftoun M (1982) Growth and chemical composition of pistachio cultivars as influenced by irrigation regimes and salinity levels of irrigation water. II. Chemical composition. Journal of Horticultural Science. 56, 227–284.

Tabatabaei SJ (2006) Effects of salinity and N on the growth, photosynthesis and N status of olive (*Olea europaea* L.) trees. Scientia Horticulturae. 108, 432- 438

Tajabadipour A, Sapaskhah A, Maftoun M (2005) Plant water relations and seedling growth of tree pistachio cultivars as influence by irrigation frequency and applied potassium. Journal of Plant Nutrition. 28, 1413-1425.

Walker RR, Torokfaluy E, Behboudian MH (1988) Photosynthetic rates and solute partitioning in relation to growth of salt treated pistachio plants. Plant Physiology. 15, 87-98

Walker RR, Torokfalvy E, Behboudian MH (1987) Uptake and distribution of chloride, sodium and potassium ions and growth of salt-treated pistachio plants. Australian Journal of Agricultural Research. 38, 383–394.