

## Short term response of grapevine grown hydroponically to salinity: Mineral composition and growth parameters

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### Summary

The response of hydroponically grown four own-rooted table grape (*Vitis vinifera* L.) cultivars ('Red Rishbaba', 'Red Sahebi', 'Dastarchin' and 'Red Sultana') to different salt concentrations (0, 25, 50 and 100 mM NaCl) was studied under greenhouse condition. Growth parameters, total chlorophyll (a+b) and proline contents were determined in leaves and roots. Cl<sup>-</sup>, Na<sup>+</sup>, K<sup>+</sup> and NO<sub>3</sub><sup>-</sup> concentrations were measured in lamina, petiole, stem and root. Shoot growth, total dry weight, total leaf chlorophyll (a+b), NO<sub>3</sub><sup>-</sup>-N and K<sup>+</sup> contents were significantly reduced ( $P \leq 0.05$ ) under NaCl stress, whereas proline, Cl<sup>-</sup> and Na<sup>+</sup> accumulation increased significantly with increasing salinity. 'Red Rishbaba' and 'Red Sahebi' showed a less decrease in total leaf chlorophyll, K<sup>+</sup> and NO<sub>3</sub><sup>-</sup> contents, while proline accumulation in these cultivars was higher than that of 'Dastarchin' and 'Red Sultana'. Also, shoots of 'Red Sahebi' and 'Red Rishbaba' accumulated lower Cl<sup>-</sup> and Na<sup>+</sup> than other cultivars. In conclusion, the overall results showed that 'Red Rishbaba' and 'Red Sahebi' were more tolerant than 'Dastarchin' and 'Red Sultana'.

**Key words:** grapevine, salt tolerance, ion content, proline, total chlorophyll.

**Abbreviations:** r-Rish: 'Red Rishbaba', r-Sah: 'Red Sahebi', Das: 'Dastarchin', r-Sul: 'Red Sultana', Chl: Chlorophyll.

### Introduction

Soil salinity is one the biggest problems for crop production in many areas of the world (MUNNS 2002). Grapevines are considered moderately sensitive to root-zone salinity (FISARAKIS *et al.* 2001) and the damage is primarily caused by chloride ions (WALKER 1994). The effects of salinity are attributed mainly to high concentrations of Cl<sup>-</sup> and Na<sup>+</sup> which negatively affect the uptake of some nutrient elements such as NO<sub>3</sub><sup>-</sup>, K<sup>+</sup> and Ca<sup>2+</sup> (HASAGAWA *et al.* 2000). GARCIA and CHARBAJI (1989) and DOWNTON (1985) showed a Na<sup>+</sup>- K<sup>+</sup> and Na<sup>+</sup>- Ca<sup>2+</sup> antagonism in grapevine respectively. Some of the grapevine rootstocks have been rated as tolerant to salinity due to their ability to prevent Na<sup>+</sup> and / or Cl<sup>-</sup> uptake and translocation to aerial parts of

the vines (GALET 1991). When used as a grapevine rootstock, 140 Ruggeri (*Vitis berlandieri* × *Vitis rupestris*) is known for its ability as a Cl<sup>-</sup> excluder, whereas K 51-40 (*Vitis champinii* × *Vitis riparia* 'Gloire') is a poor excluder and scions grafted to it accumulate high concentrations of Cl<sup>-</sup> when grown under saline conditions (TREGAGLE *et al.* 2006, WALKER *et al.* 2010). Selection of salt tolerant plants has been the main objective of many researchers. There are less studies on the relationship between Cl<sup>-</sup> exclusion capacity and salt tolerance of grapevine genotypes grown in Iran. The aim of this study was evaluation of differences between some grapevine varieties from the view point of Cl<sup>-</sup> transport to the shoot. Also, the effects of NaCl on mineral and biochemical compositions of the genotypes were investigated.

### Material and Methods

The two to four nodal, hard-wood cuttings of four own-rooted grapevine (*Vitis vinifera*) cultivars; 'Red Rishbaba', 'Red Sahebi', 'Dastarchin' and 'Red Sultana' were collected from Kahriz vineyard, Urmia, Iran. The basal part of the cuttings were soaked IBA (0.1 % w/v in 50 % ethanol) for 5-10 s. All cuttings were in a mist house with a heat-bed temperature of 25 °C and hygrometry of 70-85 %. After the appearance of leaves, rooted cuttings were transferred to hydroponic cultures in 2-L pots containing aerated Hoagland nutrient solution. At the beginning of salinity treatments, uniform plants were selected with a shoot of (average) 35 cm in length for all varieties, and exposed to various concentrations of NaCl (0, 25, 50 and 100 mM). After the stress period (2 weeks) roots, stems, petioles and leaves of plants were separately harvested. The plant materials were washed with distilled water, dried at 70 °C for 48 h, and ground to provide a powder. Experimental design was a randomized 4\*4 factorial, replicated three times. Factors tested were salinity (4 levels), and grapevine (4 variety).

**Growth analysis:** Shoot, root lengths (the longest main root) and leaf number were measured on plants from each treatment at the beginning of salinity treatments and at the end of salt period. It was calculated by using the equation: (e.g. Shoot length =  $L_f - L_i$  where L is the shoot length, and subscripts denote initial (i) and final (f) measurement.) At the end of the experiment the fresh and dry weights of shoot and root were determined.

Total chlorophyll (chlorophyll a+b) was analyzed following the method of LICHTENTHALER and WELLBURN (1985).

Proline content was determined according to (BATES *et al.* 1973). Determinations of tissue mineral contents, inorganic ions were extracted from dry matter with hot water. In the extract, the sodium and potassium concentrations were measured by flame photometer (Fater electronic 405) and the chloride by silver ion titration using a chloride meter (Corning 926).  $\text{NO}_3^-$ -N was determined by salicylic sulfuric acid method (CATALDO 1975).

Analysis of variance was performed by the statistical program SpSS version 18 and interactions between cultivar and salinity levels were determined using GLM (General Linear Model).

## Results

Shoot and root lengths were significantly reduced at all salinity levels (Tab.1). The decrease of growth in Das and r-Sul were higher than that of r-Rish and r-Sah cultivars. The accumulation of dry matter decreased more in shoots than in root. R-Rish showed higher root/shoot ratio than all cultivars (Tab. 1). The leaf number was significantly de-

creased (Tab. 1). The decrease was calculated as 79, 67, 85 and 80 % in r-Rish, r-Sah, r-Sul and Das respectively at 100 mM NaCl (data not presented). The total chlorophyll (a+b) content of leaves were significantly decreased ( $p < 0.05$ ) by salinity. However, decrease chlorophyll content in Das and r-Sul cultivars were higher than that of r-Rish and r-Sah (Tab. 1). The reduction in total chlorophyll content due to increased salt treatments from 0 to 100 mM NaCl were 44, 43, 57 and 57 % in leaves for the r-Rish, r-Sah, r-Sul and Das respectively (data not presented). Proline, as an osmoregulator in stress conditions, increased in lamina and root of four grapevine cultivars. A positive correlation was determined between proline concentrations and tissue Cl<sup>-</sup> and Na<sup>+</sup> concentrations of lamina (r: from 0.881 to 0.964,  $p < 0.01$ ) and root (r: from 0.894 to 0.967,  $p < 0.01$ ) during salt stress. However, r-Rish and r-Sah had higher proline content than the others (Tab. 1).

Vine tissue (root, stem, petiole and lamina) concentration of Na<sup>+</sup> increased significantly with increasing salinity in the nutrient solution. Roots accumulated higher amounts of Na<sup>+</sup> than other plant parts (Tab. 2). Among the cultivars, shoots (stem, petiole plus lamina) of r-Sul and Das

Table 1

The response of shoot and root lengths, leaf number, root/shoot ratio, total chlorophyll (chlorophyll a+b) and proline contents of r-Rish, r-Sah, r-Sul and Das varieties to salinity, 2 weeks after salt treatment<sup>a,b</sup>

Salinity (mMNaCl)	Shoot length (cm)	Root length (cm)	Leaf number per plant	Root/shoot ratio	Total Chl (mgg <sup>-1</sup> fw)	Lamina Proline (μgg <sup>-1</sup> dw)	Root Proline (μgg <sup>-1</sup> dw)
0	31.291 a	6.666 a	10.750 a	.186 d	20.263 a	.7694 d	1.089 d
25	20.916 b	5.250 b	8.000 b	.2096 c	16.529 b	1.6759 c	1.715 c
50	15.250 c	3.875 c	5.416 c	.2384 b	12.860 c	2.7093 b	2.591 b
100	4.541d	1.541 d	2.333 d	.2573 a	10.084 d	3.7472 a	3.142 a
Variety							
r-Rish	16 c	4.416 b	6 c	0.256 a	14.76 b	2.529 a	2.219 a
r-Sah	16.416 c	5.08 a	7.33 a	0.22 b	16.65 a	2.305 b	2.217 a
r-Sul	21.58 a	4.37 b	6.25 bc	0.19 c	14.45 b	2.075 c	1.976 b
Das	18 b	3.458 c	6.91 ab	0.225 b	13.86 c	1.99 c	2.125 a

<sup>a</sup>For salinity means calculated for all variety and for varieties means calculated for all salinity treatments.

<sup>b</sup>Different letters within columns indicate significant differences at  $P < 0.05$  (LSD,  $n = 3$ ).

Table 2

The effect of external NaCl concentrations on Na<sup>+</sup> content (% d.w.) of lamina, petiole, stem and root in r-Rish, r-Sah, r-Sul and Das, 2 weeks after salt treatment<sup>a,b</sup>

Salinity (mMNaCl)	Lamina	petiole	stem	Root
0	.110 d	.225 d	.167 d	.333 d
25	1.103 c	3.090 c	2.543 c	3.570 c
50	1.690 b	4.316 b	3.170 b	4.663 b
100	2.303 a	5.623 a	3.996 a	5.890 a
Variety				
r-Rish	1.203 b	2.913 d	2.46 a	3.913 a
r-Sah	1.118 b	3.215 c	2.505 a	3.721 ab
r-Sul	1.410 a	3.683 a	2.382 a	3.523 bc
Das	1.475 a	3.443 b	2.530 a	3.298 c

<sup>a</sup>For salinity means calculated for all variety and for varieties means calculated for all salinity treatments.

<sup>b</sup>Different letters within columns indicate significant differences at  $P < 0.05$  (LSD,  $n = 3$ ).

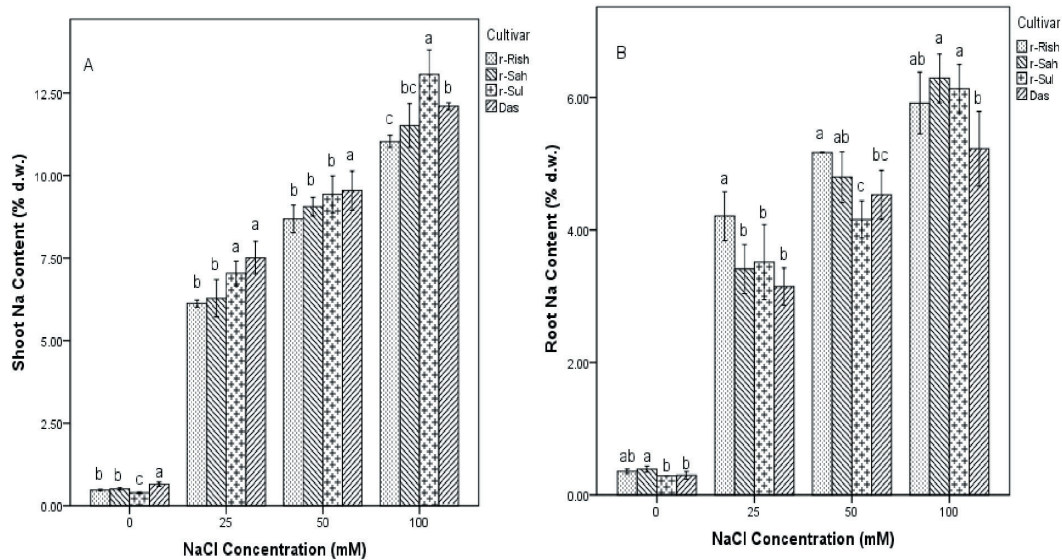


Fig. 1: Sodium concentration in shoot (stem, petiole plus lamina) (A) and root (B) of r-Rish, r-Sah, r-Sul and Das treated with different concentrations of NaCl for 2 weeks. Bars are  $\pm$  SE of the means ( $n = 3$ ) LSD  $p \leq 0.05$ . Different letters indicate significant differences between varieties at each salt concentration.

accumulated higher  $\text{Na}^+$  content than the others (Fig. 1). R-Sul and Das retained maximal  $\text{Na}^+$  concentration in petioles and lamina, while r-Sah and r-Rish retained lower  $\text{Na}^+$  concentration.

Chloride concentration in petiole, lamina, stem and root increased with increasing external salinity. Among the different parts of the plants, roots and petioles showed the highest  $\text{Cl}^-$  concentration. Leaves of r-Rish and r-Sah accumulated lower  $\text{Cl}^-$  than the other cultivars (Tab. 3). R-Sul and Das accumulated more  $\text{Cl}^-$  in the shoot (Fig. 2). However, root  $\text{Cl}^-$  content of r-Sah, r-Rish and r-Sul was similar. Das is able to maintain lower  $\text{Cl}^-$  in the root. While, it seems that r-Sah and r-Rish are partially able to restrict  $\text{Cl}^-$  transport to shoot when compared to the other cultivars.

Increasing salinity led to a decline in potassium concentration in all plant parts. Among the grapevine cultivars

the highest  $\text{K}^+$  concentration was determined in the leaf blades and petioles of r-Sah (Tab. 4). Also, shoot  $\text{K}^+$  content of r-Sah was higher than that of the others (Fig. 3). However, Das had the highest root  $\text{K}^+$  concentration (Fig. 3). Among the different parts of the plant, petioles showed the highest  $\text{K}^+$  concentration, whereas the lowest values were in the stems.

Results presented in Fig. 4 indicate that raising NaCl concentrations in solution the  $\text{NO}_3^-$ -N concentration were reduced in shoot and root. Das had significantly lower lamina and petiole  $\text{NO}_3^-$  concentrations than those of r-Sah, r-Rish and r-Sul cultivars (Tab. 4). However, shoot  $\text{NO}_3^-$  content of r-Sah and r-Rish was higher than that of the Das and r-Sul cultivars. Total  $\text{NO}_3^-$  content in roots of r-Sul was higher than that of r-Sah, r-Rish and Das. While, at 50 and 100 mM external NaCl r-Sah and r-Rish showed the highest  $\text{NO}_3^-$  concentrations, compared to other cultivars (Fig. 4).

Table 3

The effect of external NaCl concentrations on  $\text{Cl}^-$  content (% d.w.) of lamina, petiole, stem and root in r-Rish, r-Sah, r-Sul and Das, 2 weeks after salt treatment<sup>a,b</sup>

Salinity (mMNaCl)	Lamina	Petiole	Stem	Root
0	.145 d	.244 d	.180 d	.375 d
25	.451 c	.869 c	.589 c	1.675 c
50	.980 b	1.345 b	.895 b	2.310 b
100	1.504 a	2.047 a	1.358 a	2.497 a
Variety				
r-Rish	0.591 c	1.107 b	0.736 b	1.779 a
r-Sah	0.705 b	1.1 b	0.703 b	1.785 a
r-Sul	0.879 a	1.243 a	0.838 a	1.798 a
Das	0.905 a	1.055 b	0.745 b	1.496 b

<sup>a</sup> For salinity means calculated for all variety and for varieties means calculated for all salinity treatments.

<sup>b</sup> Different letters within columns indicate significant differences at  $P < 0.05$  (LSD,  $n = 3$ )

## Discussion

Increased root zone salinity led to decline in the growth of all vines. At 25 mM, the reduction in shoot dry weight (stem, petiole plus lamina) was comparable with that of the roots, indicating that shoot and roots were equally sensitive to NaCl salinity. At 50 and 100 mM, root growth was less affected than that of shoots, so that root/shoot ratio increased by 28 and 38 % respectively (Tab. 1). This result may be due to a greater ability of the roots for osmotic adjustment under stress (SHARP and DAVIES 1979). It has been well known that the roots are actively involved in salt stress adaptation (NAIK and WILDHOLM 1993). Reduction in growth due to salinity seems to be related to the sum of the total cations in the leaves (ARBAZADEH and DUTT 1987). Total chlorophyll (a+b) content was significantly decreased. Ion accumulation in leaves adversely affects Chl content (YEO *et al.* 1985). The reduction of chlorophyll contents in abiotic stress plants might possibly be due to



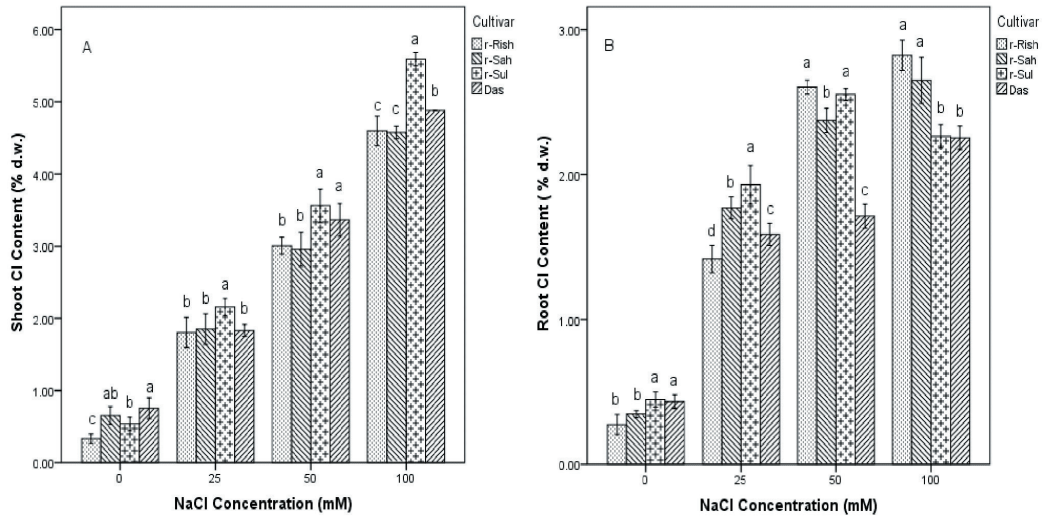


Fig. 2: Chloride concentration in shoot (stem, petiole plus lamina) (A) and root (B) of r-Rish, r-Sah, r-Sul and Das treated with different concentrations of NaCl for 2 weeks. Bars are ±SE of the means (n = 3) LSD p ≤ 0.05. Different letters indicate significant differences between varieties at each salt concentration.

Table 4

The effect of external NaCl concentrations on K<sup>+</sup> and NO<sub>3</sub><sup>-</sup> contents (% d.w.) of lamina, petiole, stem and root in r-Rish, r-Sah, r-Sul and Das, 2 weeks after all salt treatments<sup>a,b</sup>

Salinity (mMNaCl)	Potassium				NO <sub>3</sub> <sup>-</sup> -N			
	Lamina	Petiole	Stem	Root	Lamina	Petiole	Stem	Root
0	4.576 a	9.283 a	4.510 a	7.656 a	.292 a	.635 a	.272 a	.690 a
25	4.190 b	8.430 b	4.083 b	6.496 b	.268 b	.547 b	.231 b	.624 b
50	4.096 b	7.803 c	3.630 c	5.123 c	.238 c	.459 c	.190 c	.513 c
100	3.710 c	7.363 d	2.896 d	3.723 d	.207 d	.388 d	.159 d	.458 d
Variety								
r-Rish	4.176 b	8.523 a	3.643 c	5.416 c	0.284 b	0.572 a	0.206 b	.604 a
r-Sah	4.563 a	8.763 a	3.83 b	5.856 c	0.296 a	0.511 b	0.237 a	.560 b
r-Sul	3.923 c	7.937 b	3.5 c	5.483 b	0.231 c	0.521 b	0.209 b	.615 a
Das	3.91 c	7.656 b	4.136 a	6.243 a	0.194 d	0.425 c	0.201 b	.507 c

<sup>a</sup>For salinity means calculated for all variety and for varieties means calculated for all salinity treatments.

<sup>b</sup>Different letters within columns indicate significant differences at P < 0.05 (LSD, n = 3).

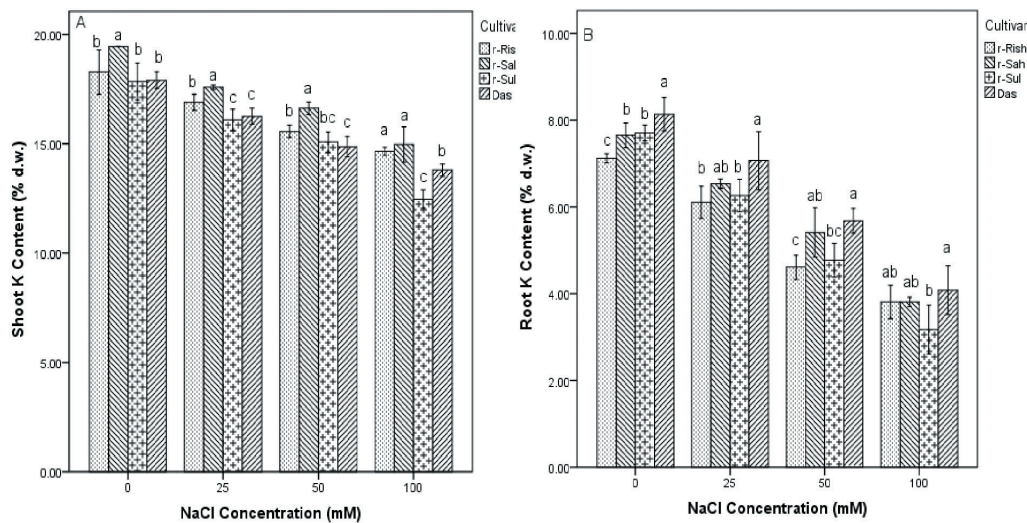


Fig. 3: Potassium concentration in shoot (stem, petiole plus lamina) (A) and root (B) of r-Rish, r-Sah, r-Sul and Das treated with different concentrations of NaCl for 2 weeks. Bars are ±SE of the means (n = 3) LSD p ≤ 0.05. Different letters indicate significant differences between varieties at each salt concentration.

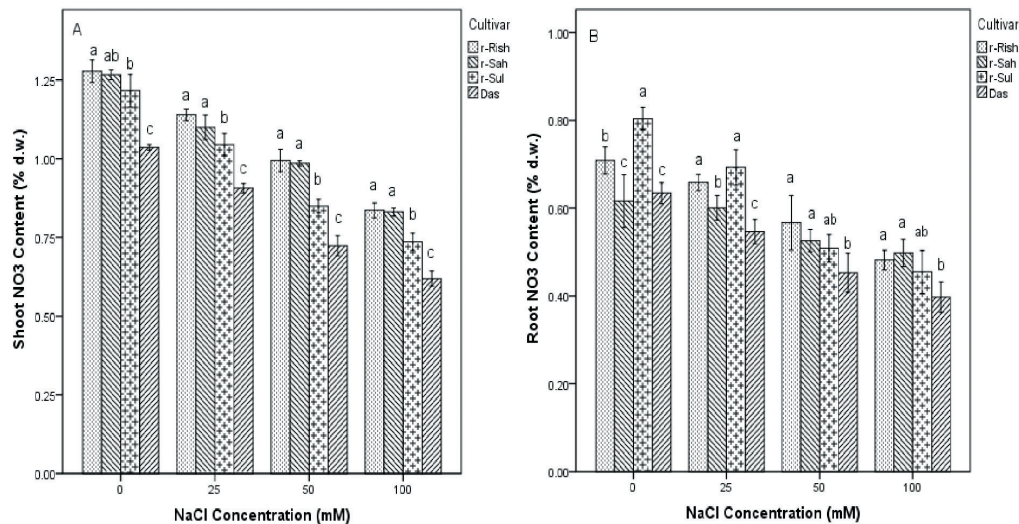


Fig. 4: Total NO<sub>3</sub><sup>-</sup> concentration in shoot (stem, petiole plus lamina) (A) and root (B) of r-Rish, r-Sah, r-Sul and Das treated with different concentrations of NaCl for 2 weeks. Bars are ± SE of the means (n = 3) LSD p ≤ 0.05. Different letters indicate significant differences between varieties at each salt concentration.

changes in the lipid protein ratio of pigment-protein complexes or increased chlorophyllase activity (PARIDA *et al.* 2004). Moreover, reduction in chlorophyll concentrations is probably due to the inhibitory effect of the accumulated ions of various salts on the biosynthesis of the different chlorophyll fractions (CHUTIPAJIT *et al.* 2008). The levels of proline in the plants mainly designate their ability to tolerate or adapt to saline conditions. Proline has also been proposed to function as molecular chaperone stabilizing the structure of proteins, and proline accumulation can provide a way to buffer cytosolic pH and to balance cell redox status (CHUTIPAJIT *et al.* 2008). Proline was accumulated by increasing salinity in most of the grapevine cultivars. Increase in the proline content in the stressed tissue under *in vitro* condition has been observed by GREENWAY and MUNNS (1980). Under saline condition, the decrease of nutrient uptake results in a lower transport rate of nutrients to the top and, therefore, in a lower shoot and root growth (FISARAKIS *et al.* 2004). The present investigation demonstrated that salinity and cultivar can modify K<sup>+</sup> and NO<sub>3</sub><sup>-</sup>-N nutrient concentration in grapevine plants. This may be due, in some cases to differential cultivar response to salinity and may have a considerable effect on growth of grapevines under saline conditions. Under salt stress, the K<sup>+</sup> concentration decreased in all vine tissues, same as in many glycophytes (GREENWAY and MUNNS 1980). Shoots and roots of all four cultivars showed an increase in Na<sup>+</sup> and a reduction in K<sup>+</sup> contents during salt treatment. Decrease of K<sup>+</sup> concentration in roots resulted in enhanced Na<sup>+</sup>/K<sup>+</sup> ratio that leads to a favorable ionic balance in the cases of increased Na<sup>+</sup> uptake (WEST 1986). Similar reductions in root K<sup>+</sup> concentrations with increasing salinity have been observed in citrus (WALKER 1986). It is well known that many K<sup>+</sup> transport systems have significant affinity for Na<sup>+</sup> (BLUMWALD *et al.* 2000). The decrease of K<sup>+</sup> concentration by salinity has been already reported (GARCIA and CHARBAJI 1993, TRONCOSO *et al.* 1999). KENT and LAUCHLI (1985) observed that the contents of K<sup>+</sup> and Ca<sup>2+</sup> were reduced in both roots and shoots of cotton seedlings by NaCl treatment. It is well

known that high NaCl concentration in the nutrient solution leads to an increase in the rate of Na<sup>+</sup> uptake. Consequently, a Na<sup>+</sup>-K<sup>+</sup> antagonism reduced K<sup>+</sup> absorption. An antagonistic effect between K<sup>+</sup> and Na<sup>+</sup> has been reported by WALKER (1994). A significant negative correlation was found between K<sup>+</sup> and Na<sup>+</sup> concentrations of leaf (r = -0.75, p < 0.01), petiole (r: -0.84, p < 0.01), and shoots (r: -0.90, p < 0.01). Salt tolerance in glycophytes is associated with the ability of a plant to limit uptake and/or transport of saline ions (mainly Na<sup>+</sup> and Cl<sup>-</sup>) from the root zone to aerial parts (GREENWAY and MUNNS 1980). At high external salinity (100 mM) the accumulation of Na<sup>+</sup> in petioles and shoots may indicate the existence of an inhibition of Na<sup>+</sup> transport to leaf laminae. Our results are consistent with those reported by DOWNTON (1985), working with grapevine rootstocks ('Dogridge', '1613', 'Harmony' and 'Ramsey') under glasshouse conditions for three consecutive growing seasons. In contrast to Na<sup>+</sup>, Cl<sup>-</sup> concentration in leaf laminae increased with salinity. The work of SYKES (1992) has shown that the ability to restrict uptake and / or root to shoot transport of Cl<sup>-</sup> is inherited as either a polygenic or monogenic trait. In crosses and backcrosses involving *V. berlandieri* and *V. vinifera*, however, hybrids segregate as either Cl<sup>-</sup> excluders or accumulators suggesting the effect of a major dominant gene for Cl<sup>-</sup> exclusion for *V. berlandieri* (SYKES 1987). Results clearly indicate that Na<sup>+</sup> concentration was higher than Cl<sup>-</sup> concentration. WALKER *et al.* (2004) showed that R<sub>3</sub>, a rootstock, was a good Cl<sup>-</sup> excluder but a poor Na<sup>+</sup> excluder. Similar results were reported for Cleopatra mandarin by WALKER (1986). Also our results about higher accumulation of Na<sup>+</sup> was similar with results reported by GARCIA and CHARBAJI (1993) for 'Cabernet Sauvignon' grapes. This suggests that in perennial plants the ability to exclude Cl<sup>-</sup> and Na<sup>+</sup> ions is due to two different mechanisms (SYKES 1992). Furthermore, while advances have been made in understanding the mechanism for Cl<sup>-</sup> exclusion in grapevines (SCHACHTMAN and THOMAS 2003, TREGGAGLE *et al.* 2010), the precise mechanism remains to be established. Rooted leaves, on

which roots are induced from the proximal end of the leaf petiole, have proved to be a suitable model system for ion transport, especially for woody perennial plants like grapevine (SCHACHTMAN and THOMAS 2003). Initial comparative data between rooted leaves of 140 Ruggeri and K 51-40 indicate similar Cl<sup>-</sup> uptake rates into roots of the two genotypes over a 3-h period in 10 mM Cl<sup>-</sup>, but lower concentrations of Cl<sup>-</sup> were measured in xylem of 140 Ruggeri (TREGEAGLE *et al.* 2010). Among the different parts of the plants, root and petioles showed the highest Cl<sup>-</sup> content. In grapevine and in soybean, it has also been shown that Cl<sup>-</sup> transport to the shoot is controlled by the root (SAUER 1968, DOWNTON 1977, LAÜCHLI 1984). The higher root/shoot Cl<sup>-</sup> concentrations ratio indicates the Cl<sup>-</sup> retaining ability of the r-Sah and r-Rish roots. GREENWAY and MUNNS (1980), MUNNS (2002), and STOREY *et al.* (2003) have proposed that the sequestration of ions in roots, and the prevention of their transport to the shoot in the xylem, is a mechanism for salinity tolerance. FISARAKIS *et al.* (2001) found consistently higher accumulations of Cl<sup>-</sup> and Na<sup>+</sup> in roots as compared to the leaves of 'Sultana' vines and suggested that capability to store Na<sup>+</sup> in roots is a tolerance characteristic of rootstocks. Certain grapevine rootstocks have Cl<sup>-</sup> restriction and exclusion capacity that can reduce Cl<sup>-</sup> uptake and transport to the shoot (WALKER *et al.* 2004). In comparison to Das and r-Sul, r-Sah and r-Rish are able to accumulate lower Cl<sup>-</sup> in the shoot (Fig. 2). However, grapevine rootstocks that lead to high concentrations of Cl<sup>-</sup> in laminae, petioles, and grape juice of scions grafted to them, e.g. rootstock K 51-40 (TREGEAGLE *et al.* 2006, WALKER *et al.* 2010), can lead to significant leaf damage and impairment to fruit development and yield of scions under saline conditions (WALKER *et al.* 2010). Studies with mature grapevines on a range of rootstocks in a range of environments and salinities have shown that the difference between a strong (e.g. 140 Ruggeri) and a poor (e.g. K 51-40) Cl<sup>-</sup> excluder, is maintained across the range of environments and salinities (WALKER *et al.* 2010). Chloride exclusion has been defined by TEAKLE and TYERMAN (2010) as the ability of plants to restrict uptake of Cl<sup>-</sup> from the soil and subsequent transport in the xylem to the shoot. WALKER *et al.* (2004) showed that 1103 paulsen (salt tolerance) is the best Cl<sup>-</sup> excluder rootstock because it had lowest Cl<sup>-</sup> concentrations in petiole, lamina and grape juice of field grown vines when compared to the others. Higher Cl<sup>-</sup> concentrations in shoots of Das and r-Sul is the most likely reason for salt sensitivity in this varieties. Genetic differences between plant species and varieties in the restriction of Na<sup>+</sup> and Cl<sup>-</sup> uptake from the soil or reduction of ion transport to the xylem could be an influential factor in diminishing the accumulation of those ions in leaves (MUNNS 2002). Increasing salinity levels resulted in decreased NO<sub>3</sub><sup>-</sup>-N concentrations in all parts of the vine, is due probably to NO<sub>3</sub><sup>-</sup>-Cl<sup>-</sup> antagonism. The reduction in availability of N significantly decreases plant growth. There is also a possibility that growth reduction is not entirely due to Cl<sup>-</sup> toxicity, but may be partially due to Cl<sup>-</sup> induced NO<sub>3</sub><sup>-</sup> deficiency (TORRES and BINGHAM 1973). There are many studies that indicate antagonism and interaction between NO<sub>3</sub><sup>-</sup> and Cl<sup>-</sup> (GUNES *et al.* 1996). The presence of Cl<sup>-</sup> inhibits the absorption of

NO<sub>3</sub><sup>-</sup> (CEREZO *et al.* 1997, PEUKE and JESCHKE 1999), while increasing NO<sub>3</sub><sup>-</sup> fertilization reduces Cl<sup>-</sup> concentration in the leaves (DEANE-DRUMMOND 1986). The inhibition of NO<sub>3</sub><sup>-</sup> uptake by Cl<sup>-</sup> could be due to interactions between both ions at the sites for ion transport (CRAM 1983) and roots of different cultivars would differ in their ability to discriminate between different available ions and show a preference for one ion over another (BANULS *et al.* 1990). TYERMAN and FINDLY (1989) found that NO<sub>3</sub><sup>-</sup> and Cl<sup>-</sup> permeate the same anion channels in tonoplast membrane, and that NO<sub>3</sub><sup>-</sup> permeability is approximately twice that of Cl<sup>-</sup>. WALLACE and BERY (1982) showed that yield reduction of wheat was due to NO<sub>3</sub><sup>-</sup> deficiency induced by Cl<sup>-</sup> toxicity. Results presented in this study showed that high external Cl<sup>-</sup> concentration probably reduces NO<sub>3</sub><sup>-</sup> accumulation in shoot and root of the four grapevine genotypes. In grapevine, it has been shown that by increasing NaCl concentration in external medium the NO<sub>3</sub><sup>-</sup>-N concentrations in all parts of the tested grapevine cultivars were decreased after 60 d (FISARAKIS *et al.* 2004). Our results are consistent with previous studies and a significant negative correlation was found between NO<sub>3</sub><sup>-</sup>-N and Cl<sup>-</sup> in all plant parts (*r*: from -0.74 to -0.89, *p* < 0.01).

## Conclusion

The growth parameters and mineral composition of four grapevine cultivars were compared under greenhouse conditions using four salt treatments. Parameters such as shoot growth, leaf number and root/shoot ratio were significantly decreased by salinity. Das and r-Sul showed the highest growth reduction. Decreasing in Total chlorophyll content in 'Dastarchin' and 'Red Sultana' cultivars were higher than that of others. Proline increased in four grapevine cultivars however, 'Rishbaba' and 'Sahebi' had higher Proline contents. Na<sup>+</sup> and Cl<sup>-</sup> contents of the shoot and root increased while there was a reduction in K<sup>+</sup> and NO<sub>3</sub><sup>-</sup> contents under different levels of salinity. Das and r-Sul were much more sensitive to salinity than r-Rish and r-Sah due to more accumulated chloride in leaves. According to the results obtained in this study salinity tolerance in the grape genotypes can be summarized as: r-Rish > r-Sah > r-Sul > Das. Generally, the maximum level of Cl<sup>-</sup> accumulated in roots of the all cultivars at 100 mM NaCl was less than the half maximum of Cl<sup>-</sup> accumulated in the shoot of the cultivars. No significant difference was observed between 25 and 50 mM NaCl. It was concluded that although r-Rish and r-Sah showed a capacity to restrict Cl<sup>-</sup> transport to the shoot particularly at 50 mM NaCl, all cultivars seem to be sensitive when exposed to salinity.

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