Adv. Hort. Sci., 2017 31(2): 97-105 DOI: 10.13128/ahs-20488

Evaluation of adaptability potential of seven Iranian pomegranate cultivars in Southern Iran, Arsenjan region

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Key words: adaptability, macro and micronutrients, *Punica granatum*, tolerance.

Abstract: Present study was carried out to compare leaf mineral composition and some physiological parameters in seven Iranian pomegranate cultivars for evaluation of their adaptability differences in Arsenjan region/Fars province/southern Iran and selecting probable more abiotic tolerant cultivars in this region. Uniform and healthy rooted plants of seven commercial pomegranate cultivars were purchased from a commercial nursery and planted in a completely randomized block design in an orchard site in Arsenjan region. After full establishment the orchard samples (fresh leaves) were taken and transferred to lab for analysis. Cultivars included: Malas Yousefkhani Saveh, Naderi Badroud, Malas Daneh Ghermez Yazd, Rabab Neiriz Fars, Shirin Shahvar Fars, Shirin Poust Daneh Ghermez and Zard Anar Arsenjan. Significant differences were found among studied pomegranate cultivars for concentrations of leaf potassium, calcium magnesium, sodium and Iron concentrations. Also parameters such as leaf total chlorophyll and carotenoids content, total sugars, relative water content and electrolyte leakage, α-tocopherol and ascorbic acid concentration were significantly different in studied cultivars. 'Zard Anar Arsenjan', an indigenous cultivar of the region, and 'Rabab Neiriz Fars' were evaluated as probable more tolerant cultivars in comparison to other cultivars. This can be attributed to their more optimized leaf mineral composition and antioxidant statues.

1. Introduction

Pomegranate *(Punica granatum* L.) is a nutrient dense and one of the most popular fruits native to Iran. With a production of 700,000 tons/year, Iran is the world's leading producer (Sarkhosh *et al*., 2009). Historical evidence reveals that the primary origin of pomegranate is Iran and that it has been spread from this region to other areas (Levin, 1996). A large number of pomegranate varieties can be found in Iran, more than 760 original, wild and decorative cultivars (Mousavinejad *et al*., 2009). Considering lack of water resources and intensification of abiotic stresses such as drought and salinity, importance of pomegranate has increased in recent years, since this species is a tolerant fruit crop and thrives well under arid and semi-arid climatic conditions (La Rue, 1980). Previous investigations indicate varied levels of tolerance to abiotic stress conditions such as drought and salinity among different pomegranate cultivars (Tabatabaei and Sarkhosh, 2006; Okhovatian-Ardakani *et al*., 2010; Ibrahim, 2016) because abiotic stress tolerance is a complex parameter and depends on both genetical and physiological properties.

Differences in adaptability potentials to prevailing environmental conditions and abiotic stresses tolerance among plant species or cultivars including pomegranate can be attributed to their varied ability for nutrients uptake and subsequent different concentration of macro and micronutrients within plant organs and tissues (Jamali *et al*., 2015). Nutrients deficiencies or imbalances exert secondary, often unpredicted influences on the growth of plants by changes in growth pattern, chemical composition, and antioxidant defense capacity of plants and particularly decrease the resistance of plants to biotic and abiotic environmental stresses (Hajiboland, 2012). Macronutrients are mainly important structural components of plants and their optimum concentrations result in improved growth (Marschner, 1995). Activity of enzymes and/or production of some metabolites involving in the plants response to their surrounding environment or even modulations in the signal transduction pathways are under micronutri-

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Received for publication 29 March 2017
Accepted for publication 25 May 2017

ents influence (Hajiboland, 2012). Cultivars with a higher capacity for macro and micronutrients absorption possess a more efficient growth and development cycle, with a better adaptability to environmental conditions (Jamali *et al*., 2015).

Previous literatures on comparison of differences in leaf mineral composition and also physiological characteristics in various Iranian pomegranate cultivars are limited (in comparison to other important crops) and more investigations for evaluation of adaptability potential in different pomegranate cultivars seem necessary. So the goal of the present study was to compare leaf mineral composition and also some physiological and biochemical parameters in seven Iranian cultivars for a deeper understanding of their adaptability differences and selecting probable more tolerant cultivars in Arsenjan region.

2. Materials and Methods

Orchard and plants

Uniform and healthy rooted plants of seven commercial Iranian pomegranate cultivars were purchased from a commercial nursery and planted in a completely randomized block design with 3 replications (each replication had 3 plants) with 3 m distance in rows and 5 m distance between rows in an orchard site in Arsenjan region (hub of pomegranate growing and production in Fars), Fars province, Iran. Average annual climate parameters in the experimental region were; precipitation: 200 mm, relative humidity (Max: 55%, Min: 23%), temperature (Max: 38, Min: 4°C). The soil of the orchard was sampled at two depths and analyzed for soil texture, mineral content, organic matter, pH and EC (Table 1). Cultivars included: Malas Yousefkhani Saveh (MYS), Naderi Badroud (NB), Malas Daneh Ghermez Yazd (MDGY), Rabab Neiriz Fars (RNF), Shirin Shahvar Fars (SSF), Shirin Poust Daneh Ghermez (SPDG) and Zard Anar Arsenjan (ZAA). After 4 years and full establishment of the collection orchard, samples (fresh leaves) were taken from the trees. Leaf samples were taken from different orientations of the trees (north, south, west and east); 25 fully expanded mature leaves from each side of all trees (100 leaves per tree as bulk samples), and transported to laboratory. Leaves were taken from shoots without terminal fruit. The leaves were of spring bloom, the middle third of the branch, at a height of between 1-1.5 m including the petiole. Leaves with abnormal symptoms such as chlorosis and mechanical lesions caused by pests or diseases were avoided. The trees were grown under drip irrigation; water quality parameters are presented in Table 2. Routine cultural practices suitable for commercial fruit production were carried out during experimental period. The following parameters were measured in studied cultivars for two consecutive years and an average was reported.

Table 2 - Analysis of quality parameters of irrigation water

Parameters	value
EC (dS m^{-1})	0.91
pH	7.1
Na (%)	34.75
Cl (mg l^{-1})	45.71
SAR	3.5
TDS (mg l^{-1})	590

Measurements

Trunk circumference. Circumference of trunks was measured 5 centimeter above ground and expressed as centimeter.

Leaf dry matter content. Three uniform leaves were selected and washed with tap and distilled water, after drying with clean towel they were weighed with digital scale and then were oven dried for 72 hours in 70°C and weighed. Leaf dry matters percentage was calculated by the following formula (Eshghi and Jamali, 2009):

Leaf dry matters (%) = [Leaf dry weight (g)/leaf fresh weight (g)] × 100

Leaf water content. Leaf relative water content (LRWC) was measured by using ten leaf discs. The leaf discs of each treatment were weighed (FW). They were then hydrated until saturation (constant

Table 1 - Analysis of soil samples in the experimental region

Soil depth (cm)	Soil texture	Soil mineral content (mg kg^{-1})							Organic carbon (%)	EC (dS m^{-1})	pH
		Nitrate	Ca	Mg	K	Fe	Zn	Mn			
0-30	Loamy clay	32	1200	150	150	8.4	1	8.7	0.75	0.82	7.6
31-60	Loamy sand	40	1150	150	140	7.32	0.95	7.1	0.65	0.71	7.6

weight) for 48 h at 5°C in darkness (TW). Leaf discs were dried in an oven (DW). Relative water content was calculated according to the following expression (Jamali and Eshghi, 2015):

$$LRWC\% = (FW-DW)/(TW-DW) \times 100$$

Leaf electrolyte leakage. Leaf electrolyte leakage (EL) was determined by recording the electrical conductivity (EC) of leaf leachates in double distilled water at 40 and 100°C. Leaf samples were cut into discs of uniform size and taken in test tubes containing 10 ml of double distilled water. The test tubes were kept at 40°C for 30 min and at 100°C in boiling water bath for 15 min and their respective electric conductivities (EC1 and EC2) were measured by conductivity-meter (METROHM Conductometer 644, Switzerland) (Jamali *et al*., 2015):

$$\text{Electrolyte leakage (\%)} = (EC1/EC2) \times 100$$

Leaf total sugars. The total leaf soluble carbohydrates were determined according to Irigoyen *et al.* (1992) and glucose (0-100 mg l^{-1}, from *MERCK*) was used as a standard. Leaf samples of 0.5 g (dry weight) were homogenized in 5 ml ethanol (95%) and centrifuged at 4500 × *g* for 15 min, the supernatant was removed from the sample and the residue was resuspended in 5 ml of 70% ethanol. Then the supernatant was centrifuged again for final extraction. Both supernatants were combined. Anthrone-sulfuric acid assay was used for determination. An aliquot of 100 µl was added to 3 ml of anthrone-sulfuric acid solution and the mixture was shaken, heated in a boiling water bath for 10 min and cooled at 4°C. The absorption at 625 nm was determined by spectrophotometer.

Leaf chlorophyll and carotenoids concentration. Leaf discs of 0.5 g were extracted in 5 ml of acetone (80%), then centrifuged for 10 min in 8,000 × *g*. The supernatant was used to make a final volume of 100 ml of the leaf extract. Extraction of leaf tissue with the buffer continued until decoloration. Absorbance of the extract was read at 470, 645 and 663 nm with a spectrophotometer and 80% acetone was used as a blank. Finally, Chlorophyll and carotenoids contents was calculated according to the following equations (Lichtenthaler, 1987):

$$\text{Chl a (mg. g}^{-1}\text{ fresh weight)}: [(12.25A663 - 2.79A645) \times v / 1000 \times W]$$

$$\text{Chl b (mg. g}^{-1}\text{ fresh weight)}: [(21.50A645 - 5.10A663) \times v / 1000 \times W]$$

$$\text{Chla + Chlb (mg. g}^{-1}\text{ fresh weight)}: [(7.15A663 + 18.71A645) \times v /1000 \times W]$$

$$\text{Carotenoids (mg. g}^{-1}\text{ fresh weight)}: 1000A470 - 1.82Chla - 85.02Chlb / 198$$

where Chla = chlorophyll a; Chlb = chlorophyll b; Chla+b = total chlorophyll; A = absorbance at ƛ (nm).

Leaf anthocyanins concentration. Leaf total anthocyanins were measured spectrophotometrically by pH differential method with two buffer systems: potassium chloride buffer, pH 1.0 (0.025 M) and sodium acetate buffer, pH 4.5 (0.4 M). 0.5 g leaf samples were extracted with 2 ml methanol: water: concentrated HCl solution (80:20:1 v/v/v). 0.4 ml of leaf extract was mixed with 3.6 ml of corresponding buffers and read against water as blank at 510 and 700 nm. Absorbance (A) was calculated as

$$A = (A515 - A700)\ \text{pH 1.0} - (A510 - A700)\ \text{pH 4.5}$$

Then total anthocyanins content was calculated using the equation:

$$\text{Anthocyanin (µg. g}^{-1}\text{ fresh weight)} = (A \times Mw \times DF \times 1000) / e$$

Where *A* is the absorbance of the diluted sample and DF is the dilution factor (10), Mw is molecular weight of cyanidin-3-glucoside (449.2) and e= 26,900 L/mol.cm, molar extinction coefficient of cyanidin-3-glucoside.

Leaf total polyphenols. Leaf polyphenols was determined with Folin-Ciocalteu reagent using gallic acid as a standard phenolic compound. In brief, 1 g of lyophilized leaf samples were placed in an eppendorf tube, with 1 ml of methanol (80%), grinded at 4°C and centrifuged at 10000 × *g* for 15 min. The extract was mixed with 0.5 ml of Folin-Ciocalteu reagent (diluted 1:1 with water) then 1 ml of a 5% sodium carbonate solution was also added. After 30 min, absorbance was measured at 725 nm and expressed as mg on g fresh $weight^{-1}$.

Leaf α-tocopherol concentration. Leaf α-tocopherol was extracted according to Chong *et al*. (2004).Two hundred mg lyophilized sample was homogenized in 1 ml acetone with a prechilled mortar and pestle at 4°C. Following the addition of 0.5 ml hexane, the homogenate was first vortexed for 30 s, then centrifuged at 1000 × *g* for 10 min. The upper hexane layer was removed while the acetone layer containing vitamin E remained in the vial. A second aliquot of 0.5 ml hexane was added, and the extraction process was repeated at least twice. α-tocopherol was estimated by the method of Kanno and Yamauchi (1997).

A 0.4-ml aliquot of 0.1% (w/v) 3-(2-pyridyl)-5,6-diphenyl-1,2,4-triazine was added to 0.2 ml of pooled extract. The volume was made up to 3 ml with absolute ethanol, 0.4 ml 0.1% (w/v) ferric chloride ($FeCl_3.6H_2O$) was added, and the content was gently mixed under dim light in a dark room to avoid photochemical reduction. After a 4 minutes reaction at room temperature, 0.2 ml 0.2 M orthophosphoric acid was added and the mixture left for another 30 min. Absorbance was determined at 554 nm spectrophotometrically and reported as µg. g fresh weight^{-1}. The blank was prepared in the same manner except that absolute ethanol was used instead of the sample.

α-tocopherol (Sigma Chemical) was used as a standard.

Leaf ascorbic acid concentration. Ascorbic acid was estimated by the method of Omaye *et al*. (1979). Briefly, to 1 g of lyophilized leaf sample, 10% ice-cold TCA was added and centrifuged for 20 min at 3500 × *g* in room temperature. One ml of the supernatant was mixed with 0.2 ml of DTC reagent and incubated for 3 h at 37°C. Then 1.5 ml of ice-cold 65% H_2SO_4 was added, mixed well and the solutions were allowed to stand at room temperature for an additional 30 min. The color developed was read at 520 nm spectrophotometrically and reported as µg. g fresh weight^{-1}.

Macro and micronutrients. Oven-dried leaf samples were used for determination of macro and micro-nutrients. Dried samples (0.5 g) were ground and ashed at 550°C in a porcelain crucible for 6 h. The white ash was mixed in 2 M hot HCl, filtered and finally made up to 50 mL with distilled water. Sodium (Na) and potassium (K) concentration of samples were determined using flame emission method using a Sherwood Scientific Ltd model 360 flame photometer. Atomic absorption spectrophotometer (AA 6200, double beam atomic absorption spectrophotometer, Shimadzu, Kyoto, Japan) was used to determine Ca, Mg and micronutrient element including Fe, Zn, Mn concentrations (Kalra, 1998). Nitrogen (N) concentration was measured using the Kjeldahl digestion method (Kalra, 1998). Phosphorus (P) concentration was determined colorimetrically (Kalra, 1998). Chlorine (Cl) was measured by precipitation titration with silver nitrate (Mohr's method) (Kalra, 1998).

Statistical analysis

Data were analyzed by SAS and means were compared using Duncan's multiple range test at 5% probability level.

3. Results

Leaf macronutrients and Na and Cl concentrations in studied pomegranate cultivars are indicated in Table 3. Leaf N, P and Cl concentrations were not statistically different in studied cultivars. RNF and ZAA had significantly higher leaf K concentration in comparison to SPDG and NB, other cultivars were not statistically different. The highest leaf Ca concentration was observed in RNF cultivar (2.53±0.06 mg g^{-1} dry weight), however MDGY, SSF and ZAA were not statistically different. This macronutrient was 22% lower in SPDG compared to RNF. Leaf Mg concentration was significantly higher in MYS, MDGY, RNF and ZAA in comparison to NB, SPDG and SSF. Leaf Na concentration was 41% higher in MYS in comparison to ZAA. Other cultivars were not statistically different.

Leaf Fe, Zn and Mn concentrations in studied pomegranate cultivars are presented in Table 4. Leaf Fe concentration is 32% higher in ZAA in comparison to NB. Other cultivars are not statistically different. No significant difference was found among studied cultivars for leaf Zn and Mn concentrations.

Leaf dry matters, total sugars, relative water content and electrolyte leakage are shown in Table 5.

Table 3 - Leaf macronutrient and Na and Cl concentrations in studied pomegranate cultivars

Cultivars	N (mg g^{-1} DW)	P (mg g^{-1} DW)	K (mg g^{-1} DW)	Ca (mg g^{-1} DW)	Mg (mg g^{-1} DW)	Na (mg g^{-1} DW)	Cl (mg g^{-1} DW)
Malas Yousefkhani Saveh	20.50±2.07 a	1.59±0.09 a	21.60±0.72 bc	1.99±0.12 cd	0.97±0.02 ab	3.53±0.47 a	1.56±0.23 a
Naderi Badroud	22.80±0.81 a	1.63±0.02 a	20.33±0.41 c	2.11±0.05 bcd	0.89±0.06 b	2.74±0.43 ab	1.48±0.28 a
Malas Daneh Ghermez Yazd	21.453±1.48 a	1.62±0.11 a	22.60±0.72 abc	2.38±0.09 abc	1.13±0.01 a	2.43±0.36 ab	1.89±0.13 a
Rabab Neiriz Fars	20.83±0.89 a	1.60±0.09 a	23.56±0.29 a	2.53±0.06 a	1.06±0.04 a	2.49±0.49 ab	1.96±0.10 a
Shirin Shahvar Fars	22.43±0.74 a	1.59±0.02 a	22.80±0.44 abc	2.21±0.08 abcd	0.88±0.06 b	2.76±0.40 ab	1.76±0.14 a
Shirin Poust Daneh Ghermez	22.50±1.41 a	1.53±0.09 a	20.91±1.21 c	1.97±0.02 d	0.86±0.07 b	2.28±0.35 ab	1.47±0.26 a
Zard Anar Arsenjan	21.78±0.87 a	1.65±0.06 a	23.55±0.27 ab	2.50±0.02 ab	1.13±0.3 a	2.07±0.07 b	1.44±0.30 a

Means followed by the same letters within columns are not different at 5% probability using Duncan's test. All data indicated are mean ± standard error (n = 3).

Table 4 - Leaf micronutrient concentrations in studied pomegranate cultivars

Cultivars	Fe (mg g^{-1} DW)	Zn (mg g^{-1} DW)	Mn (mg g^{-1} DW)
Malas Yousefkhani Saveh	58.66±2.02 ab	23.00±4.50 a	53.00±9.70 a
Naderi Badroud	43.00±5.85 b	25.00±3.46 a	49.00±3.51 a
Malas Daneh Ghermez Yazd	46.33±4.91 ab	29.33±2.84 a	49.33±5.81 a
Rabab Neiriz Fars	53.00±8.38 ab	22.33±4.33 a	55.00±9.23 a
Shirin Shahvar Fars	53.667±2.33 ab	22.33±3.66 a	50.66±3.38 a
Shirin Poust Daneh Ghermez	58.00±2.08 ab	23.00±3.05 a	48.66±0.88 a
Zard Anar Arsenjan	64.667±3.71 a	26.66±7.35 a	56.00±2.64 a

Means followed by the same letters within columns are not different at 5% probability using Duncan's test. All data indicated are mean ± standard error (n = 3).

RNF had significantly higher leaf dry matter compared to MYS, SSF and SPDG. ZAA, NB and MDGY were not statistically different. The highest leaf total sugars (47.63±4.99 mg g^{-1} dry weight) were detected in RNF which were significantly higher than SPDG, SSF, NB and MYS. ZAA showed the highest leaf relative water content (83.83±0.74%) however RNF, SSF and MDGY were not statistically different. This parameter was lower in MYS, NB and SPDG compared to ZAA. Leaf electrolyte leakage was significantly higher in MYS in comparison to all other cultivars.

Concentrations leaf pigments in studied pomegranate cultivars are indicated in Table 6. NB had significantly lower leaf total chlorophyll concentration in comparison to RNF, other cultivars were not statistically different. Leaf carotenoids concentration was significantly higher in ZAA, SPDG, SSF and RNF compared to NB and MYS. Leaf anthocyanins concentration was not different among studied cultivars. MYS showed lower chlorophyll a to b ratio in comparison to RNF, SSF, SPDG and ZAA.

Leaf total polyphenols concentration was statistically higher in RNF, SPDG and ZAA in comparison to MYS, NB and SSF (Fig. 1). The highest leaf α-toco-

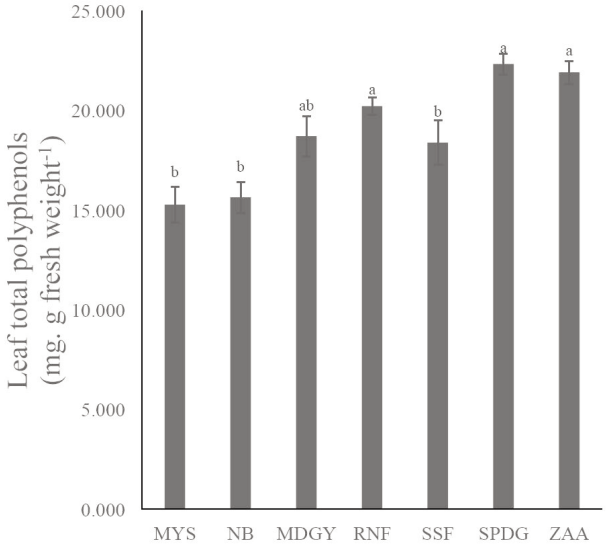

Fig. 1 - Leaf polyphenols concentration in studied cultivars. Columns with the same letters are not statistically different at 5% probability using Duncan's test. Vertical bars indicate standard errors (n=3).

Table 5 - Leaf dry matter, total sugars, relative water content and electrolyte leakage in studied pomegranate cultivars

Cultivars	Leaf dry matter (%)	Leaf total sugars (mg g^{-1} DW)	LRWC (%)	EL (%)
Malas Yousefkhani Saveh	24.33±1.20 d	22.44±2.26 d	79.05±0.67 c	24.80±1.77 a
Naderi Badroud	30.33±1.45 ab	29.50±1.45 cd	80.92±1.27 bc	20.08±1.28 b
Malas Daneh Ghermez Yazd	32.33±0.66 ab	39.63±2.56 ab	82.22±0.63 ab	19.20±1.51 b
Rabab Neiriz Fars	33.00±1.52 a	47.63±4.99 a	82.70±0.24 ab	18.66±0.50 b
Shirin Shahvar Fars	25.66±1.76 cd	35.72±3.29 bc	81.62±0.89 abc	18.55±1.24 b
Shirin Poust Daneh Ghermez	28.66±0.66 bc	29.66±1.15 cd	80.55±0.86 bc	17.20±0.49 b
Zard Anar Arsenjan	30.66±2.18 ab	42.41±1.22 ab	83.83±0.74 a	17.10±0.10 b

Means followed by the same letters within columns are not different at 5% probability using Duncan's test. All data indicated are mean ± standard error (n = 3).

Table 6 - Leaf pigments concentrations in studied pomegranate cultivars

Cultivars	Total chlorophyll	Carotenoids	Anthocyanins	Chlorophyll a/b ratio
Malas Yousefkhani Saveh	1.35±0.06 ab	0.15±0.01 c	0.25±0.004 a	0.88±0.04 c
Naderi Badroud	1.16±0.08 b	0.20±0.01 b	0.25±0.007 a	1.01±0.04 bc
Malas Daneh Ghermez Yazd	1.30±0.17 ab	0.25±0.01 ab	0.24±0.002 a	1.15±0.02 ab
Rabab Neiriz Fars	1.46±0.04 a	0.27±0.005 a	0.26±0.01 a	1.19±0.03 a
Shirin Shahvar Fars	1.44±0.04 ab	0.28±0.004 a	0.25±0.02 a	1.19±0.01 a
Shirin Poust Daneh Ghermez	1.40±0.07 ab	0.26±0.02 a	0.25±0.02 a	1.29±0.03 a
Zard Anar Arsenjan	1.34±0.10 ab	0.29±0.006 a	0.26±0.01 a	1.26±0.04 a

Means followed by the same letters within columns are not different at 5% probability using Duncan's test. All data indicated are mean ± standard error (n = 3).

pherol concentration was obtained from SPDG (229 µg. g fresh weight^{-1}), however it was not statistically different in comparison to ZAA, RNF and SSF. MYS, NB and MDGY had significantly lower leaf α-tocopherol concentration compared to SPDG (Fig. 2). Leaf ascorbic acid concentration in RNF was significantly higher in RNF in comparison to MDGY, other cultivars were not statistically different (Fig. 3). RNF had significantly higher trunk circumference in comparison to MYS. Other cultivars were not statistically different (Fig. 4).

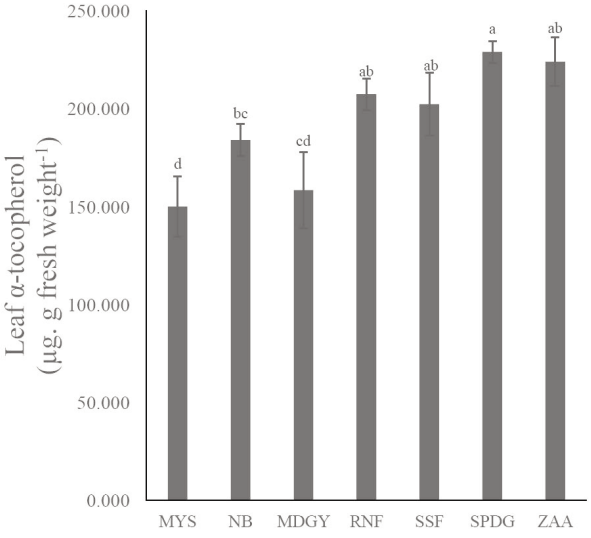

Fig. 2 - Leaf α-tocopherol concentration in studied cultivars. Columns with the same letters are not statistically different at 5% probability using Duncan's test. Vertical bars indicate standard errors (n=3).

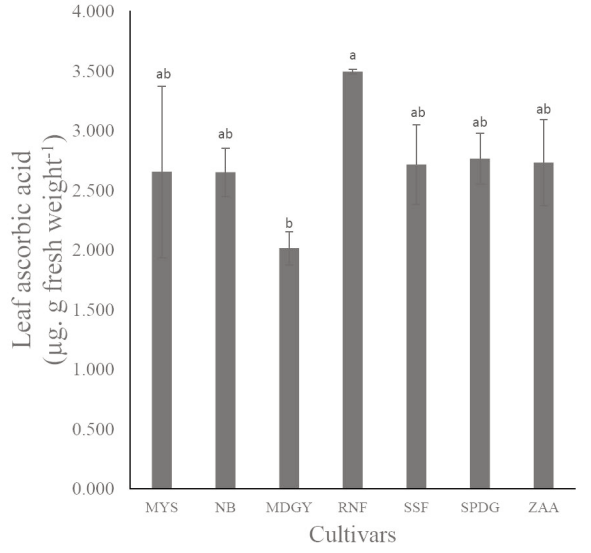

Fig. 3 - Leaf ascorbic acid concentration in studied cultivars. Columns with the same letters are not statistically different at 5% probability using Duncan's test. Vertical bars indicate standard errors (n=3).

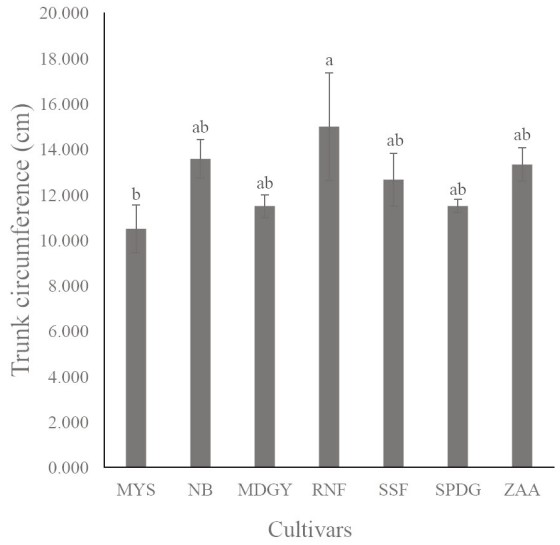

Fig. 4 - Trunk circumference in studied cultivars. Columns with the same letters are not statistically different at 5% probability using Duncan's test. Vertical bars indicate standard errors (n=3).

4. Discussion and Conclusions

In present study significant differences were found between studied pomegranate cultivars for concentrations of leaf macro and micronutrients. This was in agreement with previous studies. Giménez *et al*. (2000) compared leaf mineral composition (macro and micronutrients) in two pomegranate cultivars; MC1 (one of the most commonly grown cultivars in the south-east of Spain) and a well-known Israeli early variety. They found significant differences between these two pomegranate cultivars for concentrations of leaf macro and micronutrients. Normality ranges for N, P, K, Ca and Mg were also varied. El-Agamy *et al*. (2010), compared the salt tolerance of two important Egyptian pomegranate cultivars, Manfalouty and Nab El-Gamal *in vitro*. They found significant differences in these two cultivars under saline and non-saline conditions for absorption of N, K and Ca. Khayyat *et al*. (2014) evaluated physiological and biochemical responses of Malas Mommtaz and Shishe Kab under saline conditions; they reported with increase in salinity (from 4.61 to 7.46 dS m^{-1}) shoot Cl concentration augmented in Malas Mommtaz but decreased in Shishe Kab, also this cultivar (Shishe Kab) managed Na^+ transport into the leaves better than Malas Mommtaz. Sarafi *et al*. (2017) indicated that Wonderful and Ermioni cultivars had different ability for P, K, Ca, Mg and Zn uptake under optimum conditions. Similar differences were observed between Rabab and ShisheKab

by Hasanpour *et al*. (2015).

RNF and ZAA had higher relative water content and lower electrolyte leakage in comparison to MYS. Presence of higher K concentration in plant tissues will lead to more efficient water relationships in the leaves (Marschner, 1995). Photosynthesis, stomatal activity, transport of sugars, protein synthesis are all dependent on K (Prajapati and Modi, 2012). Ca pectates provide stability and mechanical strength to cell walls (Pilbeam and Morley, 2007; Taiz and Zeiger, 2010). Structural impairments in membrane structure are very common in Ca-deficient cells which make cell membranes very leaky and cause extensive loss of organic (*e.g*., sugars, amino acids) and inorganic electrolytes from root or leaf cells (White and Broadley, 2003). Leaf total sugars varied among studied cultivars. About half of osmotic potential in glycophte species is related to presence of sugars (Cram, 1976). Accumulation of higher total sugars has been reported to be associated with higher tolerance to salinity and/or drought in various species; grape, barley, *Zygophyllum album* and soybean (Ashraf and Harris, 2004). Five sunflower cultivars were evaluated for their salinity tolerance, more resistant lines had higher total sugars (Ashraf and Tufail, 1994). Comparison between wild populations of *Melilotus indica* (salt tolerant) and *Eruca sativa* (salt sensitive) showed that the former had higher leaf sugars (Ashraf and Harris, 2004).

Significant difference was observed between cultivars for leaf chlorophyll and carotenoids concentration in present study. In various studies the chlorophyll concentration were used as a sensitive indicator of the cellular metabolic state (Chutipaijit *et al*., 2011). Higher chlorophyll concentration is related to elevated tolerance against abiotic stresses such as drought and salinity (Hasanuzzaman *et al.*, 2013). In addition to harvesting solar energy, carotenoids play protection roles keeping integrity of photosynthesis apparatus against photo oxidative damages by scavenging free radicals (Dall'Osto *et al*., 2007; Andrade-Souza *et al*., 2011). Carotenoids are precursor of ABA which is an important phyto-hormone regulating plant responses in response to stresses. So presence of higher carotenoid concentration lead to lower photo-oxidative damage and higher potential for regulating plant growth under stress conditions (Götz *et al*., 2002; Han *et al*., 2008).

Non enzymatic antioxidants (leaf polyphenols, α-tocopherol and ascorbic acid concentration) were statistically different among studied cultivars. Varied antioxidant profile in different species and cultivars is one of the main reasons responsible for their different adaptability and abiotic tolerance potential (Munns and Tester, 2008; Jamali *et al*., 2016). Polyphenols have strong antioxidant properties and presence of an elevated level of them is associated with increased abiotic stress tolerance (Jamali *et al*., 2016). Among vitamin E family, α-tocopherol has the highest antioxidant activity (Garg and Manchanda, 2009). Several lines of evidence indicate that α-tocopherol plays a major role in keeping an adequate redox state in chloroplasts (Munne-Bosch, 2005). Deficiency of this antioxidant leads to a slightly increased susceptibility to photooxidative stress (Kanwischer *et al*., 2005). Plants have different capacity of ascorbate metabolism which is due to the variation of ascorbic acid synthesis and regeneration. Plant with higher amount of ascorbic acid content demonstrate better protection against oxidative stress. Ascorbate influences many enzyme activities, minimizing the oxidative damage through synergic function with other antioxidants (Foyer and Noctor, 2005 a, b). Ascorbic acid plays a role as a co-factor of violaxanthin de-epoxidase, thus sustaining dissipation of excess excitation energy (Pourcel *et al*., 2007).

Trunk circumference was significantly higher in RNF compared to MYS. Difference in mineral composition and ability of nutrients uptake in these cultivars could be one of the reasons responsible for this, as discussed above. Difference in enzymatic (data not shown) and non-enzymatic antioxidant responses and also growth regulators such as GA_3, zeatin and ABA (data not shown) in these cultivars are other reasons for varied growth rate observed between pomegranate cultivars in the present study.

Pomegranate is a tolerant species, however various varieties have significantly different adaptability potential. This can be contributed to varied ability for macro and micronutrients uptake, different enzymatic and non-enzymatic antioxidant profile and endogenous plant growth regulators. However, environmental conditions play an important role in this regard and might alter the final response of plant. ZAA is an indigenous variety of Arsenjan region/Fars province/Iran. High level of leaf K, Ca, Mg and Fe, enzymatic and non-enzymatic antioxidants (data not shown) and higher level of zeatin (data not shown) are the reasons responsible for better adaptation of this cultivar. Similar responses were observed in RNF. NB and MYS showed lower adaptability to the regional conditions among all studied cultivars. Further studies with same cultivars in other pomegranate growing regions seem necessary for a com-

prehensive evaluation about their adaptability capacity.

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