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Original Article

Effect of Magnetic Salinated Water on some Morphological and Biochemical Characteristics of Artichoke (*Cynara scolymus* L.) Leaves

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Abstract

Plants for optimal growth requires absorb water and nutrients absorption from the soil. Magnetic Water downward movement of minerals and makes it easy for plants to absorb nutrients and water. This study was designed to investigate the effect of magnetic salt water on some quantity and quality characteristics of artichoke leaves. The experiment was factorial based on completely randomized design with four levels of magnetic field [0, 3000, 6000 and 10000 gauss] and four levels of salinity [0, 3, 6 and 12 ds/m sodium chloride]. Results showed that the magnified water affected plant growth parameters. Fresh and dry weight of leaves and roots of plants increased as irrigated by magnified water. A significant interaction of salinity and magnetism was observed on mentioned parameters. Salinity and electromagnetic had significant effects on the most measured biochemical parameters. The highest amounts of phenolic and flavonoid contents were observed in the plants treated with magnified and saline water at medium level. The maximum antioxidant activity was observed in plant grown under 6 ds/m salinity. The highest and lowest amount s of chlorogenic acid was observed in plants irrigated with 6 ds/m saline water in which magnified fewer than 3000 gauss. The highest amount of caffeic acid [0/0044 mg/g] was recorded under 6 ds/m salinity and 3000 gauss electromagnetic conditions. Based on the results, it can be stated that the magnification increases efficiency of salty water and improve the performance and quality of the artichoke leaves.

Key words: Artichokes, Gauss, Electromagnetic field, Magnified water.

Introduction

Progressing in field soil salinity and available salty water is the most important agriculture limitation in the nearest future. The phenomenon of water treatment with an applied magnetic field has been known for many years and has been reported as being effective in numerous instances [1]. This type of physical treatment helps to prevent the use of chemicals such as polyphosphates or corrosive substances that are expensive and can be harmful to human life or disruptive to the environment [2]. According to Jones *et al* [3] they found that the electromagnetic fields amplify the plant growth regulator induced Phenylalanine Ammonia- Lyrase during cell differentiation in the suspended cultured plant cell. Magnetic fields have been reported to exert a positive effect on the germination of seeds [4,5], on plant growth and development [1-6].In principle, the water that is flowing in surface land or saving in subsurface, for the reason that passing from various levels, that have contain the cations and anions of Calcium and Magnesium, informing of carbonate, hydrogen carbonate or sulfate, is comprising the various salts, that the part of pertain to Ca2+ and Mg2+, is call water hardness [7]. Noran et al [8] observed the differences in the concentrations of K, N, P, Na and Ca + Mg in soils irrigated with magnetically treated water when compared those with normal water. They argued that magnetic treatment of water slows down the movement of minerals, probably due to the effect of acceleration of the crystallizations and precipitation processes of the solute minerals.

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It is known that plants and trees need mineral salts and microelements from the soil to function and photosynthesize properly [9]. Lin and Yotvat [10] reported an increase in water productivity in both crop and livestock production with magnetically treated water. Some studies have shown that there is an increase in number of flowers, earliness and total fruit yield of strawberry and tomatoes by the application of magnetic fields [11,12]. An increase in the nutrient uptake by magnetic treatment was also observed in tomatoes by Duarte Diaz et al [13]. Chang and Weng [14] investigated the effects of the magnetic fields (MF) on the hydrogen-bonded structure of water and found that the number of hydrogen bonds increased by approximately 0.34% when the MF strength increased from 1 to 10 T. It is found that some physical and chemical properties changed when water pass through magnetic field. Therefore the so called "magnetized water" has different chemical and physical properties and action than ordinary water [15]. Magnetic field application on water had stimulatory effect with respect to increase in seed germination because the hydrogen bond in liquid water is highly influenced by electrical and magnetic fields. Therefore magnetized water [water passed through magnetic field] bears different chemical and physical properties than ordinary water. The analysis made by Kleps [16] on soils irrigated with magnetically treated water showed higher values for mobile forms of nitrogen, phosphorus and potassium. Rokhinson et al [17] obtained a better dissolving and a deeper penetration of fertilizers in soil irrigated with magnetized water; this method is most effective in arid regions where water alkalinity is high and there is a tendency for soda salinization of soil. Artichoke is an herbaceous perennial plant (Cynara scolymus L.) belongs to the Asteraceae family cultivated in the Mediterranean area. In particular, the leaf extracts exhibits different effects: it lowers blood cholesterol, exerting a potent anticholestatic activity [18,19]. The action of phenolic compounds as antioxidant activity is mainly due to caffeoylquinic acid derivatives such as Chlorogenic acid and flavonoids such as luteolin glycosides effects [20,21].

The available studies and application of this technology in agriculture is very limited in Iran. Therefore, the present work was aimed to study the effect of irrigation with magnetized water on growth, yield and some chemical constitute of Artichoke in pot experiment.

Material and Methods

Present study was carried out as a pot experiment during 2012 at Gorgan University of Agricultural Sciences and Natural Resources, Gorgan, Iran. Four levels of magnetized water [zero [m1], 3000[m2], 6000[m₃] and 10000[m₄] Gauss] and four salinity levels [zero $[s_1]$, $3[s_2]$, $6[s_3]$ and $12[s_4]$ ds/m chloride sodium] with four replications were the experimental treatments. This experiment was carried out as a complete randomized blocks design (CRBD), in three replications. seeds were sown into round plastic pots [of 25 cm diameter and 35 cm height] filled with soil containing 54% sand, 25% silt and 21% clay [EC= 3.11 ds/m, PH= 7.9]. There were two plants per pot. The project involved greenhouse experiments and laboratory analysis of plant properties. Pots were irrigated with the tap water after magnetization through passing in magnetic device [HY5030E nutrient source DC, input 220 V, 3 Amper output tensity and teslameter device UT201 Company Hengtong]. At the harvest time parameters like, plant height (Measured ruler), number leaves, length and Width leaves (Measured ruler), leaf fresh and dry weight (gr), Root fresh and dry weight (gr) as well as Water use efficiently (WUE) of Artichoke plants was determined. Biochemical properties like Phenol, flavonoid and Antioxidant activity of leaves were determined using spectrophotometery. Chlorogenic and cafeic acids were determined with HPLC.

WUE =
$$\frac{\text{YEILD}}{\text{VW}}$$

VW= Volume of irrigation water (m^3) , YEILD= Kg per Hectare, WUE= Water use efficiently (Kg/m^3)

Extract preparation

The leaf samples were collected from the experimental plants. Well drained [at room temperature] samples were finely powdered and each sample [0.5 g] was extracted by percolation method using pure methanol [5 mL] for 24h to have a complete solvent removal extract.

Total phenolic and flavonoid contents

Total phenolic were assayed using the Folin–Ciocalteau reagent [22]. The extract of sample was added to 0.5 ml of distilled water and was mixed with 5 ml of the Folin–Ciocalteau reagent and aqueous Na_2CO_3 [4 ml, 1M]. The mixture was allowed to stand for 15 min and the phenols were determined spectrophotometrically at 760 nm. Total

phenolic content of plant parts was expressed as milligrams of gallic acid equivalents per gram of dry weight [mg GAE/g-1 DW] through the calibration curve with gallic acid. All samples were analyzed in three replications. Colorimetric aluminum chloride method was used for flavonoid determination [23]. Briefly, 0.5 ml of each plant extracts in methanol were separately mixed with 1.5 ml of methanol, 0.1 ml of 10% aluminum chloride. 0.1 ml of 1 M potassium acetate and 2 mL of distilled water and left at room temperature for 30 min; the absorbance of the reaction mixture was measured at 415 nm with Camspec M501 Single Beam Scanning UV/Vis Spectrophotometer. Total flavonoid content was calculated as quercetin from a calibration curve. The calibration curve was prepared by preparing quercetin solutions at concentration of 12.5 to 100 mg ml⁻¹ in methanol.

DPPH [diphenylpicrilhydrazyl] radical scavenging assay

DPPH was used for determination of free radicalscavenging activity of the extracts [24]. For that 1 ml of each extract was added to 1 ml DPPH. After 15 min at room temperature, absorbance was measured at 517 nm and during using methanol as blank. The antioxidant capacity was expressed as a percentage of inhibition of DPPH radical calculated according to the following equation: % inhibition of DPPH radical = $[[A_C-A_A/A_C] \times 100$, where A_C : absorbance of the control at time= 0 min; and A_A : absorbance of the antioxidant at time= 15min.

Analysis of Chlorogenic and Caffeic acid by analytical HPLC

The contents of Chlorogenic and Caffeic acid as to important substrate of Cynarin and Cynaroside were determinate using HPLC method. Standards were dissolved in methanol, the extract solutions was filtered through 0.45 μ M filter [Whatman type]. The HPLC analysis was carried out on a Merck Hitachi apparatus model Lachrom L-7100 connected to a computer analytical program HSM and an RP C18 column [250 x 4.6mm, 5 μ M] was set at 40^oC. Mobile phase included filtered H₃PO₄ [Phosphoric acid 0.5%], acetonitril and deionized water. The flow rate was kept at 1 mL per min. UV detector at 280 nm was used for Chlorogenic acid: [9 min] and Caffeic acid [14 min].

Statistical analysis

Statistical analyses were conducted using SAS program Version 9.2. The variance analyses

[ANOVA] was used to test the main effects of magnetic field and salinity and Saline ×Magnetic interaction effect. The Duncan's test was done to find the significant differences between each magnetic treatment and control at level 5%.

Results

Data presented in Table [1] show that irrigation using electromagnetized salty water significantly influenced on growth parameters of artichoke. Plant height, number of leaves, length and width of leaves, fresh and dry shoot weight, and the water consumption efficiency were relatively under magnetic water higher than compare with control plants. The improvement magnetized water treatment of 6000 Gauss with 3 ds/m salinity intensity reached to a higher value of plant height, number of leaves, length of leaves, width of leaves, fresh and dry shoot weight, dry root weight and water use efficiently, than other treatments compare with control treatment. The results revealed that fresh root weight was increased in treatment 10000 Gauss for all saline treatments compare with control. Among magnetized treatments the lowest effect was founded under 3000Gauss. A decreasing in all measured parameters was observed as salinity increased up to 3ds/m. According to the obtained results in this research it can be concluded that magnification of salty water reduce the damages of salinity and improve the water usage efficiency.

Magnetic water is considered one of several physical factors affects plant growth and its development. Results obtained in Table [2] showed that artichoke plants which irrigated with magnetic water grew taller than those irrigated with tap water. Based on the conclusion of Atak et al [25], increase all photosynthetic pigments through the increase in cytokinin synthesis which induced by MF could be the reason of the finding above. It has been showed that cytokine plays an important role on chloroplast development, shoot formation, axillary bud growth, and induction of number of genes involved in chloroplast development. Thus the stimulatory effect of the application of magnetic water on the growth parameters reported in this study may be attributed to the increase in photosynthetic pigments, endogenous promoters. Fomicheva et al [26,27] and Belyavskaya [28] reported that magnetic water significantly induces cell metabolism and mitosis meristematic cells of pea, lentil and flax. Magnetic treatment of water

may affect phyto-hormone production leading to improved cell activity and plant growth [29]. Above statements further suggest that the magnetic treatment of water probably alters something in water, makes the water more functional within plant system and therefore probably influences the plant growth at cell level. Magnetic treatment of water may affect phyto-hormone production leading to improved cell activity and plant growth [29]. Artichoke yield was increased significantly under magnetic irrigation. These results are logical to improvement growth parameters [Table 2]. The remarkable improvement induced by the magnetic treatment was consistent with the results of other studies on other crops like cereal, sunflower, flax, pea, wheat, pepper, tomato, soybean, potato and sugar beet [30-40]. In these studies the crop yield were increased.

 Table 1 Analysis of variance growth characteristic under magnetic water treatment

Treatment	df	Height	number leaves	length leaves	Width leaves	Fresh weight shoot	Dry weight shoot	Water use efficiently	root fresh weight	root dry weight
saline	3	322**	8.85^{**}	316.75**	33.33**	97.12**	1.5^{**}	2.29**	64.1**	9.26**
Magnetic Water	3	$88.88^{\ast\ast}$	3.1**	68.65^{**}	29.35**	64.32**	0.37^{**}	0.56^{**}	37.67**	1.09 ^{ns}
Saline ×Magnetic	9	0.73**	2.78**	35.73**	8.61**	16.26 ^{ns}	0.12*	0.19*	30.04**	1.21 ^{ns}
Error	45	9.47	0.58	9.47	2.57	13.31	0.06	0.09	1.64	1.7
CV	-	9.47	16.03	10.74	22.58	32.67	25.63	25.63	12.97	47.57

ns, * and ** not significant, significant at 5 and 1%, respectively.

Salinity ds/m	Magne tic water (Gauss)	Height	number leaves	length leaves (cm)	Width Leaves (cm)	Fresh weight shoot (gr)	Dry weight shoot (gr)	Water use efficientl y	Fresh weight of roots (gr)	Dry weight of roots (gr)
control	control	33.16 b	4 dc	30.87 ab	9.77 b	12.17 c	1.11 bc	1.37 c	11.18 b	3.64 a
	3000	28.75 ef	4.25 d	27.09 bc	6 ef	8.35 de	1.11 bc	1.37 c	10.02 b	1.73 c
	6000	37.25 a	5.25 bc	35.82 a	11.99 a	15.11 ab	1.52 a	1.88 a	16.91 a	2.88 ab
	10000	31.50 cd	5.75 ab	29.72 b	7.49 d	11.65 cd	0.87 d	1.07 de	10.23 de	1.92 bc
3 ds/m	control	30.37 d	5.25 bc	27.16 bc	7.45 dc	11.75 cd	1.02 c	1.26 c	12.03 bc	2.41 b
	3000	29.12 de	5.75 ab	26.58 c	5.38 ef	12.05 c	1.07 bc	1.32 c	10.12 e	1.97 bc
	6000	36.33 ab	6.75 a	34.19 a	10.33 ab	19.59 a	1.36 ab	1.67 ab	9.59 f	2.8 ab
	10000	25.31 gh	4.25 d	26.66 bc	5.07 f	12.02 c	0.94 cd	1.16 d	12.02 bc	1.86 bc
6 ds/m	control	27.57 f	5.25 bc	27.22 bc	6.74 e	10.97 cd	1.33 ab	1.64 ab	7.051	0.92 cd
	3000	32.66 c	5.25 bc	26.88 bc	6.7 e	9 d	1.06 bc	1.31 c	5.93 mn	1.08 cd
	6000	31.5 d	4.5 d	27.37 bc	8.06 c	13.56 bc	1.20 b	1.48 b	8.41 j	1.36 cd
	10000	25.96 g	5.25 bc	24.38 d	6.49 e	10.78 cd	0.68 e	0.84 e	13.63 b	1.74 c
12 ds/m	control	19 i	2.5 f	16.18 f	4.46 fg	6.63 e	0.45 f	0.56 f	8.04 k	1.16 cd
	3000	24.5 gh	4.25 d	23.9 d	7.01 de	9.94 cd	0.54 ef	0.67 ef	6.41 mn	0.98 cd
	6000	22.5 hi	4.75 c	21.35 e	5.65 f	8.35 de	0.44 f	0.54 f	4.84 n	0.55 d
	10000	23 hi	3.5 ef	20.7 ef	4.06 g	6.74 e	0.55 ef	0.68 e	11.5 cd	1.56 cd

Table 2 Effect of magnetic salty water on growth characteristic.

Means with similar letter are not significant at the 5% probability level.

Table 3 Analysis of variance chemical characteristic under saline and magnetic water treatments

Treatment	Df	Phenol	Flavonoid	Antioxidant Activity	Chlorogenic acid	Cafeic acid
saline	3	0.013**	0.021*	3593.5**	0.001*	1.68 ^{ns}
Magnetic	3	0.114^{**}	0.065^*	511.31**	0.043**	2.499 ^{ns}
Saline ×Magnetic	9	0.067^{**}	0.192^{**}	351.99***	0.011***	2.355^{*}
Error	45	0.023	0.029	80.964	0.00047	9.356
CV		9.81	6.18	15.57	13.305	2.311

ns, * and ** not significant, significant at 5 and 1%, respectively.

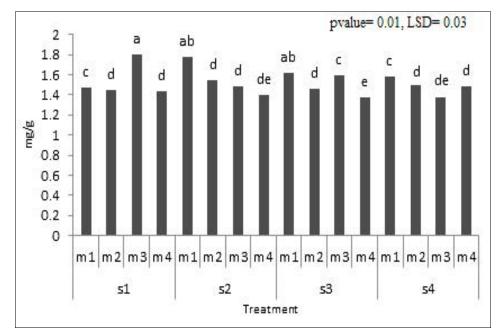


Fig. 1 Effect of magnetic saline water on phenol content of the artichoke

Means with similar letter are not significant at 5%. $[s_1, = 0 \ s_2, = 3 \ ds/m \ s_3, = 6 \ ds/m \ s_4 = 12 \ ds/m \ and \ m_1=0, \ m_2= 3000G, \ m_3= 6000G, \ m_4= 10000G]$

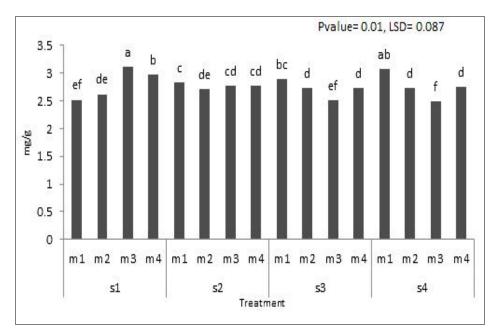


Fig. 2 Effect of magnetic salty water on flavonoid content of the artichoke

Means with similar letter are not significant at the 5% probability level.[$s_1=0$ $s_2=3$ ds/m $s_3=6$ ds/m $s_4=12$ ds/m and $m_1=0$, $m_2=3000$ G, $m_3=6000$ G, $m_4=10000$ G]

Magnetic treatment of water has been reported to change some of the physical and chemical properties of water, mainly hydrogen bonding, polarity, surface tension, conductivity, pH and solubility of salts [14,41,42]. The use of magnetically treated irrigation water increased available soil P in celery and snow pea [43]. The results obtained in this weight test allow us to conclude that magnetic treatment improves in weight plants.

Chemical constituents

In response to the irrigation with magnetized water, total phenols, flavonoids and antioxidant activity of

plants leaves at 120 days after sowing showed obvious changes than the control. Significant different at 1% of probability level, in phenols and Antioxidant activity and 5% of probability level, in flavonoid were recorded from irrigated plants with magnetized water and salty water as compared to irrigated plants with tap water. Although a significant difference of interaction at 1% of probability levels of treatments was observed all in interaction effect [Table 3].

Results of means comparison showed a decreased in phenols, flavonoids and antioxidant activity reached to increase of magnetic intensity [Fig. 1-2]. The amount of total phenolic compounds also was affected by the salty magnetic water so that the maximum of its rate (1.8 mg/g) was observed under the 6000G of magnetic field (without salinity effect) and the lowest of its amount (1.37 mg/g) was observed in combination of 6ds/m salinity+10000G magnetic field treatments. The lowest value of phenolic content was observed at 12ds/m salt treatment [Fig. 1].

Significant increases in phenol, flavonoid and antioxidant activity were recorded in artichoke plants irrigated with magnetic water compared to control treatment (Table 3). Results of means comparison show that the flavonoid content of leaf extract reached to 3.11 mg/g under the treatment of 6000G magnetized under normal water irrigation. The lowest flavonoid content (2.49mg/g) was observed when salinity increased to 12ds/m and salty water treated with 6000G magnetic intensity. The increase in the plant constituent with the magnetic treatment of water has been reported that depends to change of some of the physical and chemical properties of water, mainly hydrogen bonding, polarity, surface tension, conductivity, pH and solubility of salts [14,41,42]. It has been showed that the use of magnetically treated irrigation water increased available soil P in celery and snow pea [43]. Magnetized water increased the mobility of phosphate of soil [44].

Thus the observed changes can be concluded by the above description. The maximum of antioxidant activity (76.86 %) was observed in plants in which

treated with salty water of 6ds/m with and without magnetic treatment [Fig. 3].

Our results also showed a significant effect of magnetic water treatment on the total phenols and total flavonoid content. A potential link between MF [magnetic field] and its effects on living organisms is the fact that MF causes an oxidative stress, that is, MF can alter energy levels and spin orientation of electrons to increase the activity, concentration, and lifetime of free radicals [45-46]. Probably one of cases that can antioxidant activity increased effects is of MF on water living organisms. Increased levels of Ca was decreased the concentrations of total phenol. Although Ca led to increasing of peroxidase, polyphenoloxidase and phenylalanine amoniliase that maybe counted as phenol synthesis [47]. The decrease in the chemical substances with the magnetic treatment of salt water has been reported to change some of the physical and chemical properties of water, mainly hydrogen bonding, polarity, surface tension, conductivity, pH and solubility of salts [14,41,42].

Chlorogenic and cafeic acids

Interaction effects of magnetic and salt water were significantly different at 1% of probability level, on Chlorogenic and Cafeic acids in leaves of plants which were recorded from saline irrigated plants as compared to irrigated plants with tap water (Table 3). Fig. 4 showed that, compare to tap water irrigation, when the plants irrigated with magnetic water a decrease was observed in chlorogenic acid content. By increasing the salinity up to 6ds (0.314 mg/g) (without magnetic affect) chlorogenic acid was increased. On the other hand the chlorogenic acid of plants treated with 12 ds showed the lowest amount. Under medium salinity the content of chlorogenic acid of the plants which were irrigated with magnetized water of 10000 G strongly reduced. As presented in Fig. 4 under salinity of S2 and S3 a strong significant reduction was appeared by magnetic field density increasing. The maximum amount of chlorogenic acid (0.314 mg/g) was observed at the salinity of 6ds/m (Without magnetic treatment) [Fig. 4].

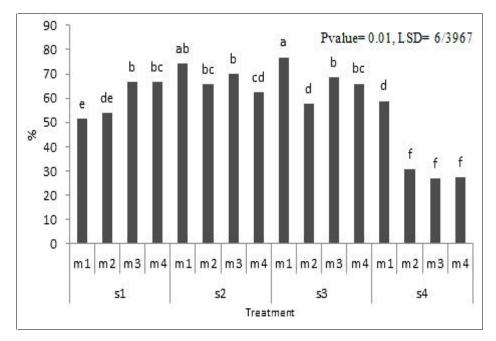


Fig. 3 Effect of magnetic salty water on antioxidant activity content of the Artichoke Means with similar letter are not significant at the 5% probability level.[s_1 = 0 s_2 = 3 ds/m s_3 = 6 ds/m s_4 = 12 ds/m and m_1 =0, m_2 = 3000G, m_3 = 6000G, m_4 = 10000G]

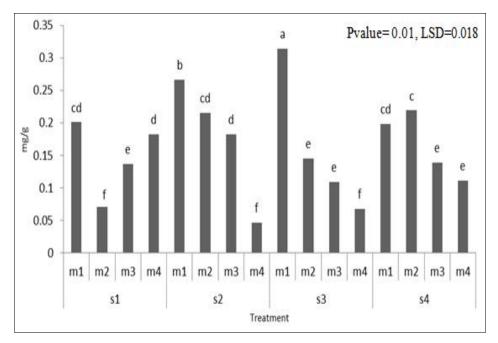


Fig. 4 Effects of magnetic salty water on the chlorogenic acid of the Artichoke

Means with similar letter are not significant at the 5% probability level. In all figures please add the mean of abbreviations used. [s_1 , = 0 s_2 , = 3 ds/m s_3 , = 6 ds/m s_4 = 12 ds/m and m_1 =0, m_2 = 3000G, m_3 = 6000G, m_4 = 10000G]

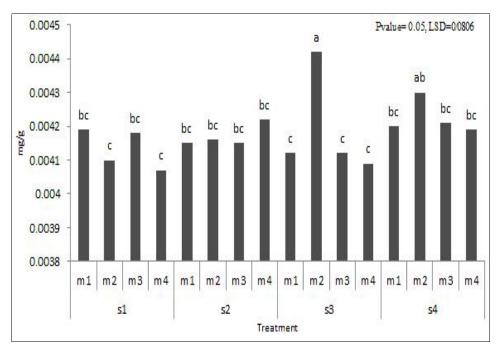


Fig. 5 Effects of magnetic salty water on the cafeic acid of the Artichoke Means with similar letter are not significant at the 5% probability level.[s_1 , = 0 s_2 , = 3 ds/m s_3 , = 6 ds/m s_4 = 12 ds/m and m_1 =0, m_2 = 3000G, m_3 = 6000G, m_4 = 10000G]

The results confirmed that 3000 gauss and 6 ds salinity was the best treatments increasing cafeic acid. As is presented in Fig. 5, the maximum cafeic acid amount (0.00442 mg/g) was observed at the salinity of 6ds/m+3000G magnetic field. Opposite to that the lowest of caffeic acid (0.00407 mg/g)accumulated in the plant samples which were irrigated with tap water in which magnetized under 10000G magnetic fields [Fig. 5]. It has been showed that polyphenolic components production strongly enhanced in response to biotic and nonbiotic stresses [48]. In fact polyphenolic components such as cafeic acid had defensive role against free radicals that produce during metabolism [49]. The reasons of increase in polyphenols in response to salinity and 3000 Gauss could be due to the changes in chemical properties of salty water. Salt stress restricts plant growth more than photosynthesis; as a result, plant diverts the synthesis of carbohydrates to produce secondary metabolites [50]. In contrast, at high salt concentration, uptake of phosphor and potassium that are principal substance of secondary metabolites such as polyphenols, decline [51]. Also, due to disturbance of enzymatic activities in high salinity, photosynthesis is declined; therefore, growth and production of polyphenols will be decreased [52].

The importance of natural acids in plants to tolerate H_2O_2 and other free oxygen radicles was proved in

Ramonda serbica [53]. Maheshwari and Grewal [43] showed that by reducing pH of magnetic water, Ca and P concentrations increase in shootsof Celery and pea. Also, it has been showed that, irrigation with magnetic water led to limitation in leading of Na and lowering its toxic effect. Probably one of the reasons why chlorogenic acid reduces under high salinity and magnetic density could refer to reduction in stress level. Reduced Na concentration in snow pea pods irrigated with magnetically treated salty water [1000 ppm NaCl] suggest restricted Na loading into snow pea pods [43]. It has been showed that compared with the different normal water, when the soil was leached with different magnetized irrigation water, at all soil depths its salinity was significantly decreased [54]. Also it has been showed that magnetic water improves the uptake of NPK [16-55]. Also it should be mentioned that the response of soil to magnetic water varied from plant to plant.

Conclusion

In summary, growth parameters, some biochemical components and yield components of tested plants were concomitantly increased when plants were treated by magnetic water. Magnetized water had significant affects on crop production and plant length increase noticeably. Treating water with static magnetic field increases the solubility of salts. Plants irrigated with magnetized water acquire more nutrients from soil. It seems that increasing of soil salts enhances the photosynthesize property of plants. Furthermore, environmental stresses could significantly enhance the chemical constituents. As a result, salt stressed plants may be interesting potential sources of polyphenols for economical uses. On the other hand, according to the limitation high quality water especially in harsh regions, pretreatments like this, improve the water usable efficiency in saline fields.

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