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Studying the effects of mycorrhiza, root juice and citric acid on *Mentha piperita* yield and root generating

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Abstract

Organic cultivate of medical plants guarantee the quality of them and reduced the probability negative effects on medical quality and yield. In order to evaluate the effects of mycorrhiza inoculation, root juice and citric acid on rooting of the medicinal plant *Mentha piperita*, this experiment was conducted in 2012 in greenhouse condition at the research field of Islamic Azad University, Karaj branch, Iran. The experiment was conducted in factorial in the form of a completely randomized design with four replications and eight treatments: *Mycorrhiza* (M_0 = 0, M_1 = 100 kg/ha), root juice (R_0 = 0, R_1 = 5 cc) and citric acid (C_0 = 0, C_1 = 5 mM). Analysis of the variances indicated that application of root juice significantly affected plant fresh weight ($P \leq 0.05$) and increased it by 41.3% compared with the control. Applying 5 mM citric acid increased ($P \leq 0.05$) root volume by 48.01% and the number of root nodules by 26.1% compared with the control. The interaction of citric acid × root juice had also a significant effect on the number of rooted nodules ($P \leq 0.05$); the highest number of root nodules was achieved in C_1R_0. The most effective treatments in this study was known citric acid.

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Introduction

Peppermint (*Mentha piperita* L.) is a hybrid plant obtained by crossing spearmint (*Mentha spicata* L.) with water mint (*Mentha aquatica* L.). Mint is an important crop which Fresh and dry leaves are used for herbal teas. Peppermint raw material is used in medicine, cosmetics and food industry, therefore this plant is widely grown around the world. So Peppermint (Mentha piperita) is one of the most popular and widely consumed aromatic and medicinal plants by people (Scora and Chang 1997; Kukreja *et al.*, 2000; Lorenzi and Matos 2002; Dambrauskienė *et al.*, 2008). Scora and Chang (1997) stated that mint is a plant that can become easily adapted to a variety of climate and soil conditions. The nutrients can be supplied through hydroponic nutrient solutions enabling rapid growth and biomass production (Garlet and Santos, 2008).

Organic cultivate of medical plants guarantee the quality of them and reduced the probability negative effects on medical quality and yield (Griffe *et al.*, 2003). Arbuscular Mycorrhizal fungi can alter the profile of VOCs emitted by roots as well as the root morphology of sorghum plants, indicating that AM fungi have the potential to help plants adapt to and alter soil environments and can be integrated in soil management to achieve low-cost sustainable agricultural systems AMF can also benefit plants by stimulating the production of growth regulating substances, increasing photosynthesis, improving osmotic adjustment under drought and salinity stresses and increasing resistance to pests and soil borne diseases (Al-Karaki, 2006; Sun and Tang, 2013; Hooker and Black, 1995). Mycorrhizal fungi occur in most of the soils and colonize roots of many plant species. The symbiotic root-fungal association increases the uptake of less mobile nutrients (Ortas *et al.*, 2001), essentially phosphorus (P) but also of micronutrients like zinc (Zn) and copper (Cu), the symbiosis has also been reported as influencing water uptake. Root juice is fully herbaceous that made for organic farming. Which compound two natural products of humic acid and alga that containing of useful nutrient of macro elements (N, P, K ...) and micro elements (Zn, Fe, Cu) that stimulate of root developed. Organic acids are source of both carbon skeleton and energy for cells and are used in the respiratory cycle and other biochemical pathways (Dasilva, 2003). Citrate and malate are among the intermediate organic acids in Krebs cycle which produces cellular energy by oxidative phosphorylation (Wills *et al.*, 1981). Soil humidity, enough light and suitable nutrients is essential for Mentha piperita cultivate. Leafs is most important of photosynthesis organs. Inclination produce the medicinal and smelling plant for natural product particularly in organic conditions are increased in the word (Carrubba *et al.*, 2002). Organic fertilizers have beneficial effects on soil structure and nutrient availability, help maintain yield and quality, and are less costly than synthetic fertilizers (Olfati *et al.*, 2012; Thy and Buntha, 2005). Using nutrients caused increasing the leaf surface and photosynthesis (Omidbeygi, 1992). So in this research, we investigate possible effects and comparison of these compounds on *Mentha piperita* yield and root generating in hydroponic conditions.

Materials and methods

The research was carried out in a Greenhouse in the research farm of agricultural university of Azad Islamic of Karaj Branch in Iran at 2012. The plants used were obtained from *Mentha spicata* L., which were placed in 128 plastic glasses that filled with perlit and Super Absorbent Hydrogels. A randomized complete block experimental design with four replicates and eight treatments were done. Two levels of Mycorrhiza (M_0 = 0, M_1 = 100 $kgha^{-1}$), two levels of root juice (R_0 = 0, R_1 = 5 cc) and two levels of citric acid (C_0 = 0, C_1 = 5 mM) used. Plants were irrigated with 5 cc citric acid and 5 cc root juice added to the one liter of nutrient solution that 10 cc given to per samples in tree time stage with every 15 days and kept under protected conditions for 45 days. All Plants cut from the soil surface and leaf, stem and root weight (Scale with 0.01 accuracy), root length (with ruler), number of root knot (counting) and root bulk (by water) were determined. All statistical evaluations were done with use of SPSS software. Analysis of

variance results were considered significant if P < 0.05.

Results and discussion

In this study variance results showed that use 5 mM citric acid increased root bulk and number of root knot of *Mentha piperita* that significant at p<0.05 respectively. Mean comparison showed citric acid increased root bulk and umber of root knot from 0.908 to 1.344 ml (48.01%) and from 6.687 to 8.437 (26.1%) figures 1 and 2. Although there is a report on increase in root yield content of Mentha, in our application dose (5 mM) citric acid We suggest part of this effect being due to increase of proton pumping rate in roots being supported or induced by organic acids supplied. Our results indicate that citric acid had exerted a profound effect on carbohydrate partitioning in basil toward secondary metabolism these findings were similar to Darandeh and Hadavi, 2012 results. This is in accordance with previous reports on effect of citric acid sprays in extension of postharvest longevity of tuberose and lilium flowers and increase of bulbil weight in lilium which have been proposed to be related to effect of citric acid on carbohydrate partitioning (Eidyan 2010; Darandeh 2010). Inductive effect of citrate on production of citrate by microorganisms is previously reported (Cantino and Goldstein, 1967), which could be the case in plants, as well. We could assume that this induction had occurred and then is transferred to roots, increasing their proton and organic acid efflux. As a result the overall ion absorption capability of plant has improved, especially for P and N which is mentioned earlier reports (Arcand and Schneider 2006; Bais *et al*., 2006; Marschener 1998), and finally had leaded to increased performance and yield in our study. There are no significant differences between different Mycorrhiza for number of stem, number of leaf, Fresh stem weight, fresh leaf and root weight per plant while the lowest values were related to plants cultivated without fertilization (Tables 1). Generally mean comparison showed used 5cc root juice increased fresh root weight from 0.940 gr To 1.329 gr (41.3%) that significant (figure 3). The mean comparison among factor-levels revealed that the highest root knot was observed in $C_1 + R_0$ (50%) figure 4. The highest fresh leaf weight and root bulk were observed in $M_0 + R_1$ treatments (figures 5 and 6). The interactive between treatments showed that fresh stem weight, number of stem, number of leafs and number of root knot were observed in $C_1 + M_0 + R_0$ that significant at p<0.05 (figures 7, 8, 9 and 10).

Table 1. Variance analysis of measurement factors of *Mentha piperita*.

Sum of squares *(Mentha piperita)*									
Source	df	Fresh stem weight	Fresh leaf weight	Number of stem	Number of leaf	Fresh root weight	Bulk of root	Root lenght	Number of root knots
Citric acid	1	0.026^{ns}	0.001^{ns}	1.613^{ns}	8.000^{ns}	0.407^{ns}	1.711*	0.801^{ns}	32.000*
Mycorrhiza	1	0.003^{ns}	0.002^{ns}	0.853^{ns}	703.125^{ns}	0.007^{ns}	0.281^{ns}	12.444^{ns}	2.000^{ns}
Root juice	1	0.003^{ns}	0.024^{ns}	0.013^{ns}	544.500^{ns}	1.441*	0.080^{ns}	1.141^{ns}	0.500^{ns}
Citric acid * Mycorrhiza	1	0.020^{ns}	0.022^{ns}	4.813^{ns}	800.000^{ns}	0.005^{ns}	0.245^{ns}	0.701^{ns}	1.125^{ns}
Citric acid * Root juice	1	0.005^{ns}	0.001^{ns}	0.213^{ns}	741.125^{ns}	0.109^{ns}	0.001^{ns}	1.628^{ns}	21.125*
Mycohrriza * Root juice	1	0.002^{ns}	0.224*	1.613^{ns}	12.500^{ns}	0.188^{ns}	0.781*	6.021^{ns}	3.125^{ns}
Citric acid * Mycorrhiza* Root juice	1	0.067*	0.092^{ns}	36.053*	3828.125*	0.107^{ns}	0.500^{ns}	1.658^{ns}	12.500*
Error	23	0.346	0.659	128.667	14918.500	5.449	4.595	173.479	77.500

*, ^{ns} show the (p<0.05 and no significant).

We conclude we could confirm that those treatments application cause growth and development of *Mentha piperita* root so cause the better absorption of nutrition from soil and increase the yield of *Mentha piperita* (Figure 11). Because of a few study about these tree biofertilizer on medical plants of *Mentha piperita* we could hope about the effect of these treatments on medicine plants.

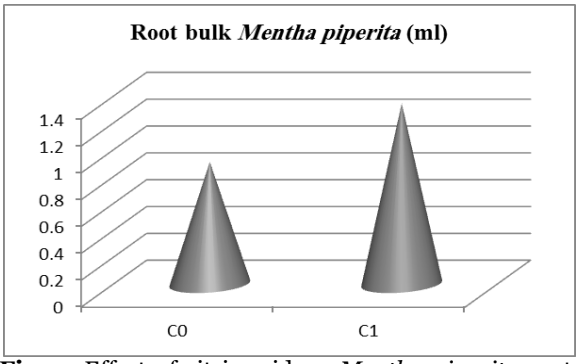

Fig. 1. Effect of citric acid on *Mentha piperita* root bulk.

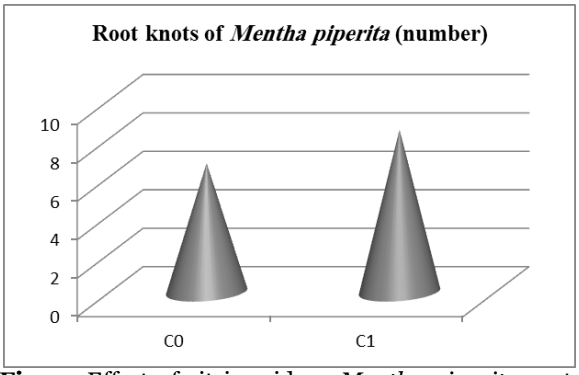

Fig. 2. Effect of citric acid on *Mentha piperita* root knots.

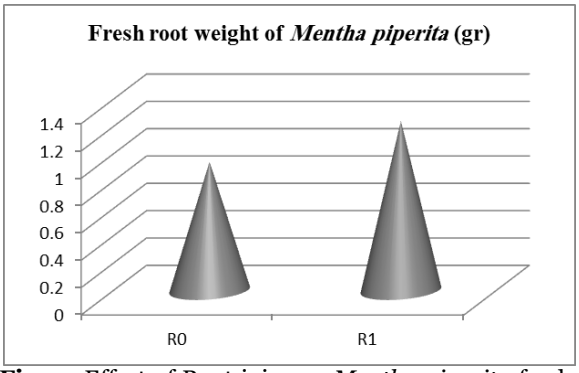

Fig. 3. Effect of Root juice on *Mentha piperita* fresh root weight.

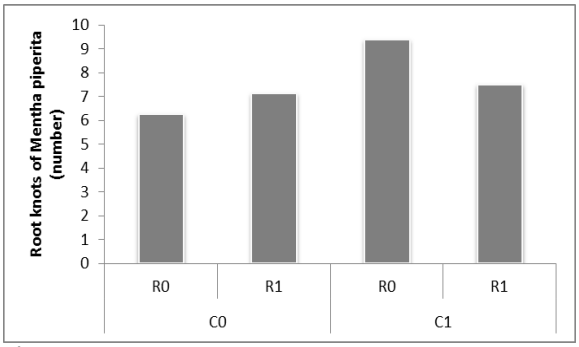

Fig. 4. Interactive of citric acid & Root juice on *Mentha piperita*.

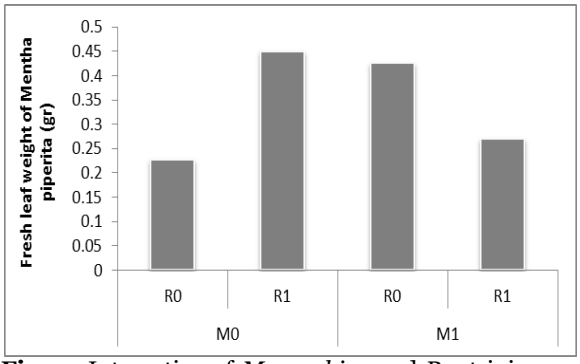

Fig. 5. Interactive of *Mycorrhiza* and Root juice on *Mentha piperita*.

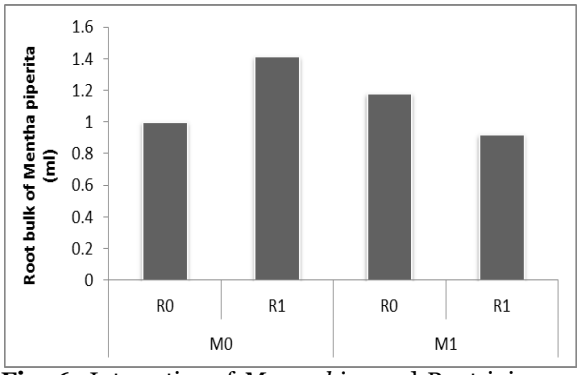

Fig. 6. Interactive of *Mycorrhiza* and Root juice on *Mentha piperita*.

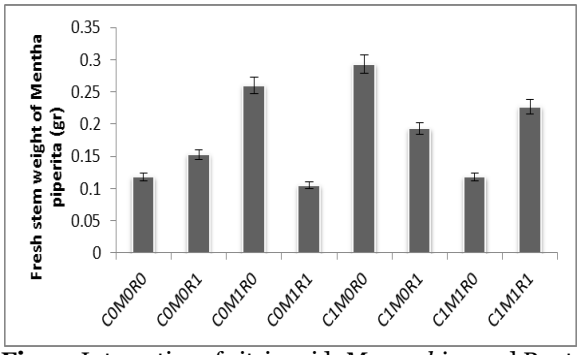

Fig. 7. Interactive of citric acid, *Mycorrhiza* and Root juice on *Mentha piperita*

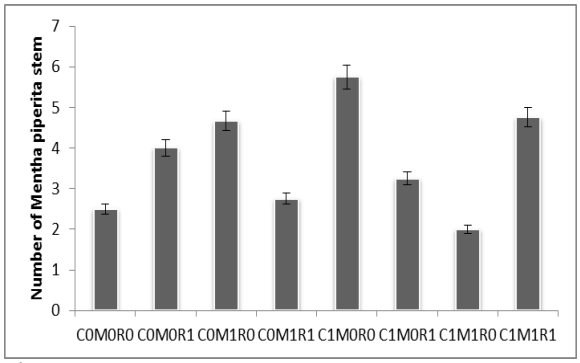

Fig. 8. Interactive of citric acid, *Mycorrhiza* and Root juice on *Mentha piperita*.

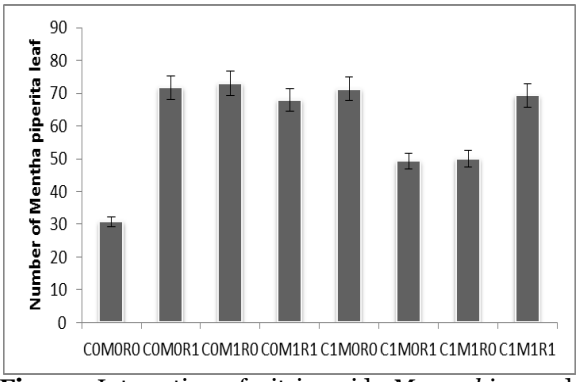

Fig. 9. Interactive of citric acid, *Mycorrhiza* and Root juice on *Mentha piperita*.

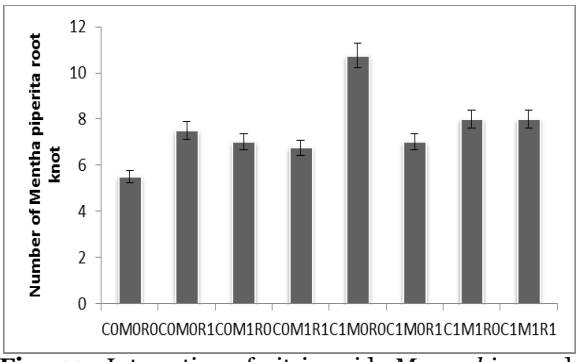

Fig. 10. Interactive of citric acid, *Mycorrhiza* and Root juice on *Mentha piperita*.

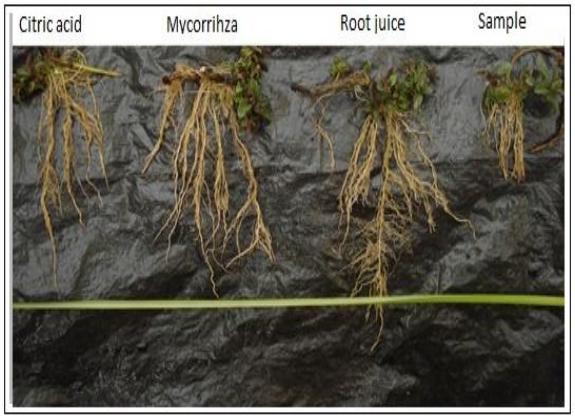

Fig. 11. Effect of treatments on *Mentha piperita* root yield.

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