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Studying the variation of active Fe content in leaves of *Satsuma mandarin* grown in a calcareous soil

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ABSTRACT

This experiment was conducted to study the variation of active iron (Fe) content in leaves of satsuma mandarin (Citrus unshiu) grown in the calcareous soils of eastern Mazandaran. The experiment was conducted in split factorial in the form of a randomized complete block design with four replications. The main factor was date (March 16, April 25, June 5 and July 16), and the sub factors were soil (three soils with different calcium carbonate content) and rootstock (sour orange, citrumelo and troyer citrange). Results indicated that bicarbonate concentration was different in four dates. Bicarbonate concentration reduced in date four and in soils one and two; however, it increased in date four and soil three, and in the second sampling date, bicarbonate concentration was higher in soil three than other soils. Generally, the highest active Fe content was achieved in date four, soil one and troyer rootstock.

Keywords: bicarbonate - Citrus unshiu- iron - rootstock.

INTRODUCTION

Evergreen citrus shrubs are from Rutaceae family with three main genera: poncirus, fortunella and citrus [18]. Poncirus has leaves with three leaflets and very bitter fruits which are not edible. Fortunella has relatively small leaves along with olive shaped fruits commonly known as kumquat. Citrus, a very well known genus of rutaceae family, includes major species such as oranges, sour oranges and mandarins. Satsuma (unshiu) is the most important early cultivar of mandarin, which is resistant to cold stress.

Selection of a suitable rootstock for each area is a very important point which must be conducted based on the cultivar of scion, environmental and soil conditions, and the common diseases of the area. The most common rootstocks in Mazandaran province of Iran are sour orange, citrange, poncirus and citrumelo [10]. In citrus cultivation, soil physical properties are important because of the effect of soil aeration and penetrability. The most favorable soil pH for citrus plants is 6-7.5. Most Iran soils have high calcium carbonate content which increases soil pH and interferes with the absorption of P, Fe and Zn; lower Fe absorption means lower yield in fruit trees, especially in citrus [8].

Results of a study indicated a sever reduction in yield of two peach cultivars (Carson and Babygold) when cultivated in calcareous soil. The yield was 50-60 t/ha in normal soil; however, reduced to 12-15 t/ha in calcareous soil [20]. In experiments conducted to detect Fe deficiency in young leaves of avocado by phenanthroline, it was indicated that Fe (II) content was higher in green leaves than in leaves with chlorosis symptoms [1]. Different experiments are conducted to study the resistance of citrus plants to Fe deficiency in soil and in nutrient solution. Results were different in soil experiments and nutrient solution experiments; overall result of these experiments indicated that sour orange and troyer rootstocks were more tolerant to Fe deficiency compared with citrumelo [4, 21]. So, the objective of this experiment was to study the variation of active Fe content in leaves of satsuma mandarin (*Citrus unshiu*) grown in calcareous soils of eastern Mazandaran province, Iran.

MATERIALS AND METHODS

This pot experiment was conducted in 2011 at the Agricultural and Natural Resources Research Station of Mazandaran Province (Pahnab Juybar Station: 36° 59' N, 52° 55' E and 11 m above the sea level), Iran. The area has a temperate climate with loam soil texture. Other soil properties are listed in Table 1.

Table 1. Physico-chemical properties of the test site soil.

Soil sample	EC (dsm^{-1})	pH	TNV (%)	OC (%)	P	K	Mg	Fe	Mn	Zn	Cu
					$Mgkg^{-1}$						
1	0.64	7.3	5	1.07	15	403	490	4.4	8.3	3	3.2
2	0.89	7.63	12	1.7	17	504	690	12.1	10.2	3.8	3.4
3	0.61	7.8	18.9	1.8	14	360	537	8.7	7.9	0.69	2.7

The experiment was conducted in split factorial in the form of a randomized complete block design with four replications. The main factor was sampling date (March 16, April 25, June 5 and July 16), and the sub factors were soil (three soils with different calcium carbonate content) and rootstock (sour orange, citrumelo and troyer citrange).
First of all, soil samples were taken from the field (0-40 cm) and were passed through a 2 mm sieve. These soil samples were used to determine the physico-chemical properties [14] on one hand, and to fill the pots on the other hands.

After filling the 20 kg pots (30 cm diameter × 30 cm dept), one year old satsuma mandarin plants grafted on sour orange, citrumelo and troyer were planted in the pots. The first sampling was conducted on March 16 from the soil around the roots in pots. Theses samples were dried, crushed and passed through a 2 mm sieve. Saturated clay was prepared from these soil samples and extraction was conducted using vacuum pump. Bicarbonate content of the extraction was measured by titration with sulfuric acid 0.05 normal.

The first sampling from young leaves was conducted on March 16. One gram of leaf blade except for the main vein was weighted and grinded in Chinese mortar. After adding orthophenanthroline 1.5% with pH of 3, and after 20 minutes, samples were ready for extraction. Fe (II) content in the extract was determined by spectrophotometer (model, Pharmacia; wavelength, 510 nm) [1]. Other samples were taken in the mentioned dates. Data were analyzed using MSTATC, the relation of soil solution bicarbonate content with leaves active Fe content in different soils was evaluated using regression equations, and finally, means were compared by Duncan's multiple range test ($P\leq0.05$).

RESULTS AND DISCUSSION

Results indicated the effect of soil on leaves Fe content ($p\leq0.01$); the effect of date and rootstock was not significant. Moreover, the interaction of date × soil, date × rootstock, and date × soil × rootstock had also a significant effect on leaves Fe content. The highest leaves Fe content (20.44 $mgkg^{-1}$) was achieved in date four, which was 71.9% higher than date two (Fig. 1). Abadia et al. [6] reported that Fe induced chlorosis in a garden varied in different times.

Among different soils, soil one had the highest effect on leaves Fe content (Fig. 2). This result is in agreement with those of Mengel et al. [11] and Tagliavini and Rombola [16]. They reported that soil texture, calcium carbonate content and organic matter affect Fe deficiency in plants. Results of this experiment indicated that troyer had the highest, and citrumelo had the lowest effect on leaves Fe content (Fig. 3). Manthey et al. [9] also reported that the ability of plant roots to absorb Fe is different and the processes in rhizosphere affect Fe deficiency.

Studying the interaction of soil × date indicated that in all soils, the highest leaves active Fe content was achieved in date four and the lowest was achieved in date two, which increased by 71.9, 58.4 and 86.5% in soils one, two and three, respectively (Table 2). Wallihan et al. [3] studied orange trees cultivated in calcium carbonate rich soils in different times and concluded that when the temperature was 16°C, Fe chlorosis was higher than the temperature of 19-22°C. They reported that the symptoms of Fe deficiency are more common in spring, mainly due to high precipitation which makes soil saturated, increased bicarbonate content and low temperature.

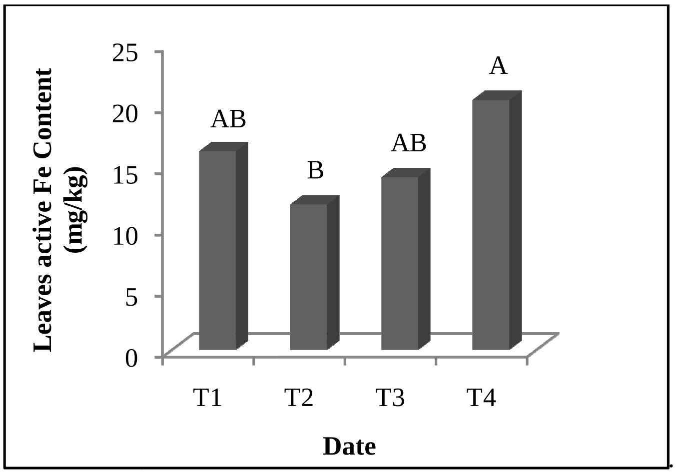

Figure 1. The effect of sampling date on leaves active Fe content.
T_1, March 16; T_2, April 25; T_3, June 5; T_4, July 16.
Same letters indicate non-significant differences between the treatments ($p \leq 0.05$).

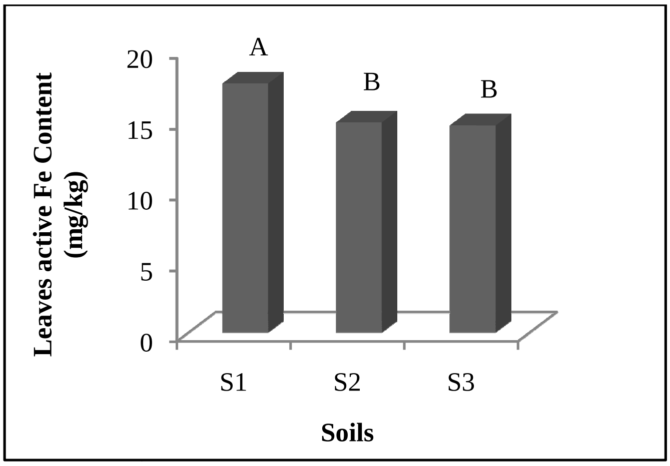

Figure 2. The effect of soil on leaves active Fe content.
S_1, soil one; S_2, soil two; S_3, soil three.
Same letters indicate non-significant differences between the treatments ($p \leq 0.05$).

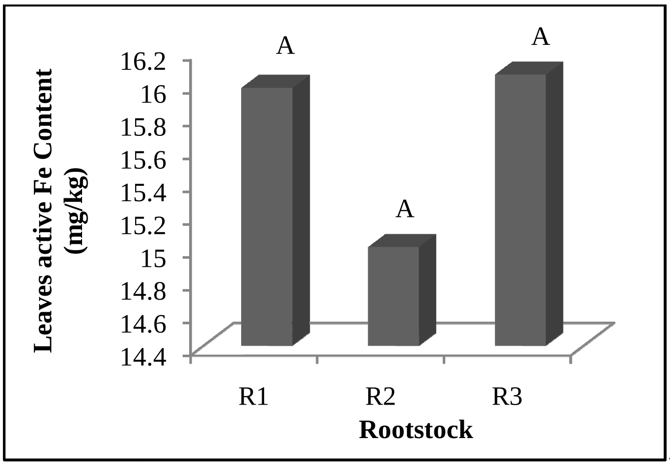

Figure 3. The effect of rootstocks on leaves active Fe content.
R_1, sour orange; R_2, citrumelo; R_3, troyer.
Same letters indicate non-significant differences between the treatments ($p \leq 0.05$).

Table 2.The effect of interaction of soil × date on leaves active Fe content (mg kg^{-1}).

Date	Soil		
	Soil one	Soil two	Soil three
March 16	19.38b	15.44c	13.91cd
April 25	13.15cd	11.71cd	10.81d
June 5	15.18c	13.61cd	13.57cd
July 16	22.61a	18.55b	20.17ab

Same letters indicate non-significant differences between the treatments (p≤0.05).

Studying the interaction of rootstock × date indicated that in all rootstocks, the highest leaves active Fe content was achieved in date four and the lowest was achieved in date two. Active Fe content was the highest in sour orange × date four and the lowest in citrumelo × date two (Fig. 4).

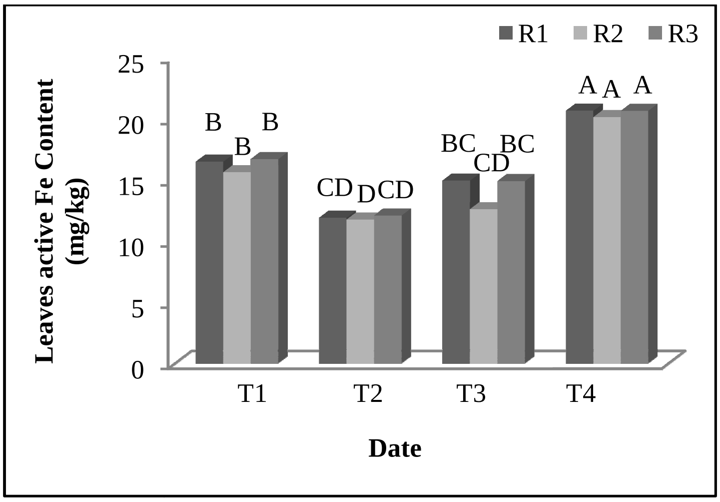

Figure 4. The effect of interaction of rootstock × dateon leaves active Fe content.
R_1, sour orange; R_2, citrumelo; R_3, troyer.
T_1, March 16; T_2, April 25; T_3, June 5; T_4, July 16.
Same letters indicate non-significant differences between the treatments (*p*≤0.05).

Studying the effects of rootstock × soil interaction showed that troyer × soil one had the highest active Fe content (18.66 mg kg^{-1}) which was 12.4% higher than sour orange × soil one. In soil two and three, sour orange had higher active Fe content but citrumelo had lower effect on Fe content (Table 3). This result was found in other experiments [5, 7, 13, 17]. Nikolic and Romheld [13] reported that high bicarbonate content in soil solution is not the only factor that increases soil pH and inactivate Fe in leaves, but root reaction in different soils is also important.

Table 3. The effect of interaction of rootstock ×soil on leaves active Fe content (mg kg^{-1}).

Rootstock	Soil		
	Soil one	Soil two	Soil three
Sour orange	16.6abc	15.39bcd	15.92bcd
Citrumelo	17.47ab	14.25cd	13.28d
Troyer	18.66a	14.85bcd	14.65cd

Same letters indicate non-significant differences between the treatments (p≤0.05).

The three-fold interaction of soil × rootstock × date indicated that leaves active Fe content was 24.51 mg kg^{-1} in date four × soil one × troyer, and reduced to 9.54 mg kg^{-1} in date two × soil three × citrumelo. In soil two, active Fe content in date four × sour orange was 87.7% higher than date three × citrumelo (Table 4). Pestana et al. [15] reported that different citrus species have different ability in Fe absorption. Maribela et al. [19] and Hamze et al. [12] also studied citrus trees in calcareous soils and categorized trees to four groups including highly resistant, partially resistant, semi resistant and non-resistant, based on chlorosis symptoms induced by lime. In their categorization, citrumelo was non-resistant and troyer was semi resistant.

Table 4. The effect of three-fold interaction of rootstock × soil × date on leaves active Fe content (mg kg^{-1}).

Date	Soil one			Soil two			Soil three		
	Sour orange	Citrumelo	Troyer	Sour orange	Citrumelo	Troyer	Sour orange	Citrumelo	Troyer
March 16	18.6b-g	18.85a-g	20.67a-e	15.24d-j	15.46ci	15.61c-i	15.52c-i	12.49ij	13.73g-j
April 25	11.61ij	14.22f-j	13.62g-j	11.62ij	11.41ij	12.11ij	12.4ij	9.54j	10.48ij
June 5	15.02e-j	14.55f-j	15.96c-i	14.96e-j	10.51ij	15.36c-j	14.78f-j	12.66h-j	13.28g-j
July 16	21.16abc	22.25ab	24.41a	19.73a-f	19.63a-f	16.31c-i	20.98a-d	18.42b-h	21.11a-c

Same letters indicate non-significant differences between the treatments ($p \leq 0.05$).

Results of this experiment indicated that date and the interaction of date × soil significantly affected leaves Fe content at $p \leq 0.01$; the effect of soil was also significant at $p \leq 0.05$. The highest bicarbonate content was observed in date one which was 31.27% higher than date two (Fig. 5). These results are in agreement with those of Zuo et al. [22].

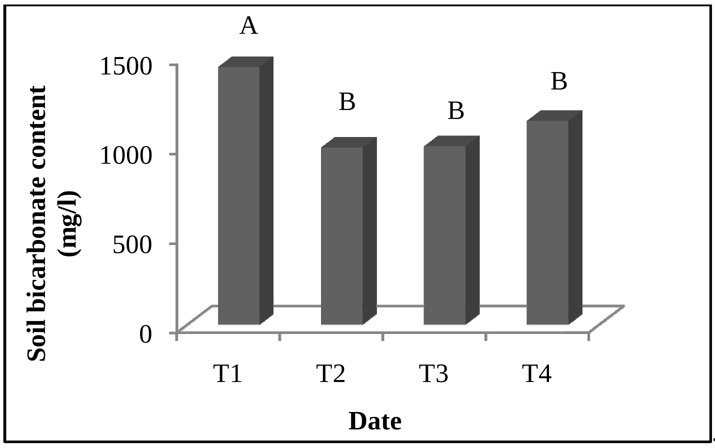

Figure 5. The effect of sampling date on bicarbonate content.
T_1, March 16; T_2, April 25; T_3, June 5; T_4, July 16.
Same letters indicate non-significant differences between the treatments ($p \leq 0.05$).

The highest bicarbonate content in soil solution was observed in soil two which reduced by 18.18% in soil three (Fig. 6). The interaction of date × soil significantly affected soil bicarbonate content; the highest bicarbonate content was observed in date one × soil two (1881 mg l^{-1}) and the lowest in date one × soil three (711.7 mg l^{-1}) (Fig. 7). Same results were reported by Alcantara et al. [2]. They studied Fe deficiency in various trees and reported that high precipitation makes the soil saturated, increases bicarbonate content, inhibits root development and reduces Fe availability to plants.

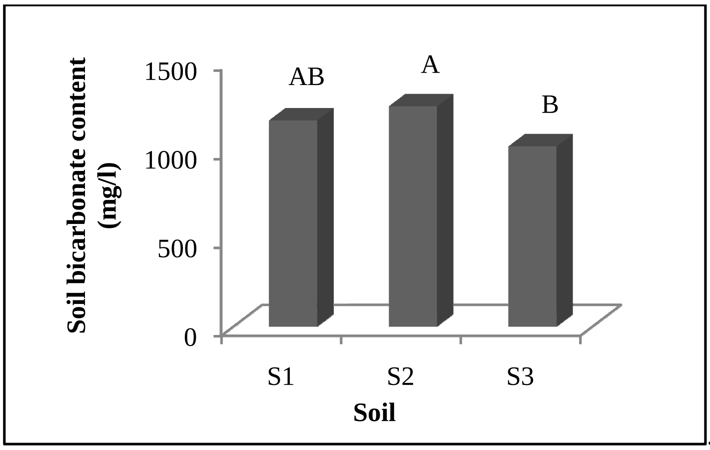

Figure 6. The effect of soil on bicarbonate content.
S_1, soil one; S_2, soil two; S_3, soil three.
Same letters indicate non-significant differences between the treatments ($p \leq 0.05$).

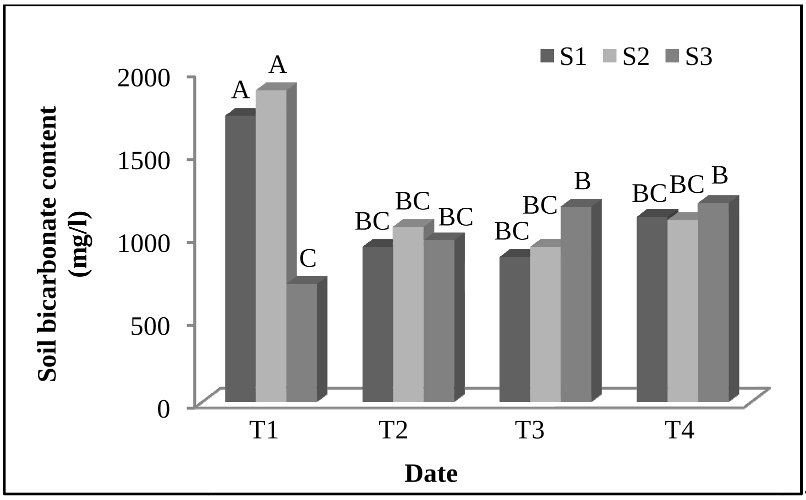

Figure 7. The effect of interaction of soil × date on bicarbonate content.
S_1, soil one; S_2, soil two; S_3, soil three.
T_1, March 16; T_2, April 25; T_3, June 5; T_4, July 16.
Same letters indicate non-significant differences between the treatments ($p \leq 0.05$).

CONCLUSION

Results of this experiment generally indicated that soil bicarbonate content varied in different times in the way that it reduced in date four in soils one and two but increased in date four in soil three. In date two, bicarbonate content was the highest in soil three. The highest active Fe content in leaves was achieved in sampling date four. Soil one had the highest effect on active Fe content. Among the rootstocks, troyer had the highest, and citrumelo had the lowest effect on Fe content. Briefly, troyer rootstock × soil one × date four had the highest effect on active Fe content in leaves, but citrumelo × soil three × date two had the lowest effect.

REFERENCES

[1] A Neaman and L Aguirre, *Journal of Plant Nutrition*, **2007**, 30, 1098–1108.
[2] E Alcantara, I Montilla, P Ramirez, P Garcia-Molina and FJ Romera, *Scientia Horticulturae*, **2012,** 138, 50–54.
[3] EF Wallihan, MJ Garber and RG Sharpless, *Hortscience*, **1976**, 9, 200–201.
[4] H Benyahia, L Beniken, FZ Omari, A Benazzouze, N Handaji, Y Msatef and P Olitrault, *African Journal of Agricultural Research*, **2011**, 6, 780–784.
[5] H Kosegarten and H Koyro, *Physiologia Plantarum*, **2001**, 113, 515–522.
[6] J Abadia, A Fernandez, A Rombola, AD Sanz, M Tagliavini and A Abadia, *Journal of Soil Science and Plant Nutrition*, **2004,** 50, 965–971.
[7] J Masalha, H Kosegarten, E Omer and K Mengel, *Journal of Biology and Fertility of Soils*, **2000**, 30, 433–439.
[8] J Saleh and MJ Malakouti, The role of sulfur in quality and yield improvement of agricultural products. Technical Bulletin of Soil and Water Research Institute, No 447. Sana Publications, Iran, **2005**. (In Farsi).
[9] JA Manthey, DL Mccoy and DE Crowley, *Plant Physiology and Biochemistry*, **1994**, 32, 211–215.
[10] K Daneshgar, A Comprehensive Guide to Citrus Cultivation, Marz-e-Danesh Publications, Iran, **2007**. (In Farsi)
[11] K Mengel, MTh Breininger and W Bubl, *Plant and Soil*, **1984**, 81, 333–344.
[12] M Hamze, J Ryan and M Zaabout, *Journal of Plant Nutrition*, **1986**, 93, 459–469.
[13] M Nikolic and V Romheld, *Plant and Soil*, **1999**, 215, 229–237.
[14] M Pansu and Y Gautheyrou, *Handbook of Soil Analysis: Mineralogical, Organic and Inorganic Methods*, USA Springer, **2006.**
[15] M Pestana, AD Varennes and EA Faria, *Food Agriculture and Environment*, **2003**, 1, 46–51.
[16] M Tagliavini and AD Rombolà, *European Journal of Agronomy*, **2001**, 15, 71–92.
[17] M Treeby and N Uren, *Journal of Plant Nutrition and Soil Science*, **1993**, 156, 75–81.
[18] N Griffin, Grow citrus (the insider secrets to growing great citrus), Craft Pty ltd, Australia, **2010**.
[19] P Maribela, V Amarilisde, A Javier and AF Eugenio, *Scientia Horticulturae*, **2005**, 104, 25–36.
[20] V Fernandez, V Delrio, L Pumarino, E Igartua, J Abadia and A Abadia, *Scientia Horticulturae*, **2008**, 117, 241–248.
[21] WS Castel and J Nunnallee, *Hortscience*, **2009**, 44, 638–645.
[22] Y Zuo, L Ren, F Zhang and RF Jiang, *Journal of Plant Physiology and Biochemistry*, **2007**, 45, 357–364.