

## Reduction of Lime-Based Iron Chlorosis in Apple Trees\*

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### Abstract

**Objective:** This study was conducted with the aim of determining the effects of different applications (Fe fertilizers, K-Humate, and sulfur) on shoot length, active and total Fe, chlorophyll a, b, and a+b, fruit Fe concentration, yield efficiency, and yield of apple.

**Materials and Methods:** In the study, which was conducted in 4 replications according to trial pattern of chance blocks in an apple orchard with a lot of lime and alkaline reactions, 13 applications (Control, FeSO<sub>4</sub>.7H<sub>2</sub>O (19% Fe), Elemental S, FeSO<sub>4</sub>.7H<sub>2</sub>O + Elemental S, K-Humate, FeSO<sub>4</sub>.7H<sub>2</sub>O + K-Humate, Fe-EDTA (%13 Fe), Fe-DTPA (%6 Fe), Fe-HBED (%6 Fe), Fe-EDDHA (o-o:2.2) (%6 Fe), Fe-EDDHA (o-o:3.5) (%6 Fe), Fe-EDDHA (o-o:4.8) (%6 Fe), and Fe-EDDHA (o-o:5.25) (%6 Fe)).

**Results:** As a result of the study, the effects of the applications on the investigated parameters were found to be statistically significant, and the maximum effect was determined by FeSO<sub>4</sub>.7H<sub>2</sub>O+K-Humate and Fe-EDDHA (o-o: 5.25) applications. Since there is no difference between these applications, Fe-EDDHA (o-o: 5.25) or Fe-chelate fertilizers can be used instead of FeSO<sub>4</sub>.7H<sub>2</sub>O+K-Humate.

**Conclusion:** In addition, it has been determined that iron intake by plants increased as ortho-ortho isomer ratios increase in Fe-chelate fertilizers. It has been determined that applying Fe-EDDHA (o-o: 5.25), one of the Fe-chelate fertilizers, is more effective than other applications in reducing and eliminating chlorosis in calcareous soils.

**Keywords:** Apple, Fe chelate fertilizers, Chlorosis, Yield

### Elma Ağaçlarında Kireç Kaynaklı Demir Klorozunun Azaltılması

#### Öz

**Amaç:** Bu çalışma, farklı uygulamalarının (Fe'li gübreler, K-Humat ve kükürt) elmanın sürgün uzunluğu, aktif ve toplam Fe, klorofil a, b ve a+b, meyve Fe konsantrasyonu, verim etkinliği ve verimine etkilerini belirlemek amacıyla yürütülmüştür.

**Materyal ve Yöntem:** Çalışmada çok fazla kireçli, alkalın reaksiyonlu bir elma bahçesinde tesadüf blokları deneme desenine göre 4 tekrarlamalı olarak yürütülen çalışmada 13 adet uygulama (Kontrol, FeSO<sub>4</sub>.7H<sub>2</sub>O (%19 Fe), Elementel S, FeSO<sub>4</sub>.7H<sub>2</sub>O + Elementel S, K-Humat, FeSO<sub>4</sub>.7H<sub>2</sub>O + K-Humat, Fe-EDTA (%13 Fe), Fe-DTPA (%6 Fe), Fe-HBED (%6 Fe), Fe-EDDHA (o-o:2.2) (%6 Fe), Fe-EDDHA (o-o:3.5) (%6 Fe), Fe-EDDHA (o-o:4.8) (%6 Fe), ve Fe-EDDHA (o-o:5.25) (%6 Fe)) yapılmıştır.

**Araştırma Bulguları:** Çalışma sonucunda, uygulamaların incelenen parametreler üzerine etkileri istatistiki bakımdan önemli bulunmuş olup en fazla etki FeSO<sub>4</sub>.7H<sub>2</sub>O+K-Humat ve Fe-EDDHA (o-o: 5.25) uygulamaları ile belirlenmiştir. Bu uygulamalar arasında fark olmaması nedeniyle bitkinin gerek demir beslenmesinde gerekse verim ve verim etkinliğinin artırılmasında FeSO<sub>4</sub>.7H<sub>2</sub>O+K-Humat yerine Fe-EDDHA (o-o: 5.25) veya Fe-kleytli gübreler kullanılabilir.

**Sonuç:** Fe-kleytli gübrelerde orto-orto izomer oranları arttıkça bitkilerce demir alımının arttığı belirlenmiştir. Kireçli topraklarda demir klorozunun azaltılması ve giderilmesinde Fe-kleytli gübrelerden

Fe-EDDHA (o-o: 5.25) uygulamasının diğer uygulamalara göre daha etkili olduğu belirlenmiştir.

**Anahtar kelimeler:** Elma, Fe kleytli gübreler, kloroz, verim

## Introduction

Iron (Fe) deficiency is seen in 40% of the soils (Zengin and Gezgin 2013), and it is seen in the form of chlorosis, especially in fruit trees grown in calcareous alkaline soils and poor in organic matter (Vigani et al., 2013), the most important reason for this is that the soils are too calcareous. (Zhao et al., 2021). It is known that apple, peach, quince, kiwi, and pear are known to be most susceptible to Fe deficiency among fruit trees (Pestana et al., 2012). Chlorosis caused by the reduction of chlorophyll content in the chloroplasts of the plant (Rajaie et al., 2018), harms the plant shoot growth (Aboutalebi and Hassanzadeh 2013), the number and size of the fruits (Rombolà and Tagliavini 2006), active Fe and total Fe content (Aras et al., 2018) and especially the yield (Şahin et al., 2017). In addition to decreasing the pH in alkaline and increasing organic matter soils in order to eliminate Fe deficiency the most effective way is to use Fe-containing fertilizers. For this purpose, inorganic compounds ( $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$ ), Fe-chelates (DTPA, EDDHA, EDTA, HBED), and natural Fe-complexes (humates, lignosulfonates, amino acids, gluconate, and citrate) are used (Abadia et al., 2011). When inorganic compounds are applied to calcareous

basic soils, they rapidly transform into a form that plants cannot take (Hansen et al., 2006), Fe-chelates have a high ability to bind Fe in plant roots and provide stability (Rodríguez-Lucena et al., 2010), and natural Fe-complexes increase Fe uptake by plants by chelating Fe, keeping it in the root rhizosphere region so that the plant can use it easily and preventing Fe from binding in the soil. Therefore, the aim of this study was to determine the effects of chlorosis caused by lime and different Fe sources ( $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$ ,  $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$  + Elemental S, K) -Humate, Fe-EDTA, Fe-DTPA, Fe-HBED, Fe-EDDHA with ortho-ortho isomer ratio of 2.2, 3.5, 4.8 and 5.25) on apple shoot length, active and total Fe, chlorophyll a, b and a+b, fruit Fe concentration, yield efficiency and yield.

## Material and Method

### Plant material and experimental site

The study was carried out in the orchard where Fe deficiency was clearly seen in the Ayrancı district of Karaman Province in 2016, and 16 year-old apple trees, Starking Delicious cultivar grafted on MM106 rootstock were grown. The year the experiment was established, the total rainfall was 246 mm, and the average temperature was 12.1 °C.

### Soil material and experimental design

The results of some physical and chemical analysis of the orchard soil taken from the depths of 0-30 cm, 30-60 cm, and 60-90 cm from the tree projection before the experiment are given in Table 1.

Table 1. Some physical and chemical analysis results of orchard soil

Soil properties	Unit	Analysis results			Method
		0-30	30-60	60-90	
pH (1: 2.5 soil-water mixture)	--	7.60	7.62	7.70	(Jackson 1962)
EC (1: 5 soil-water mixture)	$\mu\text{S cm}^{-1}$	190	184	170	
Texture	--	Clayey			(Bouyoucos 1951)
Lime		45.1	43.6	43.1	(Kacar 1994)
Active Lime	%	11.53	11.49	11.02	(Özgümüş 1999)
Organic Matter		1.07	1.03	0.87	(Smith and Weldon, 1941)
Inorg. Nitrogen ( $\text{NH}_4 + \text{NO}_3$ )		3.64	3.12	3.08	(Kacar 1994)
Retrievable K		145	116	83	(Kacar 1994)
Retrievable Ca		6343	6648	6854	
Retrievable Mg		444	445	388	
Retrievable Na		28	24	19	
Retrievable P	$\text{mg kg}^{-1}$	6.22	4.56	4.23	(Bayraklı 1987)
Retrievable Zn		0.75	0.32	0.17	(Lindsay and Norvell 1978)
Retrievable Cu		1.14	1.00	0.68	
Retrievable Fe		2.80	2.54	2.05	
Retrievable Mn		5.53	4.76	3.09	
Retrievable B		2.78	2.66	2.76	(Cartwright et al., 1983)

The experimental soil is clayey, has a slightly alkaline reaction, does not contain salinity to prevent plant growth, and has high lime and low organic matter

content. It also contains large amounts of extractable Ca and Mg, sufficient amounts of K, Mn, Cu, and B, insufficient amounts of N, Zn, and P. Although the

available Fe content of the soil varies according to the soil depth, according to the limit values (insufficient <2.5 mg Fe kg<sup>-1</sup>, 2.5-4.5 mg kg<sup>-1</sup> moderate) of Lindsay and Norvell (1978), it is cm at the medium level at 0-30 cm and 30-60, and insufficient at 60-90 cm.

Thirteen different applications carried on in the study are as follows: 1. Control, 2. FeSO<sub>4</sub>.7H<sub>2</sub>O (%19 Fe), 3. Elemental S, 4. FeSO<sub>4</sub>.7H<sub>2</sub>O + Elemental S, 5. K-Humate, 6. FeSO<sub>4</sub>.7H<sub>2</sub>O + K-Humate, 7. Fe-EDTA (%13 Fe), 8. Fe-DTPA (%6 Fe), 9. Fe-HBED (%6 Fe), 10. Fe-EDDHA (o-o:2.2) (%6 Fe), 11. Fe-EDDHA (o-o:3.5) (%6 Fe), 12. Fe-EDDHA (o-o:4.8) (%6 Fe) and 13. Fe-EDDHA (o-o:5.25) (%6 Fe) (Table 2). In addition, 2 kg of compound fertilizer per tree (15.15.15+1% Zn) and 50 g ZnSO<sub>4</sub>.7H<sub>2</sub>O (23% Zn, 11% S) were used as basic fertilization so that the insufficient N, P, and Zn according to the analysis results of the soil would not affect the experiment. In the amounts specified in Table 2, K-Humate (It has contains pH of 11, 12% Humic+fulvic acid and 3% K<sub>2</sub>O) and elemental-S dissolved in 5 liters of water were applied equally to the places at depth of 25-30 cm opened to the crown projections of the trees on February 04, 2016.

Table 2. Application resources and quantities

Applied resources	Applied Amount
Control	--
FeSO <sub>4</sub> .7H <sub>2</sub> O	95 g Fe ağaç <sup>-1</sup>
Elemental S	3 kg elemental sulphur tree <sup>-1</sup>
FeSO <sub>4</sub> .7H <sub>2</sub> O + Elemental S	95 g Fe + 3 kg elemental sulphur tree <sup>-1</sup>
K-Humate	3L K-Humat tree
FeSO <sub>4</sub> .7H <sub>2</sub> O + K-Humate	95 g Fe + 3L K-Humat tree <sup>-1</sup>
Fe-EDTA	12 g Fe tree <sup>-1</sup>
Fe-DTPA	12 g Fe tree <sup>-1</sup>
Fe-HBED	12 g Fe tree <sup>-1</sup>
Fe-EDDHA (o-o: 2.2)	12 g Fe tree <sup>-1</sup>
Fe-EDDHA (o-o: 3.5)	12 g Fe tree <sup>-1</sup>
Fe-EDDHA (o-o: 4.8)	12 g Fe tree <sup>-1</sup>
Fe-EDDHA (o-o: 5.25)	12 g Fe tree <sup>-1</sup>

The study was established according to the randomized blocks experimental design with 4 replications and was carried out on 104 trees (52 plots) with 2 trees in each replication.

#### Data collection and analysis

In the transition phase of apple trees to the generative period, the shoot length was measured with the help of meters from all sides of the tree (north-south-east-west) one month after the shoots emerged from the main branches and trunk reached full maturity (Atılgan 2009). Leaf samples were taken from young leaves that have completed their growth from all four sides of the tree as stated by Kacar (2014). The samples taken were washed first with tap water, then with distilled water, 0.2 N HCl solution, and distilled water, respectively and removed excess water on blotting paper. In these fresh samples, active Fe and chlorophyll a, b, a+b analyses were made according to Horwitz (1970) according to Takkar and Kaur (1984). The fruit of the tree are then harvested and the fruit

of the tree is harvested (kg tree<sup>-1</sup>). The dried and ground leaf and fruit samples were dissolved in a microwave device (MarsXpress, CEM Corp., USA) under high pressure (200 PSI) and total Fe concentrations were determined in the ICP-AES (Varian, Australia Pty Ltd. Mulgrave, Australia) device. In addition, the yield values of the harvested fruits were calculated using the formulas 1, 2, and 3 below (Küçükler et al., 2011).

$$\text{Tree Trunk Diameter (r)(cm)} = \text{tree circumference (C)}/\pi \quad (1)$$

$$\text{Trunk cross - sectional area (cm}^2\text{)} = \pi r^2 \quad (2)$$

$$\text{Trunk cross-sectional area (cm}^2\text{)} = \text{yield per / trunk cross-sectional area} \quad (3)$$

#### Statistical analysis

The data obtained within the scope of the experiment were subjected to statistical evaluation with the MSTAT-C statistical package program by the Randomized Complete Block Design. The variance analysis was performed to determine the differences between the shoot length, active Fe, total Fe, chlorophyll concentration of the leaf sample, the fruit of Fe concentration, yield, and yield efficiency belonging to each application group.

#### Results and Discussion

##### Shoot length of apple trees

The effects of the applications on the shoot length of apple trees are given in Table 3. The effect of applications on plant shoot length was found to be statistically significant at the level of 1%. This significant effect is an indication that the plant shoot length changes depending on the applications and increases in accordance with the control. Shoot length varies in accordance with the control (46.9 cm), with an increase of 3% (Fe-DTPA) to 39% (FeSO<sub>4</sub>.7H<sub>2</sub>O + K-Humate). Although the plant shoot length increased with only Fe fertilizer applications compared to the control it has been determined that Fe-EDDHA applications were more effective, and their effectiveness increased as the ratio of ortho-ortho isomer increased.

Table 3 Effects of applications on shoot length of apple trees

Applied resources	Applied resources (cm)
Control	46.9 <sup>D</sup>
FeSO <sub>4</sub> .7H <sub>2</sub> O	55.0 <sup>BC</sup>
Elemental S	51.2 <sup>BCD</sup>
FeSO <sub>4</sub> .7H <sub>2</sub> O + Elemental S	54.6 <sup>BC</sup>
K-Humate	58.6 <sup>AB</sup>
FeSO <sub>4</sub> .7H <sub>2</sub> O + K-Humate	65.2 <sup>A</sup>
Fe-EDTA	53.4 <sup>BCD</sup>
Fe-DTPA	48.3 <sup>CD</sup>
Fe-HBED	53.8 <sup>BCD</sup>
Fe-EDDHA (o-o: 2.2)	57.7 <sup>AB</sup>
Fe-EDDHA (o-o: 3.5)	55.3 <sup>BC</sup>
Fe-EDDHA (o-o: 4.8)	58.8 <sup>AB</sup>
Fe-EDDHA (o-o: 5.25)	56.9 <sup>B</sup>
<b>Average</b>	<b>59.6</b>

A, B: p<0.01

As a matter of fact, in their studies (Rombola and Tagliavi 2006; Abay 2017), it is reported that ferrous fertilizer applications increased plant shoot length, and Fe-chelated fertilizers were more effective than other fertilizers.

#### Active Fe, total Fe, and chlorophyll concentrations of leaf samples

The effects of the applications on the active and total Fe, chlorophyll a, b, and a+b concentrations of apple leaves are statistically significant at the level of 1% and are given in Table 4. According to active and total Fe ( $\text{Fe}^{+2}+\text{Fe}^{+3}$ ) concentrations controls (10.3 and 102.6  $\text{mg kg}^{-1}$ ) of the leaves, the least increase was determined by Elemental S application while it was determined the highest by FeEDDHA (o-o:5.25) applications. In all applications, the total Fe content of the leaves was sufficient according to the limit values (50-300  $\text{mg kg}^{-1}$ ) specified for the apple plant by Jones et al., (1991). Similarly, in studies conducted on apple (Bozkurt et al., 2000; Mordoğan and Ergün 2002), peach (Çelik and Katkat 2007), vineyard (Gezgin and Er 2001) and strawberry (Özden and Ayanoglu 2002) plants, it has even been found that the total Fe concentrations of the leaves were sufficient when Fe chlorosis was observed. And even total Fe concentrations of the leaves with chlorosis were

higher than the leaves that did not show any signs of chlorosis (Horuz et al., 2016). However, although there is sufficient total Fe in the plant leaves in all applications since the signs of Fe deficiency are still observed, total Fe should not be evaluated alone as a criterion in feeding the plants with Fe, as well as the ratio of active Fe in active Fe and total Fe, should be taken into account (Sönmez and Kaplan, 2005; Çelik and Katkat, 2007; Akgül et al., 2013). As a matter of fact, in the studies (Gezgin and Er 2001; Yılmaz et al., 2021), it is reported that the ratio of  $\text{Fe}^{+2}$ ,  $\text{Fe}^{+2}+\text{Fe}^{+3}$ , and  $\text{Fe}^{+2}/\text{Fe}^{+2}+\text{Fe}^{+3}$  varies according to applied to Fe applications in plant varieties in order for Fe to fulfill its metabolic functions in plants. The active Fe / total Fe ratio of the leaves varies between 0.21 and 0.25, and this ratio was determined the highest by  $\text{FeSO}_4.7\text{H}_2\text{O}$  + Elemental S and Fe-HBED applications, while the lowest was determined in control. This is due to the fact that the total Fe content of the leaves increases more than the active Fe content of the leaves with different applications, and it changes depending on the applications. In addition, in soils with calcareous basic reactions, the plants cannot get enough Fe, and chlorosis symptoms are seen in the leaves (Fig.1).

Table 4 Effects of applications on active Fe, total Fe, active Fe / total Fe, and chlorophyll concentrations of apple leaves

Applications	Active Fe	Total Fe	Active Fe/Total Fe	Chlorophyll a	Chlorophyll b	Chlorophyll a+b
	(mg kg <sup>-1</sup> )					
Control	10.3 <sup>C</sup>	102.6 <sup>E</sup>	0.10	11.0 <sup>F</sup>	6.4 <sup>D</sup>	17.4 <sup>H</sup>
FeSO <sub>4</sub> .7H <sub>2</sub> O	30.3 <sup>BC</sup>	129.8 <sup>A-D</sup>	0.23	12.7 <sup>EF</sup>	12.2 <sup>C</sup>	24.9 <sup>EF</sup>
Elemental S	27.6 <sup>BC</sup>	116.8 <sup>DE</sup>	0.24	12.4 <sup>EF</sup>	6.7 <sup>D</sup>	19.1 <sup>GH</sup>
FeSO <sub>4</sub> .7H <sub>2</sub> O + Elemental S	31.2 <sup>ABC</sup>	126.2 <sup>BCD</sup>	0.25	14.7 <sup>DE</sup>	7.3 <sup>D</sup>	22.0 <sup>FG</sup>
K-Humate	33.0 <sup>AB</sup>	138.8 <sup>A-D</sup>	0.24	18.3 <sup>BC</sup>	11.8 <sup>C</sup>	30.1 <sup>D</sup>
FeSO <sub>4</sub> .7H <sub>2</sub> O + K-Humate	31.2 <sup>BC</sup>	143.8 <sup>ABC</sup>	0.22	17.5 <sup>BC</sup>	10.9 <sup>C</sup>	28.4 <sup>DEF</sup>
Fe-EDTA	29.5 <sup>BC</sup>	127.7 <sup>BCD</sup>	0.23	14.9 <sup>DE</sup>	6.8 <sup>D</sup>	21.7 <sup>GH</sup>
Fe-DTPA	28.8 <sup>BC</sup>	123.2 <sup>CDE</sup>	0.23	14.6 <sup>DE</sup>	7.5 <sup>D</sup>	22.1 <sup>GH</sup>
Fe-HBED	30.6 <sup>BC</sup>	122.6 <sup>CDE</sup>	0.25	16.5 <sup>CD</sup>	11.8 <sup>C</sup>	28.3 <sup>DE</sup>
Fe-EDDHA (o-o: 2.2)	31.3 <sup>ABC</sup>	139.7 <sup>A-D</sup>	0.22	17.1 <sup>CD</sup>	26.1 <sup>A</sup>	43.2 <sup>BC</sup>
Fe-EDDHA (o-o: 3.5)	31.5 <sup>ABC</sup>	139.5 <sup>A-D</sup>	0.23	18.9 <sup>BC</sup>	23.3 <sup>B</sup>	42.2 <sup>C</sup>
Fe-EDDHA (o-o: 4.8)	33.1 <sup>AB</sup>	149.5 <sup>AB</sup>	0.22	19.7 <sup>B</sup>	26.8 <sup>A</sup>	46.5 <sup>B</sup>
Fe-EDDHA (o-o: 5.25)	35.8 <sup>A</sup>	152.2 <sup>A</sup>	0.24	25.4 <sup>A</sup>	28.1 <sup>A</sup>	53.5 <sup>A</sup>
<b>Average</b>	<b>13.6</b>	<b>131.7</b>	<b>0.22</b>	<b>16.5</b>	<b>14.6</b>	<b>30.7</b>

A, B: p<0.01

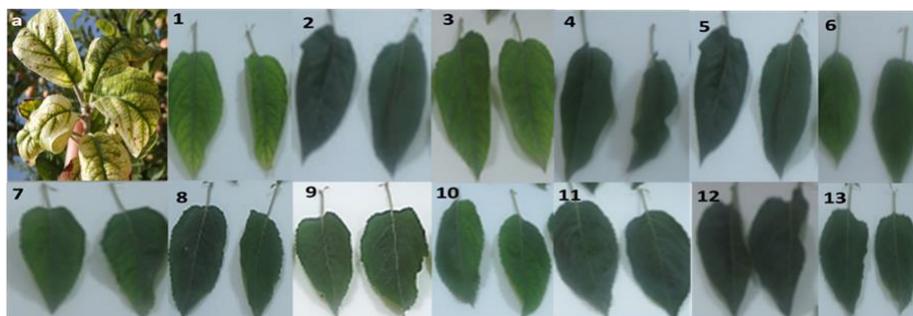


Fig. 1 Picture of apple leaves [a. Before application, 1. Control, 2. FeSO<sub>4</sub>.7H<sub>2</sub>O, 3. Elemental S, 4. FeSO<sub>4</sub>.7H<sub>2</sub>O + Elemental S, 5. K-Humate, 6. FeSO<sub>4</sub>.7H<sub>2</sub>O + K-Humate, 7. Fe-EDTA, 8. Fe-DTPA, 9. Fe-HBED, 10. Fe-EDDHA (o-o:2.2), 11. Fe-EDDHA (o-o:3.5), 12. Fe-EDDHA (o-o:4.8), 13. Fe-EDDHA (o-o:5.25)]

Chlorophyll a, b, and a+b concentrations of the leaves were determined by Fe-EDDHA (o-o: 5.25) application with 2.3-fold, 4.4-fold and 2.9-fold increases, respectively, compared to the controls (11.0, 6.4 and 17.4 mg L<sup>-1</sup>), depending on the applications. This was followed by other Fe-cleated fertilizers. Although Fe is not involved in the structure of the chlorophyll molecule, it plays an important role in the synthesis of chlorophyll-protein compounds in the chloroplasts in the plant (Taiz and Zeiger 2008). Low chlorophyll concentration (chlorosis) in young leaves is the most visible symptom of Fe deficiency (Marschner 1995). Although the amount of chlorophyll in plant leaves changes depending on the Fe content, the amount of chlorophyll increases depending on the increase in Fe. In studies conducted on peach (Molassiotis et al., 2005), blueberry (Michel

et al., 2019), strawberry (Pestana et al., 2012), and apple (Şahin et al., 2017), ferrous fertilizer applications were found to be effective in removing Fe chlorosis. In the studies by Mengel (2001), Karaman (2002), and Yılmaz et al., (2012), they reported that of different Fe compounds can be used in the prevention of Fe chlorosis in plants and that Fe-cleated fertilizer applications are more effective because the plants increase the active Fe/total Fe ratio, chlorophyll a, chlorophyll b and chlorophyll a+b.

#### Fruit Fe concentration, yield, and yield efficiency

Although it varies depending on the applications, the Fe concentration of the apple, the yield per tree, and the yield efficiency are given in Table 5, and were found to be statistically significant at the level of 1% (Table 5).

Table 5 Effects of applications on Fe concentration, yield, and yield efficiency

Applications	Fe (mg kg <sup>-1</sup> )	Yield (kg tree <sup>-1</sup> )	Yield efficiency (kg cm <sup>-2</sup> )
Control	22.6 <sup>D</sup>	27.75 <sup>G</sup>	1.14 <sup>C</sup>
FeSO <sub>4</sub> .7H <sub>2</sub> O	25.4 <sup>BCD</sup>	36.4 <sup>DEF</sup>	1.2 <sup>BC</sup>
Elemental S	23.3 <sup>D</sup>	33.7 <sup>F</sup>	1.2 <sup>BC</sup>
FeSO <sub>4</sub> .7H <sub>2</sub> O + Elemental S	29.6 <sup>B</sup>	40.1 <sup>DE</sup>	1.3 <sup>BC</sup>
K-Humate	39.0 <sup>A</sup>	45.5 <sup>BC</sup>	1.4 <sup>BC</sup>
FeSO <sub>4</sub> .7H <sub>2</sub> O +K- Humate	26.3 <sup>BCD</sup>	53.4 <sup>A</sup>	1.8 <sup>A</sup>
Fe-EDTA	24.3 <sup>CD</sup>	40.4 <sup>CD</sup>	1.3 <sup>BC</sup>
Fe-DTPA	24.1 <sup>CD</sup>	35.1 <sup>DEF</sup>	1.3 <sup>BC</sup>
Fe-HBED	25.3 <sup>BCD</sup>	35.0 <sup>EF</sup>	1.3 <sup>BC</sup>
Fe-EDDHA (o-o: 2.2)	25.9 <sup>BCD</sup>	33.5 <sup>F</sup>	1.2 <sup>C</sup>
Fe-EDDHA (o-o: 3.5)	27.4 <sup>BCD</sup>	38.8 <sup>DEF</sup>	1.5 <sup>B</sup>
Fe-EDDHA (o-o: 4.8)	28.2 <sup>BC</sup>	40.1 <sup>DE</sup>	1.3 <sup>BC</sup>
Fe-EDDHA (o-o: 5.25)	28.6 <sup>BC</sup>	48.1 <sup>AB</sup>	1.5 <sup>B</sup>
<b>Average</b>	26.9	39.1	1.3

A, B: p<0.01

Although apple Fe concentration increased between 3% (Elemental S) and 73% (K-Humate) compared to the control (22.6 mg kg<sup>-1</sup>), it varies according to the amount of fertilizer applied and the sources (Table 4). It has been determined that the Fe concentration of apples is the highest (28.6 mg kg<sup>-1</sup>) with Fe-EDDHA (o-o: 5.25) application with only Fe fertilizer applications, but Fe-cleated applications are more effective, and their efficiency increases as the ortho-ortho isomer ratios increase. In the studies (Özcan et al., 2012; Çoşkun and Aşkın 2016), it is reported that the Fe concentration in Starking Delicious apple varied between 6.84 - 9.77 mg kg<sup>-1</sup> and increased depending on the application of ferrous fertilizers. Although the yield of apple per tree generally increased compared to the control (27.75 kg tree<sup>-1</sup>), the most (53.4 kg tree<sup>-1</sup>) was obtained with the application of 95 g FeSO<sub>4</sub>.7H<sub>2</sub>O and 3 L K-Humate per tree, followed by Fe-EDDHA (o-o: 5.25)>K-Humate>Fe-EDTA>FeSO<sub>4</sub>.7H<sub>2</sub>O+Elemental S=Fe-

EDDHA (o-o: 4.8> Fe-EDDHA (o-o: 3.5)>FeSO<sub>4</sub>.7H<sub>2</sub>O>Fe-DTPA>Fe-HBED>Elemental S>Fe-EDDHA (o-o: 2.2) applications respectively. In addition, since there was no statistical difference between FeSO<sub>4</sub>.7H<sub>2</sub>O + K-Humate application, which has the highest yield per tree, and Fe-EDDHA (o-o: 5.25) application, it has been determined that Fe-cleated fertilizers can be used instead of K-Humate fertilizers. It has been determined that fertilizers with high o-o isomer among Fe-chelate fertilizers has higher shoot length, active Fe, total Fe, chlorophyll a, chlorophyll b, chlorophyll a+b, and yield efficiency than other applications. In addition, statistically significant relationships were determined between the effect of applications on yield per tree and shoot length (r=0.567\*\*), active Fe (r=0.394\*), total Fe (r=0.526\*\*), chlorophyll an (r=0.579\*\*), chlorophyll a+b (r=0.314\*), Fe concentration of fruit (r=0.456\*\*) and yield efficiency (r=0.678\*\*). In their studies, it is reported that the yield per tree in Starking Delicious

variety was 13.20 kg tree<sup>-1</sup> (Çulha, 2010), 13.40 kg tree<sup>-1</sup> and (Soylu et al., 2003), 33.06 kg tree<sup>-1</sup> (Kaith et al., 2011). Álvarez-Fernández et al. (2005 and 2006) stated that Fe deficiency in trees would significantly reduce fruit yield and that potential yield loss could be up to 50% even in trees with moderate Fe chlorosis and stated that ferrous fertilizers should be used to increase yield. Schulte and Kelling (2004) have determined that by increasing Fe intake from the soil by Fe fertilization, inorganic Fe sources (Frossard et al., 2000), and chelated Fe sources (Akrinde 2006) cause increases in yield, and this supports the results of the study. In the leaves where Fe chlorosis is seen, active leaf iron (Fe<sup>+2</sup>) and total iron (Fe<sup>+2</sup>+Fe<sup>+3</sup>) contents were low (Table 4) in control (1), FeSO<sub>4</sub>.7H<sub>2</sub>O (2) and FeEDDHA (o-o: 2.2) (10) applications (Fig.1) when compared to other applications (Table 4) and the yield per tree (Table 5) was low as well. It was determined that the yield efficiency increased with the increase in yield per tree, and the yield efficiency decreased as the trunk cross-sectional area calculated depending on the tree diameter increased, however, the yield efficiency increased as well as the yield per tree with the Fe-chelate fertilizer applications. The significant correlation (r=0.678\*\*) between yield efficiency and yield per tree is an indicator of this. Westwood (1995) stated that the simplest way to express yield according to tree size is to determine yield per trunk cross-sectional area. In addition, it has been reported that the efficiency calculated from the yield per trunk cross-section area can be used in determining the effects of the applications on plant development and yield due to the fact that they are perennial trees, in a short time or in the next year. As a matter of fact, since the study is single year, the efficiency effectiveness, which is the expression of the yield per tree instead of the perennial yield, as well as the yield per tree unit trunk cross sectional area, has been determined, and it has been determined that it varies depending on the applications and that Fe fertilizer applications are effective in increasing the yield efficiency.

### Conclusion

Applications for the removal of Fe chlorosis seen in apple trees in calcareous soils [Control, FeSO<sub>4</sub>.7H<sub>2</sub>O, Elemental S, FeSO<sub>4</sub>.7H<sub>2</sub>O + Elemental S, K-Humate, FeSO<sub>4</sub>.7H<sub>2</sub>O + K-Humate, Fe-EDTA, Fe-DTPA, Fe-HBED, Fe-EDDHA (o-o:2.2), Fe-EDDHA (o-o:3.5), Fe-EDDHA (o-o:4.8), Fe-EDDHA (o-o:5.25)], plant shoot length, active and total Fe, chlorophyll a, b, and a+b concentrations were effective on apple Fe

concentration, yield per tree and yield efficiency. Although the shoot length of the apple tree, active (Fe<sup>+2</sup>) iron, total (Fe<sup>+2</sup>+Fe<sup>+3</sup>) iron, chlorophyll a, chlorophyll b, and chlorophyll a+b concentrations, and yield increased compared to the control, the maximum yield was 95 g FeSO<sub>4</sub>.7H<sub>2</sub>O per tree together with 3 L K-Humate (FeSO<sub>4</sub>.7H<sub>2</sub>O+K-Humate) and 12 g Fe-EDDHA (o-o: 5.25) applications. Since there is no statistical difference between these applications, it has been determined that Fe-chelate fertilizers can be used instead of Fe-containing fertilizers together with K-Humate. In Fe-fertilizer, it was found that Fe-EDDHA applications were more effective on the investigated parameters examined, and their effectiveness increased as the ortho-ortho isomer ratios increased. For this reason, it has been determined that in apple orchards where chlorosis with alkaline reaction is seen, in order to reduce the effect of chlorosis or to increase the yield by eliminating the effect of chlorosis, it is necessary to use Fe-containing fertilizers, especially Fe-clay fertilizers with high Fe binding capacity. In addition, it was found that the application of Fe-EDDHA fertilizer with 5.25 ortho-ortho isomer was the most effective application.

### Conflict of Interest

Authors declare that they have no competing interests.

### Author Contribution

All authors participated in this study the arranging and development. This study was taken from a part of AK's Master's thesis. AK contributed to this work in the experimental design and setup, lab processing of samples, data analysis, manuscript writing, and discussion. FGY, MH, and SG data interpretation, article writing, and discussion. The authors approved the final manuscript.

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