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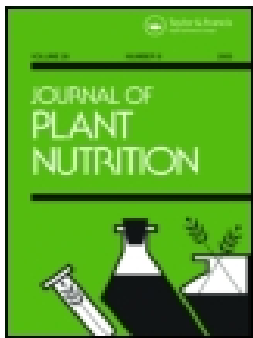


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Nano iron fertilization significantly increases tomato yield by increasing plants' vegetable growth and photosynthetic efficiency

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ABSTRACT

Nano fertilization is a precision approach to sustain economically viable and environmentally compatible crop production. The objective of our study was to evaluate the effects of different sources and rates of iron (Fe) fertilization as an essential micronutrient on the growth, physiological processes, and yield of greenhouse-grown tomatoes (*Solanum lycopersicum* cv. Bigdena F1). A factorial experiment in randomized complete block design with four replications was conducted using three sources (Conventional, FeCl₃·6H₂O [Conv-Fe]; Chelated with 6% Fe [Che-Fe]; and Nano Fe₂O₃ [Nano-Fe], alpha, 99%, 30–50 nm) and rates of Fe (0, 50, and 100 mg/kg soil) during 2015 and 2016 growing seasons. Averaged across Fe rates, the Nano-Fe significantly increased plant height, leaf number and area, shoot and root fresh and dry weights, and Soil Plant Analysis Development (SPAD) readings. Nano-Fe at 100 mg/kg significantly increased plant height, leaf number and area, fresh and dry weights of shoot and roots, gas exchange parameters i.e., photosynthesis (Pn), stomatal conductance (gs) and transpiration (E) rates, total yield, and yield components of tomatoes in comparison to the other rates. While there were no significant simple and interactions of Fe on plant physiological processes, the Nano-Fe had the highest Pn with an associated decrease in E and an increase in gs. Nano-Fe, when applied at 100 mg/kg, produced the highest fruit diameter, fruit numbers/plant, total fruit weight/plant, mean fruit weight/plant, total fruit numbers/ha, and total fruit weight/ha, followed by Nano-Fe at 50 mg/kg as compared with other Fe sources and rates. In conclusion, Nano-Fe fertilization is more effective than Conv-Fe and Che-Fe fertilizations across growing seasons to improve growth characteristics and metabolic processes of tomato plants, as Fe plays an important role in photosynthates accumulation and translocation. Nano-Fe increased tomato yield by 11% compared to Conv-Fe and Che-Fe fertilizers and will greatly improve farmers' profitability.

Abbreviations: Conv-Fe; Conventional iron; Che-Fe; Chelated iron; Nano-Fe; Nano iron; SPAD; Soil Plant Analysis Development; Pn; Photosynthesis : C; Stomatal conductance; E; Transpiration:

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Introduction

Iron is one of the micronutrients essential for all living organisms in terrestrial ecosystems Hochmuth (2011). Due to its oxidation-reduction properties, Fe plays a critical role in various physiological and biochemical pathways in plants such as DNA synthesis, respiration, and photosynthesis processes (Rout and Sahoo 2015). Fe also serves as a component of several vital enzyme cofactors that carry out electron and O₂ transport functions, facilitate chemical transitions, regulate protein stability, and is thus required for a wide range of biological functions (Rout and Sahoo 2015; Connorton, Balk, and Rodríguez-Celma 2017).

Iron is a major limiting nutrient for plant growth and metabolism, primarily due to its low solubility in aerobic environments (Zuo and Zhang 2011; Samaranayake, Bd, and Dssanayake 2012). An imbalance between the Fe availability in soil and its demand by the plant are the primary causes of Fe chlorosis of plants. While abundant in most soils, the ionic activity of Fe is low (more oxidized forms) because it forms insoluble Fe⁺³ compounds at neutral and higher pH levels in terrestrial ecosystems (Rout and Sahoo 2015).

Fe deficiency is widespread among many different crops. To alleviate Fe deficiency of plants, Fe application in conventional mixed fertilizers is still the most prevalent to improve crop yields especially in greenhouse production systems; however, Fe applied with conventional fertilizers are often ineffective with a low nutrient-use efficiency (Laurie et al. 1991; Connorton, Balk, and Rodríguez-Celma 2017). Chelated Fe (Che-Fe) has been recommended as an alternate approach for assuring increased Fe uptake by growing plants (Ronan 2007). The Che-Fe is an organic Fe salt, primarily prepared to make Fe soluble in water, making it more suitable for plant uptake. Currently, Che-Fe fertilization is one of the most popular methods for treating Fe chlorosis of plants. Che-Fe is quite popular because the compound has stabilized Fe ions, ideally preventing it from being oxidized and, in turn, precipitating in the soil solution. While the Che-Fe compounds are stable at a pH of below 6, almost 50% of the Fe becomes unavailable at levels above 6.5, which suggests that Che-Fe is also ineffective in high pH soils or alkaline growth media. Additionally, the Che-Fe has a high affinity for calcium that limits its use in soil, growth media, or water with higher calcium concentration ((Incesu et al. 2015).

Nanotechnology has been researched in agriculture, aiming to reduce the use of reactive chemicals, minimize nutrient losses, and increase economic crop yields by precision nutrient management practices (Lakshmi 2017). Fe nanoparticles are being investigated as a substitution for Conv-Fe and Che-Fe fertilizers. Several recent studies have reported that Fe nanotechnology is more effective in supplying Fe to plants, compared to the commonly used Fe fertilizers/chemicals in agriculture production systems (Li et al. 2013; Cheng et al. 2016; Connorton, Balk, and Rodríguez-Celma 2017; El-Desouky et al. 2018)

Nanoparticles are generally below 100 nm within the transition zone between individual molecules and the corresponding bulk materials, in which they exert both positive and negative effects on living cells (Nel et al. 2006). Substituting nano fertilizers for conventional fertilization is expected to release essential nutrients into the soil or growing media in a controlled way, thus preventing eutrophication and pollution of freshwater resources (Pramanik et al. 2020; Moaveni and Kheiri 2011). Karimi, Pourakbar, and Feizi (2014) reported that using Nano-Fe as foliar spraying increased bean growth and productivity. Likewise, (Pourjafar, Zahedi, and Sharghi 2016) indicated that Nano-Fe application increased canola growth, yield, and yield components. (Rizwan et al. 2019) stated that using Fe nanoparticles increased plant height, spike length, and dry weights of shoots, roots, spikes, and grains, when applied with N-P-K fertilizers. Despite all these potential advantages, the use of Nano-Fe in the agricultural sector is still relatively limited.

Our hypothesis is that Nano-Fe fertilization will support an economically viable and environmentally compatible greenhouse tomato production, compared to Conv-Fe and Che-Fe fertilizations. The objective of the research was to determine the effects of different rates of Nano-Fe

fertilization on the dynamics of plant growth, physiological processes, and yield of greenhouse-grown tomatoes with respect to the Conv-Fe and Che-Fe fertilizations.

Material and methods

A greenhouse experiment using tomato (cv. Bigdena F1) as a test plant was conducted at The Ohio State University South Centers, Piketon, Ohio, USA. Three types of Fe fertilizers: Che-Fe (Ethylene di-amine-N, N'-bis (2-hydroxyphenylacetic acid, EDDHA), Conv-Fe fertilizer ($\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$), and Nano-Fe (Fe_2O_3) particles were applied at three different levels (control [0], 50 and 100 mg/kg) to the commercial grow media in plastic bags 10 days following the transplant of tomato seedlings during the 2015–2016 and 2016–2017 growing seasons.

Plastic bags (30 cm wide x 45 cm tall) were filled with commercial grow media “Promix px” (5.44 kg), which consisted of Canadian sphagnum peat moss (75–85%), perlite horticultural grade, vermiculite horticultural grade, dolomitic and calcitic limestone (pH adjuster) wetting agent, and starter nutrients. The characteristics of the growing media were pH 4.7, EC 08 mS, and P 118, K 617, Ca 112, Mg 31, S 84, Fe 2.5, Mn, 3.7, Cu 1.5, Zn 0.7, B 3.1, Mo 0.3, and Al 6 mg/kg. The water quality parameters were pH 7.3, ECw 0.71 mS, and P 6.5, K36, Ca 208, Mg 24, S 20, Fe 7.7, Mn 1, Cu 1.6, Zn 1.4, B 0.2, Cl^- 0.3, and Al 13 mg/kg.

Seed germination and plant growth

In first week of October 2015 and July 2016, tomato seedlings were started in a growth chamber in a seed starter mix and then moved to the greenhouse early in the third week of October 2015 and early in the second week of July 2016. Late in the second week of November 2015 and during the first week of August 2016, randomly selected healthy tomato seedlings were transplanted to the growing media in the plastic bags. Five drip irrigation emitters were inserted to a depth of 10 cm in the growing media of each plastic bag. Irrigation was applied as required. Growing media temperature was measured throughout the season.

Temporal growth characteristics of tomato plants such as height, leaf numbers/plant, and leaf area/plant were measured. Leaf chlorophyll, as a measure of N content and uptake, was measured using a SPAD-502 Plus at several growth stages of the tomato plants. Fresh and dry weights of both shoots and roots at 70 days after transplantation (at harvest) were determined.

Tomato plant gas exchange parameters as physiological processes and yield attributes

Physiological processes, such as photosynthesis (P_n as $\mu\text{M}/\text{m}^2/\text{s}$), transpiration (E as $\text{mM}/\text{m}^2/\text{s}$), and stomatal conductance (g_s as $\text{mM}/\text{m}^2/\text{s}$) were measured on fresh leaves using a handheld Photosynthesis System CI-340 (CID Bio-Science, Inc., Camas, Washington, USA) at different growth stages of the tomato plants. Tomato fruit diameter, length, fruit number/plant, fruit weight/plant, mean fruit weight/plant, total fruit number/ha, and total fruit weight/ha were determined. Harvest index was calculated by dividing the economic yield (tomato fruits) with the total production (tomato biomass and yield).

Statistical analysis

The effects of independent variables (Fe types and rates) on growth and biomass production, SPAD, P_n , E , and C , and yield of tomatoes over time were statistically analyzed using SAS 9.3 (SAS 2010, SAS Institute, Cary, North Carolina, USA). The F-protected simple and interactive treatment means were separated by Tukey's significant difference test, when the ANOVA showed

Table 1. Effects of iron sources and doses on plant characteristics at maximum vegetative growth stage of tomatoes under greenhouse during 2015–2016 (first season).

Fe source	Fe-dose (mg/kg)	Plant ht. (cm)	Leaf no./plant	Leaf area/plant (cm ²)	SPAD reading
Conv-Fe		98.4c ^y	21bc	2253.5b	34.4b
Che-Fe		101.5b	22b	2506c	35.3a
Nano-Fe		105.3a	24a	2964.4a	35.4a
	0	98.3z ^e	23x	2147.6z	34.4y
	50	102.6y	23x	2664.9y	35.2x
	100	104.3x	23x	2911.4x	35.6x
Fe fertilizer x rate					
Conv-Fe	0	98.1*	21*	2158.5*	33.4 ns
	50	97.1	22	2260.6	34.8
	100	100	21	2341.3	34.9
Che-Fe	0	98.8	22	2123.4	34.8
	50	102.9	22	2545.6	35.3
	100	102.7	22	2849	35.9
Nano-Fe	0	98.0	21	2160.7	34.9
	50	107.8	25	3188.5	35.4
	100	110	27	3544	36

^yMeans separated by same lower-case letters in each column is not significantly different among iron sources at $p < 0.05$.

^eMeans separated by same lower-case letters in each column is not significantly different among iron doses at $p < 0.05$.

*indicates significant interaction between iron source x dose at $p < 0.05$. ns means Non-significant.

significant predictor effects on dependent variables at $p \leq 0.05$, unless otherwise mentioned. Regression and correlation analyses were performed by using SigmaPlot® (Systat Software Inc., San Jose, California).

Results and discussion

Tomato plant growth and biomass production

Fe fertilizer source and dose were shown to significantly affect tomato plant growth and biomass production during both years [Tables 1–6](#). The Nano-Fe significantly increased plant height, total leaf area, and SPAD readings, compared to Che-Fe and Conv-Fe fertilizers in the first (2015–2016) growing season. A similar response was observed in the second growing season (2016–2017), where Nano-Fe significantly increased plant height, leaf number/plant, SPAD readings, and total leaf area, followed by Che-Fe and Conv-Fe. The effect of 100 mg/kg Nano-Fe was more effective than other Fe rates. The Fe source and dose showed effects on tomato plant height and number of leaves in the first season, exhibiting significant interactions. In addition, the Fe types x rates exerted a significant interaction on tomato plants' SPAD readings and soil temperature in the second season only. During the first and second growing seasons, Nano-Fe at 100 mg/kg soil treatment produced the highest plant height, leaf number, and SPAD readings followed by Nano-Fe 50 mg/kg soil. The Fe source x dose clearly indicated a significant increase of tomato plant total leaf area/plant. Significant effects of Nano-Fe, especially at 100 mg/kg soil on total leaf area, were reflected on the efficiency of photosynthesis by accumulating more assimilates and high rates of their translocation. Thereby, significant increases in total fruit yields may be expected.

Regarding the combined growing seasons results, Nano-Fe source significantly increased plant height, number of leaves, and total chlorophyll (SPAD readings) compared to the Che-Fe and Conv-Fe sources. Higher growth of tomato plants was found at 100 mg/kg soil, followed by 50 mg/kg soil and then the control. Application of higher concentrations of Fe increased plant height, number of leaves, and total chlorophyll. The Fe source x dose showed significant interactions on tomato leaf number, total chlorophyll, and SPAD. The highest total leaf area existed with Nano-Fe, followed by Che-Fe and Conv-Fe fertilizations. The most effective dose to increase

Table 2. Effects of iron sources and doses on plant characteristics at maximum vegetative growth stage of tomatoes under greenhouse during 2016–2017 (second season).

Iron Source	Fe-dose (mg/kg)	Plant ht. (cm)	Leaf no. /plant	Leaf area /plant (cm ²)	SPAD reading
Conv-Fe		128.4c [¥]	26bc	2253.5b	45.3c
Che-Fe		132.8b	27b	2421.5b 49.0 b	
Nano-Fe		137.6a	29a	2964.4a	51.1a
	0	121.2y [€]	25y	2115.9z 42.7z	
	50	138.6x	28x	2617.9y 50.0 y	
	100	139.0x	29x	2837.7 × 52.7x	
Fe fertilizer x rate					
Conv-Fe	0	120.5*	22*	2104.9*	42.1*
	50	132.3	26	2273.8	46.4
	100	132.3	28	2320.4	47.4
Che-Fe	0	118.6	26	2105.8	43.8
	50	140.2	28	2430.5	50.3
	100	139.7	28	2728.4	52.8
Nano-Fe	0	124.4	26	2136.9	42.1
	50	143.3	30	3149.5	53.4
	100	145.2	31	3464.2	57.8

[¥]Means separated by same lower-case letters in each column is not significantly different among iron sources at $p < 0.05$.

[€]Means separated by same lower-case letters in each column is not significantly different among iron doses at $p < 0.05$.

*indicates significant interaction among iron source x dose at $p < 0.05$. ns means non-significant.

Table 3. Effects of iron sources and doses on plant characteristics at maximum vegetative growth stages of tomatoes under greenhouse during 2015–2017 (averaged across seasons).

Fe Source	Fe-dose (mg/kg)	Plant ht. (cm)	Leaf no. /plant	Leaf area /plant (cm ²)	SPAD reading
Conv-Fe		113.4c [¥]	23c	2243.3c	39.8c
Che-Fe		117.2b	25b	2463.8b	42.1b
Nano-Fe		123.7a	27a	2940.6a	43.3a
	0	112.0y [€]	23z	2131.7z	38.5z
	50	120.6x	25y	2641.4y	42.6y
	100	121.7x	26x	2874.5x	44.1x
Fe fertilizer x rate					
Conv-Fe	0	109.3 ^{ns}	22*	2131.7*	37.8*
	50	114.7	24	2267.2	40.6
	100	116.2	24	2330.9	41.2
Che-Fe	0	108.7	24	2114.6	39.3
	50	121.6	25	2488.0	42.8
	100	121.2	25	2788.7	44.3
Nano-Fe	0	118.0	24	2148.8	38.5
	50	125.6	27	3169.0	44.4
	100	127.6	29	3504.1	46.9

[¥]Means separated by same lower-case letters in each column is not significantly different among iron sources at $p < 0.05$.

[€]Means separated by same lower-case letters in each column is not significantly different among iron doses at $p < 0.05$.

*indicates significant interaction among iron source x dose at $p < 0.05$. ns means Non-significant.

total leaf area was 100 mg/kg soil, followed by 50 mg/kg soil and the control. Also, results showed that total leaf area increased as Fe concentration increased. In other words, there is a parallel increase between Fe concentration and the leaf area obtained. The Fe source x dose significantly increased the total leaf area/plant.

The data presented in Tables 5–7 showed the effects of Fe sources and dose on biomass production, especially root and shoot growth characteristics in both the 2015–2016 and 2016–2017 growing seasons. Nano-Fe had the highest shoot and root dry weights, followed by Che-Fe, while Conv-Fe recorded the lowest shoot and root dry weights. However, the Conv-Fe fertilization significantly increased the shoot: root in the first season while there were no significant differences between sources in the second season. For Fe rates, in the first season, 50 mg/kg gave the highest shoot fresh weight, meanwhile, 100 mg/kg gave the highest shoot dry weight and both root fresh

Table 4. Effects of iron sources and doses on shoot and root weights of tomato plants at 70 days after transplanting (harvest) under greenhouse during 2015–2016 (first season).

Fe Source	Fe-dose (mg/kg)	Root fresh wt. (Mg/ha)	Root dry wt. (Mg/ha)	Shoot fresh wt. (Mg/ha)	Shoot dry wt. (Mg/ha)	Shoot/ root
Conv-Fe		2.0b [¥]	0.4b	6.3ab	1.2a	2.9a
Che-Fe		2.2b	0.5ab	6.2b	1.2a	2.6bc
Nano-Fe		2.7a	0.6a	6.5a	1.4a	2.4c
	0	1.9y [€]	0.4y	6.3y	1.1y	3.0x
	50	2.4xy	0.5xy	6.6x	1.4x	2.6y
	100	2.6x	0.6x	6.0z	1.4x	2.3z
Fe fertilizer type x rate						
Conv-Fe	0	1.8 ^{ns}	0.3 ^{ns}	5.6*	1.1 ^{ns}	3.2*
	50	2.3	0.6	6.4	1.3	2.4
	100	2.4	0.6	6.5	1.3	2.2
Che-Fe	0	1.8	0.4	6.6	1.1	3.0
	50	2.2	0.4	6.7	1.3	3.0
	100	2.2	0.5	5.7	1.2	2.6
Nano-Fe	0	2.1	0.4	6.6	1.1	3.0
	50	2.8	0.6	6.9	1.5	2.3
	100	3.2	0.8	5.8	1.6	2.0

[¥]Means separated by same lower-case letters in each column is not significantly different among iron sources at $p < 0.05$.

[€]Means separated by same lower-case letters in each column is not significantly different among iron doses at $p < 0.05$.

* indicates significant interaction between iron source x dose at $p < 0.05$. ns means non-significant.

Table 5. Effects of iron sources and doses on shoot and root weights of tomato plants at 70 days after transplanting (harvest) under greenhouse during 2016–2017 (second season).

Fe Source	Fe-dose (mg/kg)	Root fresh wt. (Mg/ha)	Root dry wt. (Mg/ha)	Shoot fresh wt. (Mg/ha)	Shoot dry wt. (Mg/ha)	Shoot/ root
Conv-Fe		1.2a [¥]	0.4a	4.4b	0.8a	2.2a
Che-Fe		1.2a	0.4a	4.3b	0.8a	2.1a
Nano-Fe		1.3a	0.5a	5.2a	1.0a	2.0a
	0	1.1x [€]	0.3x	3.7y	0.7y	2.0x
	50	1.2x	0.4x	5.0x	0.9xy	2.1x
	100	1.3x	0.5x	5.3x	1.0x	2.1x
Fe fertilizer type x rate						
Conv-Fe	0	1.1 ^{ns}	0.3 ^{ns}	4.1*	0.8*	2.2*
	50	1.2	0.4	4.6	0.9	2.2
	100	1.2	0.4	4.6	0.8	2.1
Che-Fe	0	1.1	0.4	3.5	0.7	2.0
	50	1.2	0.4	4.4	0.8	2.0
	100	1.2	0.5	5.0	1.0	2.2
Nano-Fe	0	1.0	0.4	3.6	0.7	1.8
	50	1.3	0.5	5.9	1.1	2.1
	100	1.6	0.6	6.2	1.2	2.1

[¥]Means separated by same lower-case letters in each column is not significantly different among iron sources at $p < 0.05$.

[€]Means separated by same lower-case letters in each column is not significantly different among iron doses at $p < 0.05$.

*indicates significant interaction among iron source x dose at $p < 0.05$. ns means non-significant.

and dry weights. In the second season, 100 mg/kg gave the highest shoot fresh weight, shoot dry weight, and root fresh and dry weights. The root fresh weight did not show significant difference in response to the Fe source x dose in the first and second growing seasons, however, it is important to mention that Nano-Fe recorded the highest values at 100 mg/kg, but the shoot fresh weight showed a significant difference by Fe source x dose in the first and second seasons. The Fe source x dose clearly showed a synergistic effect to increase tomato root and shoot dry weights, and the highest shoot and root dry weights were recorded when Nano-Fe was applied 100 mg/kg soil.

The significant effects of Nano-Fe may be due to the synergistic roles played by Nano-Fe particles on increasing total chlorophyll, photosynthesis, number of leaves, plant height and biomass production than that of the control. In the case of combined growing seasons, among the Fe sources, the shoot fresh weight had the highest value with the treatment of Nano-Fe, followed by

Table 6. Effects of iron sources and doses on shoot and root weights of tomato plants at 70 days after transplanting (harvest under greenhouse during 2015–2017 (averaged across seasons).

Fe Source	Fe-dose (mg/kg)	Root fresh wt. (Mg/ha)	Root dry wt. (Mg/ha)	Shoot fresh wt. (Mg/ha)	Shoot dry wt. (Mg/ha)	Shoot/root
Conv-Fe		1.6b ^y	0.4a	5.4b	1.0a	2.5a
Che-Fe		1.7b	0.5a	5.2b	1.0a	2.3ab
Nano-Fe		2.0a	0.5a	5.8a	1.2a	2.2b
	0	1.5y [€]	0.4x	5.0y	0.9y	2.5x
	50	1.8xy	0.5x	5.8x	1.1xy	2.3xy
	100	2.0x	0.5x	5.7x	1.2x	2.2y
Fe fertilizer type x rate						
Conv-Fe	0	1.5 ^{ns}	0.3 ^{ns}	4.9*	0.9 ^{ns}	2.7*
	50	1.7	0.5	5.5	1.1	2.3
	100	1.8	0.5	5.6	1.1	2.2
Che-Fe	0	1.4	0.4	5.0	0.9	2.5
	50	1.7	0.4	5.5	1.1	2.5
	100	1.7	0.5	5.3	1.1	2.4
Nano-Fe	0	1.5	0.4	5.1	0.9	2.4
	50	2.1	0.6	6.4	1.3	2.2
	100	2.4	0.7	6.0	1.4	2.0

^yMeans separated by same lower-case letters in each column is not significantly different among iron sources at $p < 0.05$.[€]Means separated by same lower-case letters in each column is not significantly different among iron doses at $p < 0.05$.*indicates significant interaction among iron source x dose at $p < 0.05$. ns means non-significant.**Table 7.** Effects of iron sources and doses on tomatoes plant photosynthesis (Pn), transpiration (E), and stomatal conductance (C) under greenhouse conditions during 2015–2016 (first season).

Fe Source	Fe-Dose (mg/kg)	Photosynthesis $\mu\text{mol}/\text{m}^2/\text{s}$	Transpiration $\text{mmol}/\text{m}^2/\text{s}$	Stomatal Conductance $\text{mmol}/\text{m}^2/\text{s}$
Conv-Fe		6.5b ^y	2.8a	546.7a
Che-Fe		5.4b	2.2a	482.7a
Nano-Fe		6.9a	2.2a	457.9a
	0	5.5y [€]	2.4x	466.3x
	50	7.2x	2.4x	476.3x
	100	6.1y	2.4x	544.7x
Fe fertilizer type x rate				
Conv-Fe	0	6.5*	2.7 ^{ns}	467.9 ^{ns}
	50	6.4	2.8	577.1
	100	5.3	2.1	519.3
Che-Fe	0	5.0	2.2	480.9
	50	6.0	2.2	453.8
	100	5.4	2.1	516.9
Nano-Fe	0	4.7	2.1	445.3
	50	9.7	2.3	406.3
	100	6.2	2.3	522.2

^yMeans separated by same lower-case letters in each column is not significantly different among iron sources at $p < 0.05$.[€]Means separated by same lower-case letters in each column is not significantly different among iron doses at $p < 0.05$.*indicates significant interaction among iron source x dose at $p < 0.05$. ns means non-significant.

Conv-Fe and then Che-Fe; the highest value of shoot dry weight and root fresh and dry weights were recorded with the Nano-Fe, followed by Che-Fe and Conv-Fe, respectively. Averaged across seasons, there was no significant differences between Conv-Fe and Che-Fe treatments on shoot: root and the same between Che-Fe and Nano-Fe. Averaged across sources, Fe levels significantly affected the shoot fresh and dry weights and root fresh weight of tomato where Fe was applied at 50 and 100 mg/kg, respectively, and showed a significant difference in comparison to the control level. Root dry weight was increased by the Fe when applied at 100 mg/kg and 50 mg/kg Fe compared to the control. The Fe source x dose showed that the increase of shoot dry weight and root

fresh and dry weights with Nano-Fe was likely due to the increases in higher leaf areas and enhanced photosynthetic capacity.

The effectiveness of nanoparticle treatment may be caused by providing more availability of Fe for the plants, as when the insoluble ferric (Fe^{+3}) form is reduced, it is converted to a ferrous (Fe^{+2}) form in the soil and is then absorbed by plants. The balance between the solubility of Fe in soil and the demand for Fe by the plant could be a main factor in maintaining a healthy plant and higher tomato yields. In contrast, Fe deficiency induces chlorosis, which leads to reduced yield and fruit quality. This is because Fe is essential for the biosynthesis of cytochromes and other heme molecules, including chlorophyll, the electron transport system, and the construction of Fe-S clusters (Briat, Curie, and Gaymard 2007; Hänsch and Mendel 2009). The increase of plant height, leaf numbers, and growth characteristics could be attributed to the positive effect of Nano-Fe on increasing photosynthetic rates and other metabolic activities leading to an increase in various plant metabolites responsible for cell division and elongation Hochmuth (2011). That it could be preceded with a positive alteration in the hormonal profile especially the promoter's ones, i.e., auxins, gibberellins and cytokinin's, as well.

A significant variation in growth, physiological processes, and yield (as affected by Fe sources) is due to the variable effects of Fe on tomato plants. The positive effects of Fe, such as an increase in stem length on spinach plant, were due to uptake of Fe_2O_3 nanoparticles (Jeyasubramanian et al. 2016). Such results agree with those obtained by (Shankramma et al. 2016). (Alireza et al. 2012), who reported that spinach leaf area index and growth rates indicated that Nano-Fe has a positive effect on all plant growth traits. These results are consistent with those reported by other studies (El Feky et al. 2013; Shankramma et al. 2016). (Alireza et al. 2012) reported similar positive effects of Nano-Fe on fresh and dry weights of spinach. Sharifi (2016) illustrated that the use of Fe nanoparticles increased corn total dry biomass El-Desouky et al. (2018) indicated that cucumber yield was applied of Nano Fe at 50 and 100 mg/l.

Tomato plant gas exchange parameters (physiological processes)

The results presented in Tables 8–10 show that Fe source and dose influenced the physiological processes of tomato plants during 2015–2016 (first season). Among the Fe sources, Nano-Fe significantly increased photosynthesis (Pn) rates; however, the Pn rates did not vary between Che-Fe and Conv-Fe sources, meanwhile transpiration (E) and stomatal conductivity (gs) did not vary among Fe sources. In contrast, Fe doses significantly affected the Pn of tomato plants, where 50 mg Fe/kg soil showed a significant response over the 100 mg/kg and control levels. The Fe source x dose showed a significant interaction of Pn rates. The Nano-Fe fertilization at 50 mg/kg had the highest Pn rates with decreased E rates, compared to other treatment combinations. During 2016–2017 (second season), Nano-Fe source significantly increased tomato Pn rates, followed by Che-Fe and Conv-Fe, while the E and C rates showed the same response to the different Fe sources. The Fe level of 100 mg/kg was the most effective. The Fe source x dose did not show significant interactions on tomato gas exchange parameters i.e., physiological processes (i.e., Pn, E, and gs rates); however, Nano-Fe at 100 mg/kg gave the highest Pn rates with decreased E rates associated with increased gs rates. Significant effects of Nano-Fe, especially at 50 mg/kg, that increased Pn rates with reduced E rates were due to beneficial effects of Nano-Fe particles at dose balance optimization. Nano-Fe, at 50 mg/kg, was expected to act as a catalyst to enhance Pn activities compared to other treatments. An increase in Pn rates was expected to increase the water-use efficiency of plants (Osmond, Björkman, and Anderson 2012). Previous studies have shown a similar response of Nano-Fe on diverse crops in this respect (Kazemi 2013; Mer and Ama 2014; Karimi, Pourakbar, and Feizi 2014). The present results agreed with those obtained by Kazemi (2013), who showed that foliar application of Che-Fe at 50 and 100 mg/kg significantly increased Pn and gs rates of cucumber plants. Fe was promoting too, and decreased E rates. Regarding

Table 8. Effects of iron sources and doses on tomatoes plant photosynthesis (Pn), transpiration (E), and stomatal conductance (C) under greenhouse conditions during 2016–2017 (second season).

Fe Source	Fe-Dose (mg/kg)	Photosynthesis $\mu\text{mol}/\text{m}^2/\text{s}$	Transpiration $\text{mmol}/\text{m}^2/\text{s}$	Stomatal Conductance $\text{mmol}/\text{m}^2/\text{s}$
Conv-Fe		7.6b ^y	3.2a	627.0a
Che-Fe		8.3b	3.2a	638.6a
Nano-Fe		9.5a	3.3a	630.4a
	0	7.8y [€]	3.2x	642.3x
	50	7.9y	3.2x	609.7x
	100	9.7x	3.3x	644.1x
Fe fertilizer type x rate				
Conv-Fe	0	6.8 ns	3.1 ns	634.2 ns
	50	7.8	3.4	686.6
	100	9.1	3.4	650.7
Che-Fe	0	7.8	3.1	625.7
	50	8.7	3.3	634.3
	100	8.3	3.3	674.1
Nano-Fe	0	9.3	3.4	677.4
	50	8.0	3.3	573.1
	100	11.7	3.2	634.5

^yMeans separated by same lower-case letters in each column is not significantly different among iron sources at $p < 0.05$.

[€]Means separated by same lower-case letters in each column is not significantly different among iron doses at $p < 0.05$.

*indicates significant interaction among iron source x dose at $p < 0.05$. ns means Non-Significant.

2015–2017 (combined) growing seasons, the highest Pn rates were recorded with Nano-Fe source. Also, the 100 mg/kg dose gave the highest Pn with an increase in gs rates. Nanoparticles improve Pn rates in tomatoes by improving carbonic anhydrase activity and synthesis of photosynthetic pigments (Siddiqui and Al-Whaibi 2014). Also, (Liu et al. 2005) reported that Nano-Fe increased Pn and gs rates in peanut plants. Generally, Fe is essential for many important compounds and physiological processes in plants. Although Fe is required by plants in small concentrations, Fe is involved in chlorophyll synthesis, and it is necessary for certain enzyme functions. Fe required in chlorophyll synthesis is the reason for the chlorosis associated with Fe deficiency (Hochmuth 2011).

Iron is involved in the biosynthesis of chlorophylls, and it is critical for the preservation of chloroplast structure and function, approximately 80% of Fe is found in photosynthetic cells anywhere it is crucial for the biosynthesis of cytochromes and other heme molecules, including chlorophyll, the electron transport system. Iron has big role for porphyrin construction of chlorophyll, and is therefore a main component of chloroplastides. Also, iron is straight intricate in the photosynthetic activity of plants and, consequently, their productivity Briat, Curie, and Gaymard (2007). Through evidence so far that nutrient accumulation induced by Nano-Fe is related with H⁺-ATPase Biosynthesis, improved photosynthesis process by PM H⁺-ATPase causes an accumulation of sugar in plants Okumura et al. (2016). Therefore, in addition to the present mechanisms that cause increased plant growth by nano-Fe by increasing bioavailable Fe, plant hormones and antioxidant enzymes. plant Using of nano-fertilizer induces the slow release of nutrients from nanoparticles, which increase the uptake efficiency of plants Achari and Kowshik (2018) and Yoon et al. (2019).

In this respect Fe helpful in changing stomatal conductance, transpiration rate and net CO₂ fixation rate. plants by optimize the allocation of resources in order to reserve a balance between enzymatic (i.e., Rubisco) and light harvesting (i.e., Chl) capabilities across a wide range of light and nutrient regimes. When Fe-increase in leaves such a balance may form part of an adaptive

Table 9. Effects of iron sources and doses on tomatoes plant photosynthesis (Pn), transpiration (E), and stomatal conductance (C) under greenhouse conditions during 2015–2017 (averaged across seasons).

Fe Source	Fe-Dose (mg/kg)	Photosynthesis $\mu\text{mol}/\text{m}^2/\text{s}$	Transpiration $\text{mmol}/\text{m}^2/\text{s}$	Stomatal Conductance $\text{mmol}/\text{m}^2/\text{s}$
Conv-Fe		7.2b ^y	3.1a	600.2a
Che-Fe		7.3b	2.9a	586.7a
Nano-Fe		8.6a	2.9a	572.9a
	0	7.0y ^ε	2.9x	583.6x
	50	7.6x-y	2.9x	565.2x
	100	8.5x	3.0x	610.9x
Fe fertilizer type x rate				
Conv-Fe	0	6.7 ns	3.0 ns	570.8 ns
	50	7.4	3.2	652.1
	100	8.1	3.1	616.1
Che-Fe	0	6.9	2.9	581.6
	50	7.9	2.9	579.4
	100	7.3	2.9	619.4
Nano-Fe	0	7.6	2.9	593.0
	50	8.5	2.9	517.5
	100	9.9	2.9	597.1

^yMeans separated by same lower-case letters in each column is not significantly different among iron sources at $p < 0.05$.

^εMeans separated by same lower-case letters in each column is not significantly different among iron doses at $p < 0.05$. ns means non-significant.

Table 10. Effects of iron sources and doses on tomato yield characteristics under greenhouse during 2015–2016 (first season).

Fe Source	Fe-dose (mg/kg)	Fruit no. /plant	Fruit wt. /plant (kg)	Mean fruit wt. (kg)	Fruit dia. (cm)	Total fruit wt. (ton/ha)
Conv-Fe		33ab ^y	8.2c	0.3a	3.9b	29c
Chel- Fe		32b	10.1b	0.3a	4.1ab	34.4b
Nano-Fe		35a	15.1a	0.4a	4.2a	51a
	0	32x ^ε	6.6y	0.2y	3.7y	22.6z
	50	35x	13.1x	0.4x	4.2x	44.5y
	100	34x	14.0x	0.4x	4.3x	47.7x
Fe fertilizer x rate						
Conv-Fe	0	33 ^{ns}	6.5*	0.2*	3.7*	22.2*
	50	36	10	0.3	4	34.1
	100	31	9.1	0.3	4.1	30.8
Che-Fe	0	33	7.4	0.2	3.6	25
	50	30	11.1	0.4	4.4	37.6
	100	32	12	0.4	4.5	40.7
Nano-Fe	0	29	6	0.2	3.7	20.4
	50	39	18.2	0.5	4.4	61.9
	100	39	21.1	0.5	4.5	71.6

^yMeans separated by same lower-case letters in each column is not significantly different among iron sources at $p < 0.05$.

^εMeans separated by same lower-case letters in each column is not significantly different among iron doses at $p < 0.05$.

*indicates significant interaction among iron source x dose at $p < 0.05$. ns means non-significant.

mechanism to enable plants to increased stomatal conductance and survive more CO_2 and increasing photosynthesis efficiency. Thereby, net assimilation rate will be increase, Larbi et al. (2006).

Tomato yield attributes

The data in Tables 11–12 show that in both seasons, Nano-Fe recorded the highest fruit diameter, total fruit weight/plant, mean fruit weight/plant, and total fruit weight/ha, followed by Che-Fe and Conv-Fe. There was no significant difference among the effects of Fe sources on fruit numbers; however, Nano-Fe gave the highest fruit number/plant. The Fe levels affected tomato yield

Table 11. Effects of iron sources and doses on tomato yield characteristics under greenhouse during 2016–2017 (second season).

Fe Source	Fe-dose (mg/kg)	Fruit no. /plant	Fruit wt. /plant (kg)	Mean fruit wt. (kg)	Fruit dia. (cm)	Total fruit wt. (ton/ha)
Conv-Fe		29b ^y	7.8c	0.3b	4.2b	26.5c
Che-Fe		30b	9.2b	0.3b	4.3ab	31.2b
Nano-Fe		34a	15.0a	0.4a	4.7a	50.9a
	0	27y ^e	6.9y	0.3y	4.2y	23.5z
	50	33x	12.0x	0.4x	4.4xy	40.7y
	100	33x	13.1x	0.4x	4.6x	44.4x
Fe fertilizer x rate						
Conv-Fe	0	27.5*	6.9*	0.3*	4.1 ^{ns}	23.6*
	50	30	8.2	0.3	4.30	28
	100	29	8.2	0.3	4.30	27.9
Che-Fe	0	30	7.5	0.2	4.10	25.6
	50	35	11.1	0.3	4.40	37.6
	100	26	8.9	0.3	4.40	30.3
Nano-Fe	0	24	6.3	0.3	4.3	21.2
	50	35	16.7	0.5	4.7	56.6
	100	43	22.1	0.5	5	75

^yMeans separated by same lower-case letters in each column is not significantly different among iron sources at $p < 0.05$.

^eMeans separated by same lower-case letters in each column is not significantly different among iron doses at $p < 0.05$.

*indicates significant interaction among iron source x dose at $p < 0.05$. ns means non-significant.

characteristics in both seasons, while 100 mg Fe/kg gave the highest fruit diameter, total fruit weight/plant, mean fruit weight/plant, and total fruit weight/ha, followed by 50 mg Fe/kg and the control. The 50 mg Fe/kg gave the highest fruit number/plant in both seasons. The Fe source x dose exerted significant interactions of tomato yield characteristics in both seasons, except the fruit number/plant; however, it did not show any significant interactions in the first season. The fruit diameter was not influenced by Fe source x dose in the second season. Nano-Fe, when applied at 100 mg/kg, had produced the highest total fruit weight/plant, fruit mean weight/plant, fruit diameter, and total fruit weight/ha followed by Nano-Fe, when applied at 50 mg/kg, in the first season.

Nano-Fe, when applied at 50 mg/kg, produced the highest fruit number/plant in the first season. In the case of 2015–2017 (combined) growing seasons, Nano-Fe showed increase in total fruit weight/ha that equals (meets) about 84% more than in Conv-Fe. For Fe levels, 100 mg/kg gave the highest tomato fruit diameter, total fruit weight/plant, mean fruit weight/plant, and total fruit weight/ha, followed by 50 mg/kg, while the control ranked last in this respect. Also, 50 mg/kg gave the highest tomato fruit number/plant, followed by 100 mg/kg and the control. In addition, Fe source x dose has shown that Nano-Fe, at 100 mg/kg, had the highest fruit diameter (4.8 cm), tomatoes fruit number/plant (41), total fruit weight/plant (21.6 kg), Mean fruit weight/plant (0.5 kg) and total fruit weight/ha (73.3), followed by Nano-Fe applied at 50 mg/kg.

Figure 1 showed that tomato harvest index (HI) significantly increased with Nano-Fe when compared to Conv-Fe and Che-Fe during 2015–2016, 2016–2017, and averaged across seasons. Regarding Fe levels, 100 mg/kg had the highest values when compared with 50 mg/kg and the control at harvest in the first, second, and combined seasons. Also, dry biomass increased in 2016–2017 compared to 2015/2016. The HI represents the efficiency of using both natural resources and anthropogenic inputs (water, CO₂, soil, and inputs, especially fertilizers) to produce a harvestable product; for that reason, it is important both from an economic and ecological aspect (Gur et al. 2010).

The increase of fruit number/plant, mean fruit diameter, total fruit weight/plant and mean fruit weight with Nano-Fe treatment could be attributed to the role of Fe nanoparticles to provide more soluble Fe. This leads to more surface area for the metabolic reactions of the plant and an increase in Pn rates, dry-matter and yield because Fe acts as an electron carrier in respiration and photosynthesis, in the production and detoxification of free O₂ radicals, O₂ transport, and

Table 12. Effects of iron sources and doses on tomato yield characteristics under greenhouse during 2015–2017 (combined seasons).

Fe Source	Fe-dose (mg/kg)	Fruit no. /plant	Fruit wt. /plant (kg)	Mean fruit wt. (kg)	Mean fruit dia. (cm)	Total fruit yield (ton/ha)
Conv-Fe		31a [*]	8.2c	0.3b	4.1a	27.8c
Che-Fe		31a	9.7b	0.3b	4.2a	32.8b
Nano-Fe		34a	15.1a	0.4a	4.4a	51.1a
	0	29y ^ε	6.8y	0.2y	3.9y	23y
	50	34x	12.6x	0.4x	4.3x	42.6x
	100	33x	13.6x	0.4x	4.5x	46x
Fe fertilizer x rate						
Conv-Fe	0	30*	6.7*	0.2*	3.9*	22.9*
	50	33	9.1	0.3	4.1	31.1
	100	30	8.6	0.3	4.2	29.3
Che-Fe	0	32	7.4	0.2	3.8	25.3
	50	32	11.1	0.3	4.4	37.6
	100	29	10.5	0.4	4.4	35.5
Nano-Fe	0	26	6.1	0.2	4	20.8
	50	37	17.4	0.5	4.5	59.2
	100	41	21.6	0.5	4.8	73.3

^{*}Means separated by same lower-case letters in each column is not significantly different among iron sources at $p < 0.05$.

^εMeans separated by same lower-case letters in each column is not significantly different among iron doses at $p < 0.05$.

*indicates significant interaction among iron source x dose at $p < 0.05$. ns means non-significant.

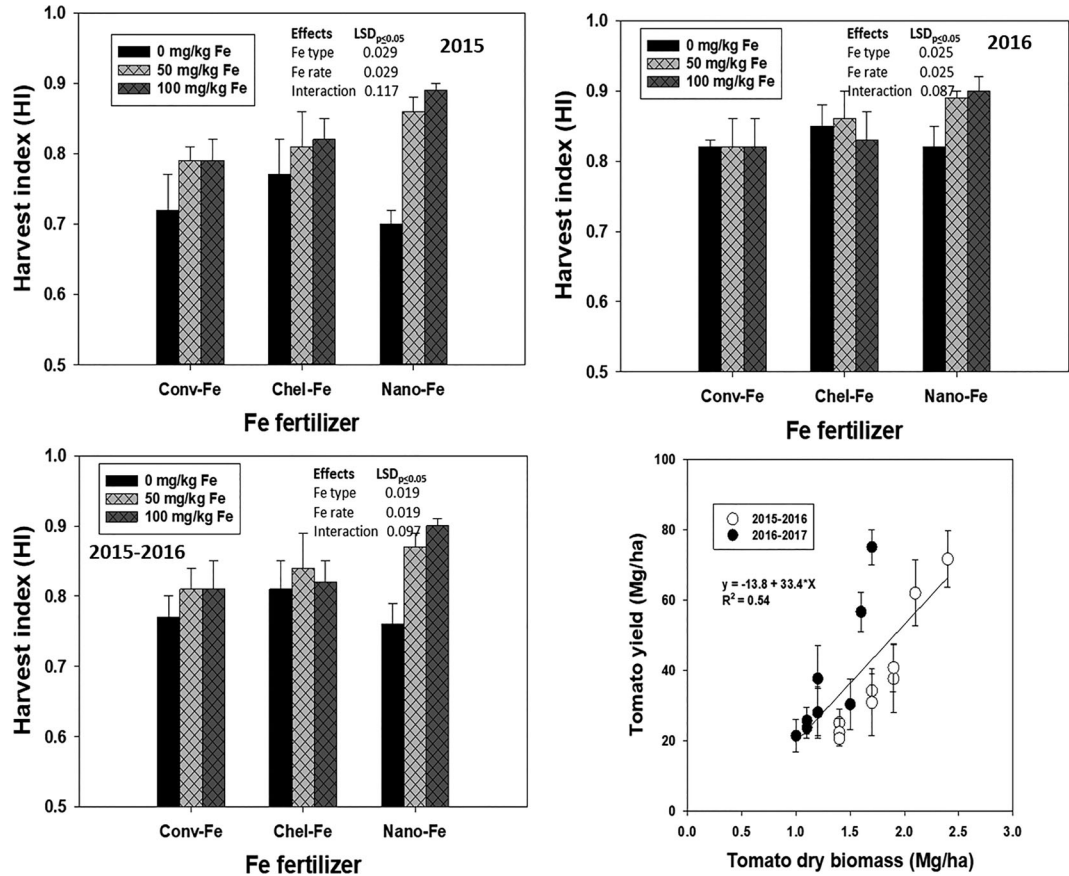


Figure 1. Effects of iron fertilizer types and rates on tomato harvest index (HI) and relationship between tomato dry biomass (shoot and root) and fruit yield (2015 to 2017 growing seasons).

reduction and mono oxygenase reactions. (Nadi, Ayneband, and Mojaddam 2013) reported that Nano-Fe, when applied in optimum concentrations, had a positive and significant effect on fava bean yield; the same Fe level effect on the previous yield characteristics was observed in the second season, in addition to the fruit number/plant and number/ha. These results show that Nano-Fe fertilizers are more efficient or effective than Che-Fe and Conv-Fe fertilizers, as they improved plant growth and increased metabolic efficiency such as photosynthesis, which leads to higher photosynthates accumulation and translocation to the plant economic parts (tomatoes fruit) and total yield. Therefore, selection of the proper concentration of nano source is important for getting higher benefits. In this respect, several previous studies have reported a similar response on diverse crops by different Fe sources (Sheykhabglou et al. 2010; Nadi, Ayneband, and Mojaddam 2013; Alireza, Alireza, and Elham 2015).

Conclusions

Results from our two-year study have shown that Fe fertilization using Nano-Fe, Che-Fe, and Conv-Fe had significantly increased tomato growth and yield. Among the Fe sources, Nano-Fe improved tomato physiological processes and growth, compared to Conv-Fe and Che-Fe fertilization. Nano-Fe was more advantageous than Conv-Fe and Che-Fe fertilizers, as they economically increased tomato yield and yield attributes.

Conflict of interest

There were no competing financial interests or personal relationships.

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