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The influence of iron chelate and zinc sulfate on the growth and nutrient composition of chickpea grown on a calcareous soil

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ABSTRACT- The effects of iron (Fe) and zinc (Zn) treatments on the growth and nutrient composition of chickpea were studied in a greenhouse experiment arranged in a completely randomized design. While the application of Fe decreased mean shoot dry weight of chickpea, that of Zn had no significant effect on chickpea shoot dry weight. Increasing Fe levels drastically decreased Mn concentration and uptake in chickpea shoot. Addition of Zn, however, had no significant effect on Mn concentration and uptake in chickpea shoot. Although the addition of 10 mg Fe kg⁻¹ significantly increased mean Fe concentration in chickpea shoot, the effect of Fe application on mean Fe uptake was negligible. Although the application of Zn had no significant effect on mean Fe concentration or uptake, it increased mean Zn uptake in chickpea shoot. While the application of Fe increased mean Zn concentration, application of 5 mg Fe kg⁻¹ had no significant effects on mean Cu concentration or uptake in chickpea tissues although 10 mg Fe kg⁻¹ increased mean Cu concentration and uptake in chickpea tissues. Addition of Zn, however, had no significant effect on Cu uptake. A significant negative correlation was observed between Mn uptake and Fe rates demonstrating a reduction in shoot Mn uptake following Fe application. Although shoot dry weight of chickpea was negatively correlated with Fe rates, it showed significant positive correlations with the uptakes of Zn and Mn indicating that the patterns of changes in dry matter was in coordination with changes in Zn and Mn uptakes. Since Fe chelate addition may cause nutrient imbalance and growth reduction in chickpea, it appears that the use of Fe and Zn efficient genotypes should be considered as an appropriate practice for chickpea grown on calcareous soils low in available Fe and Zn.

INTRODUCTION

Iron (Fe) deficiency is a constraint for crop grown on calcareous soils (Ghasemi Fasaei et al., 2003). Fe deficiency can cause yield losses in bean cultivars in soils low in available Fe (Zaiter et al., 1992). High level of bicarbonate is usually the most common cause of Fe deficiency in calcareous soils (Pestana et al., 2005).

Iron chelate (Fe-EDDHA) is one of the most effective Fe sources which has been widely used in correcting Fe deficiency (Mortvedt, 1991). Application of Fe fertilizers may be futile in increasing crop growth and yield since it may cause nutritional disorders throughout antagonistic effects of Fe with other metal micronutrients. Ghasemi Fasaei et al. (2003, 2005) reported that application of Fe deceased Mn contents of soybean genotypes and chickpea due to the presence of antagonistic relationships among these nutrients. It appears that Fe interferes with Mn translocation from root to shoot. Roomizadeh and Karimian (1996) reported that application of Fe either had no significant effect on dry matter of soybean or decreased it. They concluded that the negative effect of Fe application was attributed to the interference of Fe with Mn. Also, Alam et al. (2000) reported that Fe and Mn competed for the same transport sites.

Zinc (Zn) deficiency may be observed in calcareous soils. This element can be toxic under high concentrations (Nan et al., 2002). The main reason for Zn deficiency is low availability of this metal micronutrient to plant roots rather than low content of this nutrient in soils (Kalayci et al., 1999).

Ghasemi Fasaei and Ronaghi (2008) reported that application of 8 mg Fe Kg⁻¹ as Fe-EDDHA decreased mean Zn uptake in wheat grown on a calcareous soil by 31.9%. Interactions between Fe and Zn have also been reported by Verma and Tripathi (1983) for submerged paddy and by Kaya et al. (1999) for two tomato cultivars. Zhao et al. (2011) studied the effect of Fe and Zn on micronutrient levels in wheat in a nutrient solution experiment and observed that Zn, Cu and Mn concentrations were negatively correlated with leaf Fe concentrations. They reported that application of 5 mg Fe L⁻¹ reduced the concentration of Zn, Cu and Mn in wheat by 49%, 34% and 56%, respectively. They also observed that application of 10 mg Zn L⁻¹ decreased Fe concentration by about 8%, but had no significant effect on the concentration of Fe or Cu.

Ghasemi Fasaei and Ronaghi (2008) observed that soil application of 8 mg Fe Kg^{-1} as Fe-EDDHA increased mean Fe: Zn ratio by 3.2-fold. Foliar

application of a 2% Fe sulfate increased mean Fe:Zn ratio by 8.5-fold. Increased micronutrients deficiency over recent decades is a result of a decrease in the quality of poor people's diets (Graham et al., 2001). Legumes are a source of many minerals including Fe and Zn that are found only in low amounts in the cereals or root crops (Wang et al., 2003).

The main objective of the present study was to investigate the influence of the levels of Fe and Zn on growth and nutrient composition of chickpea grown on a calcareous soil low in available Zn and Fe.

MATERIALS AND METHODS

A greenhouse experiment was conducted on soil low in iron (Fe) and zinc (Zn). The soil was collected from Chitgar series (Fine- loamy, carbonatic, thermic, Typic Calcixerepts) located in Sarvestan area, Fars province,*: Iran. The studied soil was a calcareous soil with a pH of 7.8, electrical conductivity (ECe) of 1 dS m⁻¹, calcium carbonate equivalent (CCE) of 60%, organic matter (OM) of 1%, and sodium bicarbonate extractable phosphorus (P) of 15 mg kg⁻¹. The values of DTPA extractable Fe, Zn, Cu, and Mn were 2.5, 1, 1 and 7 mg kg⁻¹. The experiment was a 3×3 factorial arranged in a completely randomized design with three replications.

Treatments consisted of three levels of Zn (0, 5, and 10 mg Zn kg⁻¹ as Zn-sulfate) and three levels of Fe (0, 5, and 10 mg Fe kg⁻¹ as Fe-EDDHA). Three kg of soil was placed in plastic pots and watered with distilled water to field capacity and maintained at this moisture level throughout the experiment by watering the pots to a constant weight. Nitrogen (N), phosphorus (P), potassium (K), and manganese (Mn) were uniformly added by 120, 50, 25, and 10 mg kg⁻¹, respectively to all pots based on the results of soil testing. Six chickpea (*Cicer arietinum* L. var. Flip 84-42) seeds were planted on the 8th of June, 2012 about 2-cm deep in soil and thinned to three uniform stands 1 week after emergence. Air temperature was kept at 25 ± 5 °C throughout the experiment.

Six weeks after planting, the shoots were harvested on the 23rd of July, 2012 rinsed with distilled water, dried at 65 °C for 48 hours, weighed, ground, and dry ashed at 550 °C. Metal micronutrients including Fe, Mn, Cu, and Zn in plant shoot were determined by atomic absorption spectrophotometer. Dry matter as well as metal micronutrients concentration and uptake were considered as plant responses. Data were analyzed statistically using Excel and SPSS software packages.

RESULTS AND DISCUSSION

Results related to the influence of Fe and Zn levels on the dry matter weight of chickpea are given in Table 1. Application of Fe decreased mean shoot dry weight. Application of both 5 and 10 mg Fe kg⁻¹ decreased mean dry weight of chickpea by about 23% (Table 1). Roomizadeh and Karimian (1996) reported that application of Fe either had no significant effect on dry matter of soybean or decreased it. They concluded that the negative effect of Fe application was attributed to the interference of Fe with Mn absorption or translocation. Application of Zn had no significant effect on shoot dry weight (Table 1). The highest dry weight of chickpea was obtained at 10 mg Zn kg⁻¹ treatment in the absence of Fe addition (Table 1).

 Table 1. Influence of Fe and Zn levels on dry matter weight of chickpea (g/pot)

		•)			
Zn levels	Fe	Fe levels (mg kg ⁻¹)			
$(mg kg^{-1})$	0.00	5.00	10	Mean	
0	1.10 ab*	0.77 bc	0.88 bc	0.92 A	
5	1.05 bc	0.72 c	0.80 bc	0.86 A	
10	1.31 a	0.80 bc	0.73 c	0.94 A	
Mean	1.15 A	0.76 B	0.80 B		

* Means followed by the same letter indicate no significant difference at $p \le 0.05$.

Application of Fe drastically decreased Mn concentration and uptake in chickpea shoot. Addition of 5 or 10 mg Fe kg⁻¹ decreased mean Mn concentration in chickpea shoot by 78% and 85%, respectively (Table 2). Application of 5 or 10 mg Fe kg⁻¹ decreased mean Mn uptake in chickpea shoot by 86% and 89%, respectively (Table 2). Ghasemi-Fasaei et al. (2003) reported that antagonistic effect of Fe with the absorption and/or translocation of Mn following Mn addition might be responsible for the decrease in Mn content of soybean genotypes. Ghasemi-Fasaei et al. (2005) reported that Fe interfere with Mn translocation from the root to the shoot of chickpea. Addition of Fe decreased both shoot dry matter yield and shoot Mn uptake (Tables 1 and 2) demonstrating that a decrease in the amount of Mn uptake might be responsible for the decrease in Mn content of chickpea shoot. Addition of Zn, however, had no significant effect on Mn concentration or uptake in chickpea shoot (Table 2).

Table 2 .Influence of Fe and Zn levels on Mn concentration $(\mu g/g)$ and uptake $(\mu g/pot)$ in chickpea shoot.

Zn levels Fe levels (mg kg ⁻¹)						
(mg kg ⁻¹)	0	5	10	Mean		
	Mn	on concentration	on			
0	59.3 a	9.82 b	9.85 b	26.3 A		
5	73.5 a	11.8 b	10.3 b	31.9 A		
10	65.0 a	22.1 b	9.45 b	32.2 A		
Mean	65.9 A	14.6 B	9.86 B			
		<u>Mn uptake</u>				
0	64.0 b*	7.67 c	9.45 c	27.2 A		
5	76.9 ab	8.45 c	8.26 c	31.2 A		
10	83.6 a	16.1 c	7.21 c	35.6 A		
Mean	75.0 A	10.72 B	8.31 B			

* Means followed by the same letter indicate no significant difference at $p \le 0.05$.

In a similar manner with the dry weight of chickpea, the highest Mn uptake in chickpea shoot was obtained following the application of 10 mg Zn kg⁻¹ in the absence of Fe addition (Table 2).

Although addition of 10 mg Fe kg⁻¹ significantly increased mean Fe concentration in chickpea shoot, the effect of Fe application on mean Fe uptake in chickpea shoot was negligible (Table 3).

However Zn addition had no significant effect on mean Fe concentration or uptake in chickpea shoot (Table 3).

Table 3. Influence of Fe and Zn levels on Fe concentration $(\mu g/g)$ and uptake $(\mu g/pot)$ in chickpea shoot.

Zn levels	Zn levels Fe levels (mg kg ⁻¹)						
$(mg kg^{-1})$	0	5	10	Mean			
	Fe	concentrat	ion				
0	65.9 ab*	65.0 ab	96.3 a	72.3 A			
5	64.1 ab	71.1 ab	101 a	75.7 A			
10	45.6 b	82.4 ab	88.8 a	78.8 A			
Mean	58.6 B	72.9 B	95.4 A				
		Fe uptake					
0	72.1 a	51.2 a	85.7 a	69.7 A			
5	65.6 a	47.1 a	80.5 a	64.4 A			
10	59.5 a	68.2 a	64.4 a	64.1 A			
Mean	65.8 A	55.5 A	76.8 A				

* Means followed by the same letter indicate no significant difference at $p \le 0.05$

Application of 5 and 10 mg Fe kg⁻¹increased mean Zn concentration by 79% and 114%, respectively (Table 4). Application of Zn had no significant effect on mean Zn concentration as compared to that of the control group.

Mean Zn uptake in chickpea tissues following the application of Fe levels did not show any trend (Table 4).

Table 4. Influence of Fe and Zn levels on Zn concentration $(\mu g/g)$ and uptake $(\mu g/pot)$ in chickpea shoot.

7 1 1							
Zn levels Fe levels $(mg kg^{-1})$							
$(mg kg^{-1})$	0	5	10	Mean			
	1	Zn concentr	ation				
0	20.5 d*	21.5 d	21.6 d	34.1 AB			
5	38.5 c	18.4 d	57.1a	27.2 B			
10	43.2 bc	41.7 bc	51.7 ab	43.5 A			
Mean	21.2 B	38.0 A	45.5 A				
		Zn uptak	<u>ke</u>				
0	21.8 cd	16.6 d	18.9 d	19.1 C			
5	40.3 b	13.2 d	45.7 ab	33.1 B			
10	56.0 a	33.0 bc	37.8 b	42.4 A			
Mean	39.4 A	21.0 B	34.2 A				

* Means followed by the same letter indicate no significant difference at $p \le 0.05$

Application of Zn, however, increased mean Zn uptake in chickpea shoot (Table 4). Application of 5 or 10 mg Zn kg⁻¹ increased mean Zn uptake of aboveground tissues by 73% and 123%, respectively (Table 4). Ghasemi-Fasaei et al. (2005) reported that mean shoot Zn concentration or uptake in chickpea was not affected by Fe- chelate application.

Addition of 5 mg Fe kg⁻¹ had no significant effect on mean Cu concentration or uptake in chickpea tissues, whereas, application of 10 mg Fe kg⁻¹ increased mean Cu concentration and uptake in chickpea tissues by about 90% and 36%, respectively (Table 5). Addition of Zn, however, had no significant effect on Cu concentration or uptake in chickpea shoot (Table 5).

Table 5. Influence of Fe and Zn levels on Cu concentration $(\mu g/g)$ and uptake $(\mu g/pot)$ in chickpea shoot.

Zn levels Fe levels (mg kg ⁻¹)						
$(mg kg^{-1})$	0	10	Mean			
	<u>Cı</u>	1 concentrat	tion			
0	7.27 bc*	7.40 bc	8.58 bc	7.75 A		
5	5.50 bc	7.33 bc	13.40 a	8.74 A		
10	4.13 c	8.52 bc	10.22 ab	7.62 A		
Mean	5.63 B	7.75 B 10.73 A				
		Cu uptake				
0	7.53 ab	5.70 b	7.63 ab	6.96 A		
5	5.68 b	5.06 b	10.4 a	7.023 A		
10	5.41 b	6.77 ab	7.41 ab	6.53 A		
Mean	6.21 B	5.84 B	8.47 A			

* Means followed by the same letter indicate no significant difference at $p \le 0.05$.

Correlation coefficients between Fe- and Zn- rates and responses of chickpea are given in Table 6. A significant negative correlation was observed between Mn uptake and Fe rates demonstrating a reduction in shoot Mn uptake following Fe application. Such antagonistic relationships can also be observed in Table 2. In a similar manner with Mn uptake, shoot dry weight of chickpea was also negatively correlated with Fe rates (Table 6). Shoot dry weight of chickpea showed significant positive correlations with the uptakes of Zn and Mn which revealed that the patterns of changes in dry matter yield was in coordination with those of Zn and Mn uptakes (Table 6). Zhao et al. (2011) studied the effect of Fe and Zn on micronutrient levels in wheat in a nutrient solution experiment and observed that Zn, Cu and Mn concentrations were negatively correlated with leaf Fe concentrations, but positively correlated with stem Fe concentrations. They also reported that leaf Mn concentrations were negatively correlated with Zn concentration in different parts of wheat.

CONCLUSIONS

Addition of Fe decreased both shoot dry matter yield and shoot Mn uptake demonstrating that a reduction in the amount of Mn uptake might be responsible for the decrease in the chickpea shoot weight. Application of Zn, however, had no significant effect on mean shoot dry weight and Mn uptake of chickpea. Although the addition of 10 mg Fe kg⁻¹ significantly increased mean Fe concentration in chickpea shoot, the effect of Fe application on mean Fe uptake was negligible. Application of Zn had no significant effect on mean Fe concentration or uptake in chickpea shoot. Iron addition increased mean Zn concentration in chickpea shoot. Zinc addition increased mean Zn uptake in chickpea shoot. Application of the highest Fe level increased mean Cu concentration or uptake in chickpea tissues. Addition of Zn, however, had no significant effect on

Cu concentration or uptake in chickpea shoot. Both shoot dry weight and Mn uptake of chickpea were negatively correlated with Fe rates. Shoot dry weight of chickpea showed significant positive correlations with the uptakes of Zn and Mn indicating that the patterns of changes in these responses were almost similar. Since Fe chelate addition can cause nutrients imbalance such as reduction in Mn uptake and may result in a decrease in plant growth, it appears that the use of Fe efficient cultivars should be considered as an appropriate practice for crops grown on Fe-deficient calcareous soils. It appears that the selection of varieties with stable expressions of grain high in Fe and Zn levels across diverse environments is as vital as increasing the concentration of these nutrients in the grain (Oikeh et al., 2004).

Table 6.	Correlation	coefficients	between Fe-	and Zn-rates	and plar	nt response	s of chickpea

	Zn-rates	Fe-rates	Shoot dry weight	Zn-uptake	Cu-uptake	Fe-uptake
Mn-uptake	0.106	-0.845**	0.744**	0.470*	-0.178	0.051
Fe-uptake	-0.101	0.198	0.312	0.294	0.691**	
Cu-uptake	-0.074	0.392*	-0.009	0.292		
Zn-uptake	0.628**	-0.141	0.518**			
Shoot dry weight	0.053	-0.596**				

*, and **: significant at $p \le 0.05$ and $p \le 0.01$, respectively.

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اثر کلات آهن و سولفات روی بر رشد و ترکیب شیمیایی نخود در یک خاک آهکی

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> **واژه های کلیدی:** روی آهن برهمکنش

عدم توازن تغذيه اي

چکیده- اثر تیمارهای آهن و روی بر رشد و ترکیب شیمیایی نخود طی یک آزمایش گلخانه ای مطالعه گردید. کاربرد آهن سبب کاهش میانگین وزن خشک اندام هوایی نخود گردید. اما کاربرد سولفات روی اثر معنی داری بر میانگین وزن خشک اندام هوایی نخود نداشت. مصرف سطوح آهن سبب کاهش قابل ملاحظه ای در غلظت و جذب کل منگنز شد. اما سطوح روی اثر معنی داری بر غلظت و جذب کل منگنز نداشت. کاربرد سطوح روی یا آهن تاثیر معنی داری بر میزان جذب کل آهن اندام هوایی نخود نداشت. افزودن آهن سبب افزایش میانگین غلظت روی و آهن گردید. افزودن سطوح روی سبب افزایش جذب کل روی گردید. اگر چه افزودن ۵ میلی گرم آهن در کیلوگرم تاثیر معنی داری بر میانگین غلظت و جذب مس نداشت اما کاربرد ۱۰ میلی گرم آهن در کیلوگرم سبب افزایش معنی دار میانگین غلظت و جذب مس گردید. افزودن سطوح روی تاثیر معنی داری بر میزان غلظت و جذب كل مس اندام هوايي نخود نداشت. همبستكي منفي معنى داري بين ميزان جذب منگنز اندام هوایی نخود و سطوح آهن افزوده شده به دست آمد که نشان دهنده کاهش در میزان جذب منگنز با افزایش سطح آهن مصرفی بود. وزن خشک اندام هوایی نخود نیز همبستگی منفی معنی داری با سطوح آهن افزوده شده نشان دادند. همبستگی های مثبت معنی داری بین وزن خشک اندام هوایی نخود با مقادیر جذب منگنز و روی به دست آمد که نشان دهنده تطابق بین روند تغییرات وزن خشک اندام هوایی نخود با روند تغییرات جذب منگنز و روی اندام هوایی این گیاه بود. نظر به اینکه مصرف روی سبب افزایش وزن خشک نخود نگردید و مصرف کلات آهن ممکن است منجر به عدم توازن در وضعیت عناصر غذایی و به دنبال آن کاهش رشد گیاه گردد به نظر می رسد که استفاده از ژنوتیپ های آهن کارا و روی کارا می تواند به عنوان راه حل مناسب جهت کاشت گیاهان در خاکهای با کمبود این عناصر در نظر گرفته شود.