

## MORPHO-AGRONOMICAL RESPONSE OF RICE (*ORYZA SATIVA* L.) TO FOLIAR IRON

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### Abstract

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Rice (*Oryza sativa* L.) is a crop set to be in high demand of market. Among the nutrients, iron is an essential element in plants' phenological cycles. Assessment of this element effects could lead to improving the plant growth as well as the quantity and quality of yield. To achieve this, the effect of foliar application of iron was examined on yield and components of rice. The experiment was conducted based on randomized block design with three replicates. The morphologic and agronomic feedbacks of the rice were analyzed by exposing the rice seedlings to different concentrations of Fe solution (0, 50, 100 and 150 mgL<sup>-1</sup>). It was revealed that spraying Fe had significant effect on grain yield (GY), grains per plant (GP) and grain length (GL). Estimation of Correlation coefficients showed that, GY was positively correlated with GL and GP, while it was negatively correlated with unfilled grains (UG). According to multiple regression analysis using the stepwise method, it was concluded that increasing in the GY was mainly due to increase of GP and also reduction of UG amount. Consequently, the results suggest that, spraying Fe could be considered as a suitable technique to increase rice grain yield.

*Key words:* chemical fertilizers, micro-nutrients, iron concentration, rice

*Abbreviations:* Fe - Iron foliar; FG - number of filled grains; GL - grain length; GP - grains per plant; GY - grain yield; GW - 1000 grains weight; PB - paddy breadth; PH - plant height; PL - panicle length; UG - number of unfilled grains

### Introduction

Nowadays, although rice (*Oryza sativa* L.) is an important food source of more than two billion people across the world, but many countries cannot produce enough rice from land and water resources. In Iran due to some limitation factors such as rainfall, rice is usually planted in northern parts of Iran. Therefore, it has kept ahead of rising demand.

Like other agricultural crops, for the best growth and yield of rice, adequate macro and micronutrients must be available for the plant (Colangelo and Guerinot, 2004). Iron (Fe) is one of the most important micronutrients, which significantly affects plant growth and yield. Iron promotes the formation of chlorophyll, enzyme mechanism, which operates the respiratory system of cells, reactions involving cell division and growth (Colangelo and Guerinot, 2004).

Plants require macro and micro elements to grow. In soil, micro nutrients are not always present in the solution and their availability is limited due to several factors, which mainly limit their solubility. The main factors determining the level of soluble macro and micro nutrients in soil solution are: Oxidation-reduction reactions and the pH. The solubility of Fe depends on the changes of pH inside the soil, and thus any changes in soil pH may affect Fe availability to the plant, which covers 30% of agricultural soils in the world (Mori, 1999) and containing Fe as oxide and hydroxide compounds with little solubility (Lindsay and Schwab, 1982). In such soils, Fe uptake considerably diminishes and signs of chlorosis appear on the plant, resulting in substantial yield cut (Marschner, 1995). Thus, spraying Fe on leaves may be more effective than the application of Fe inside the soil.

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Along with activation, stomatal absorbance may be important in the foliar application of nutrients, since most of the sprayed nutrients are absorbed through the stomata. Eichert and Burkhardt (2001) reported a model showing that stomatal pathway is for large anions such as uranine and for small cations such as  $\text{Fe}^{3+}$ . This may further prove the affectivity of foliar Fe on rice growth and yield. In addition, the physico-chemical properties of compound may also be a determining factor in foliar application. Besides their role in producing organic molecules, it is found that macro- and micro-nutrients may act as signal molecules in plant, regulating plant functions (Coruzzi and Bush, 2001).

Researchers have indicated that different plant species have dissimilar strategy to iron uptake. Non-graminaceous plants in encounter to low concentration of Fe act three different functions; (i) production of  $\text{H}^+$  into the rhizosphere by  $\text{H}^+$ -ATPase, resulting in decreased soil pH and higher  $\text{Fe}^{3+}$  uptake ((Romheld, 1987; Colangelo and Guerinot, 2004), (ii) reducing  $\text{Fe}^{3+}$  to  $\text{Fe}^{2+}$  because of the ferric chelate reductase action of FRO2 (Robinson et al., 1999), and (iii) transport of  $\text{Fe}^{2+}$  into the plant by IRT1, the main root transporter of iron (Eide et al., 1996; Vert et al., 2002).

The grasses (Takagi et al., 1984), bacteria and fungi (Guerinot, 1994), use the second strategy to iron uptake, chelating Fe rather than cutting it, with excreting phytosiderophores, from mugineic acid family, into the soil and thus chelating  $\text{Fe}^{3+}$ , which is later incorporated into plant organic molecules using special transporters (Colangelo and Guerinot, 2004; Ogo et al., 2006).

Application of acid and Fe-fortified organic materials has been developed over the past ten years due to (i) the acidifying property of the material with Fe, (ii) the chelating property with Fe, and (iii) the carrying property of acidifiers. To reduce Fe deficiency the following solutions may be suggested (Wallace, 1991) proper soil management, right balance of nutrients such as P and micronutrients, using Fe chelates, use of ferrous materials for spray application, or urea-acid sulfate-Fe, or gradually-released Fe chelates especially at seeding.

Due to this issue, the present study was aimed to investigate the effects of foliar iron on rice (*Oryza sativa* L.) considering yield and its related traits. Despite this, the growth quality and yield of the rice under foliar iron on the paddy fields should be subjected to further investigations in the future as this case has not been scrutinized yet.

## Materials and Methods

### Germination and growth condition

The rice seeds were provided from the Rice Research Institute of Iran, Rasht, Iran. The seeds were germinated in

greenhouse to provide nursery. After one month of germination, the seedlings were cultivated in the farmers' fields at the Mobarake region of Isfahan province, Iran. Three and five weeks after cultivation, 272 kg.ha<sup>-1</sup> (125 N kg.ha<sup>-1</sup>) of urea was applied in the greenhouse. After 45 days of germination, the seedlings were cultivated in the farmers' fields at the Mobarake region of Isfahan province, Iran. After planting the seedlings, urea and ammonium phosphate were applied at 160 and 300 kg.ha<sup>-1</sup> in the main field, respectively. The field was irrigated and weeds, pests, and diseases were controlled, chemically and mechanically.

### Experimental design

We carried out an experiment based on randomized complete block design (RCBD) with five concentration of iron (0, 50, 100, and 150 mgL<sup>-1</sup>) and three replicates during March 2010 - April 2011. In this study, the Fe soluble in water with free amino acids (Trade Corp Company, Spain) as a bio-nutrient with functional L-a-amino acids was used. After two months the Fe solutions were sprayed onto the rice plants at the stem producing stage. The experimental unit for each treatment was 3 × 3 meter with a 20×20 cm<sup>2</sup> space for each seedling.

At the physiological maturity, the plants were harvested from surface of 1m (including 25-28 plants in each plot). Ten plants from each plot randomly were selected and data on grains per plant (GP), panicle length (PL cm), plant height (PH cm), number of filled (FG), and unfilled (UG) per 100 grains, paddy breadth (PB mm), grain breadth (GB mm), grain length (GL mm), and 1000-grain weight (GW g), seed moisture were measured.

### Statistical analysis

At first, the raw data were tested for normality distribution by using the SAS software No. 9 and the main data were analyzed for measured traits using analysis of variance and the Duncan's multiple range test ( $P \leq 0.01$ ). Analysis of variance and mean comparison were done using Proc GLM of SAS software.

## Results

Analysis of variance showed that spraying Fe on leaves significantly affected GY, PH, GL, PB and GP (Table 1). The variation due to year was significant for GY, PH, PL, FG, UG, PB and GP (Table 1). The coefficient of variation was varied from 1.13 for GL to 23.93 for UG. The interaction year and Fe had no significant effect on studied traits (Table 1). In general, traits such as GY, PH, PL, FG, GL, PB and GP were higher in the first year of experiment; while the amount

of UG was lower (Table 2). During two years and based on combined data, application of foliar Fe improved yield and some important yield components such as GW and GP (Figure 1). The highest yield and its components were obtained at 150 mgL<sup>-1</sup> of Fe concentration. There was no significant difference among 0 and 50 mgL<sup>-1</sup> concentrations on GY and GP at. Grain yield means were 710.92, 720.67, 754.83 and 793.25

gm<sup>-2</sup> at 0, 50, 100 and 150 mgL<sup>-1</sup> concentrations respectively, showing a gradual raise concurrent with increase of Fe concentration. The maximum raise for GY with 11.6% or 82.33 gm<sup>-2</sup> belonged to 150 mgL<sup>-1</sup> concentration. According to these results, there was a significant positive linear regression between GY and different level of Fe concentrations (Table 3). The corresponding regression model fitted for grain yield

**Table 1**  
Combine analysis of data derived from the experiment of foliar Fe on studied morphological traits in rice

S.O.V	df	Mean Square								
		GY	GW	PH	PL	FG	UG	GB	GL	GP
Y	1	109620.17**	1.42 <sup>ns</sup>	222.35**	8.76**	436.91**	10.41**	0.004 <sup>ns</sup>	0.029*	230068.92**
Fe	3	8357.03**	1.35 <sup>ns</sup>	56.68*	0.78 <sup>ns</sup>	16.96 <sup>ns</sup>	0.24 <sup>ns</sup>	0.003 <sup>ns</sup>	0.067**	21487.81**
Fe × Y	3	207.03 <sup>ns</sup>	0.07 <sup>ns</sup>	2.81 <sup>ns</sup>	0.047 <sup>ns</sup>	7.45 <sup>ns</sup>	0.58 <sup>ns</sup>	0.001 <sup>ns</sup>	0.008 <sup>ns</sup>	1040.91 <sup>ns</sup>
Error	14	163.57	0.36	11.70	0.34	120.56	1.08	0.005	0.005	1362.21
C.V, %		1.72	2.60	4.00	2.84	13.28	23.93	2.86	1.13	3.02
Mean		744.92	22.99	85.44	20.64	82.68	4.34	2.44	6.13	1222.55

\*\* and \* mean that the corresponding F value is significant at the 0.01 and 0.05 levels, respectively

Y: year, Fe: Iron foliar, R: replication, CV: coefficient of variation, GY: grain yield (g.m<sup>-2</sup>), GW: 1000 grains weight (g), PH: plant height (cm), PL: panicle length (cm), FG: number of filled grains, UG: number of unfilled grains, GL: grain length (mm), PB: paddy breadth (mm), GP: grains per plant

**Table 2**  
Mean comparison of rice morphological traits under impact of year factor

Year	Means						
	GY	PH	PL	FG	UG	GL	GP
2010	812.50a	88.49a	21.24a	86.95a	3.68b	6.17a	1320.46a
2011	677.33b	82.40b	20.03b	78.417b	5.00a	6.10b	1124.64b

GY: grain yield (g.m<sup>-2</sup>), GW: 1000 grains weight (g), PH: plant height (cm), PL: panicle length (cm), FG: number of filled grains, UG: number of unfilled grains, GL: grain length (mm), PB: paddy breadth (mm) and GP: grains per plant

**Table 3**  
Correlations coefficients between rice morphological traits under effect of different concentrations of foliar Fe

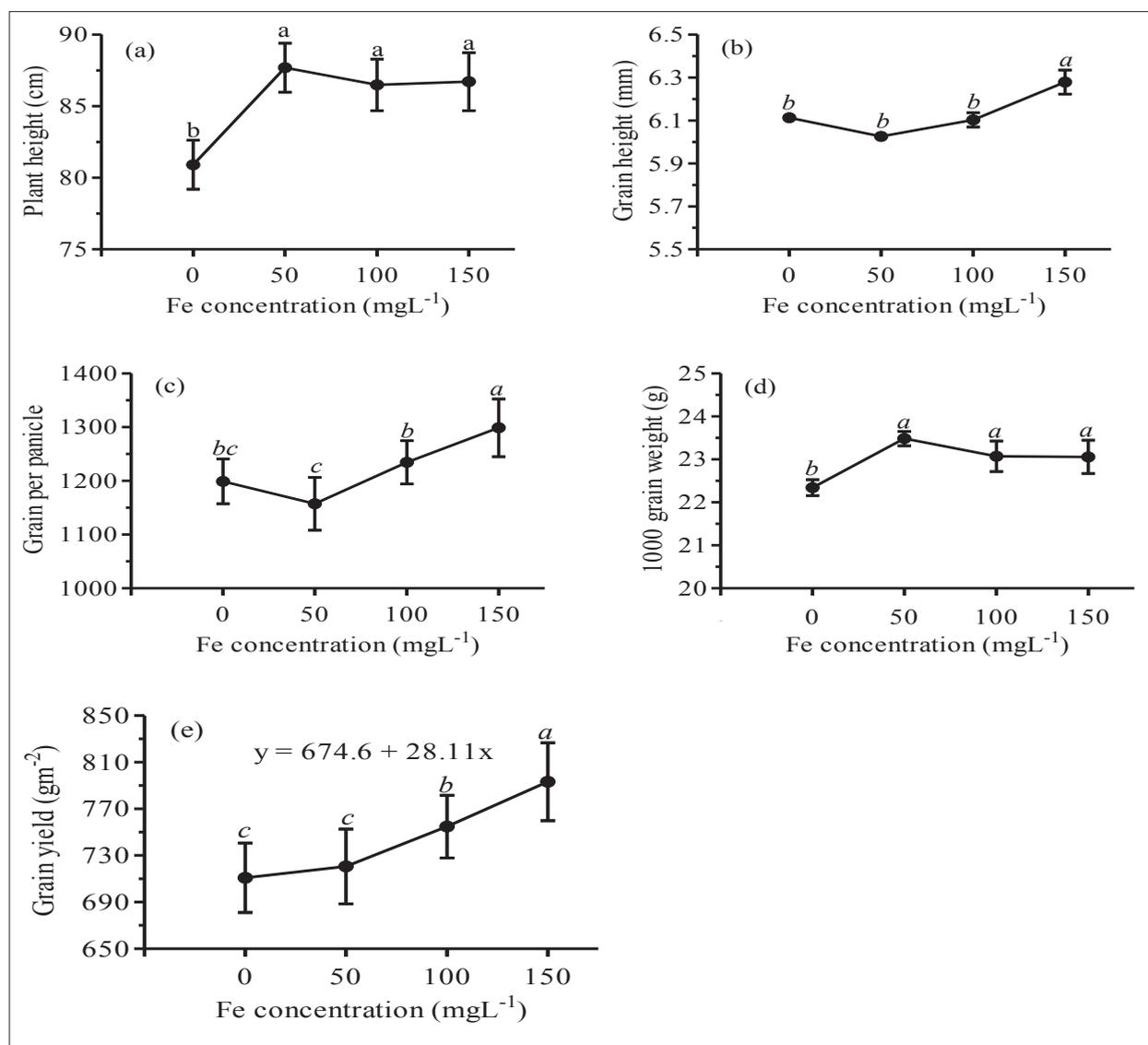
	GY	GW	PH	PL	FG	UG	GB	GL	GP
GY	1								
GW	0.354	1							
PH	0.665**	0.541**	1						
PL	0.669**	0.166	0.454*	1					
FG	0.298	0.126	0.400	0.614**	1				
UG	-0.512*	0.282	-0.134	-0.340	-0.213	1			
GB	0.060	0.216	0.167	0.323	0.539**	-0.129	1		
GL	0.551**	-0.131	0.337	0.392	-0.048	-0.144	-0.124	1	
GP	0.946**	0.032	0.533**	0.655**	0.273	-0.640**	-0.015	0.639**	1

\*\* and \* mean that correlation is significant at the 0.01 and 0.05 levels, respectively (2-tailed). GY: grain yield (g.m<sup>-2</sup>), GW: 1000 grains weight (g), PH: plant height (cm), PL: panicle length (cm), FG: number of filled grains, UG: number of unfilled grains, GL: grain length (mm), PB: paddy breadth (mm), GP: grains per plant

was  $GY=702.7 + 0.562 F$  with  $R^2=0.946$ , which shows a suitable goodness of fit which is of important in terms of estimating the expected grain yield for a given Fe concentration (Figure 1).

Similarly, GP as one of the important yield components showed an increase by 8.3% (or 99.63 grains per plant) at 150  $mgL^{-1}$  concentration in comparison with control. The corresponding values for GP were equal to 1199.24, 1157.50, 1234.60 and 1298.87, respectively showing a relative reduc-

tion at second Fe concentration level (50  $mgL^{-1}$ ) followed by a gradual raise at 100 and 150  $mgL^{-1}$  levels. Also GW 'as one of the other important yield components' showed a raise equal to 5.15% (or 1.15 g). Also, spraying Fe increased the GL. The maximum raise in GL values (0.17 mm) observed at 150  $mgL^{-1}$  of Fe, compared with control. At the different level of Fe concentrations (0, 50, 100 and 150  $mgL^{-1}$ ) grain length mean were equal to 6.11, 6.03, 6.11 and 6.28 mm, respectively showing that the variation trend was similar to GP. The re-



**Fig. 1.** Effect of different levels of foliar Fe concentration on plant height (a), grain height (b), grain per panicle (c), 1000 grain weight (d) and grain yield (e) in rice. Mean values  $\pm$  S.E are from three independent replicates and values superscripted by different letters are significantly different by Duncan's multiple range test ( $p \leq 0.01$ )

sults suggested that increase of Fe concentration from 50 to 150 mgL<sup>-1</sup> had a slightly negative effect on GW (Figure 1).

Estimation of correlation coefficients between studied traits showed, under impact of foliar Fe, GY was positively correlated with GL, GP, PL and PH while was negatively correlated with UG. Also some positive and significant correlations were found between GW-PH, GP-PH, PL-FG, PL, GP, FG-GB, and GL-GP while the correlation between UG and GP was negative (Figure 2).

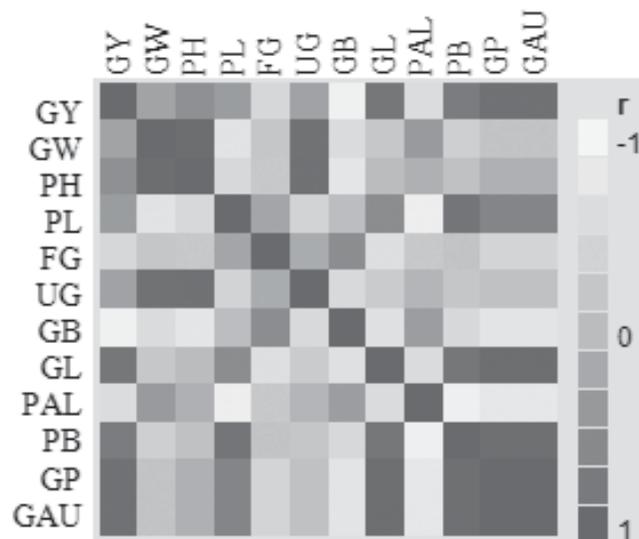
Stepwise linear regression (SLR) analysis showed that GY was positively regressed with GP, GW and was negatively regressed with PH. Standardized coefficients showed that GP had the highest impact on GY. The corresponding regression model fitted for grain yield was  $GY = 740.81 + 0.617 GP + 32.99 GW - 0.314 PH$  with  $R^2 = 0.99$ .

## Discussion

According to the results, the interaction of year Fe was not significant showing that effect of foliar Fe on improvement of quantity of yield and its components was independent from the impact of year. The reduction of GY observed in second year was mainly due to the significant raise in the amount of UG and also significant decrease of GP. Foliar application of nutrients especially micronutrients such as iron may be an efficient way to provide the crops with needed nutrients, especially at regions where soil pH is a limiting factor for nutrient solubility and uptake. Marschner and Crowley (1998) found that foliar application of Fe could provide the barley stressed plants with adequate Fe. According to Eichert and Burkhardt (2001) most parts of the solutes sprayed onto the leaves are absorbed through the stomatal pathway.

The results of this study suggested that spraying foliar Fe fertilizer had a positive effect to improve rice grain yield. Also, application of Fe at 150 mgL<sup>-1</sup> was the proper concentration to increase rice grain yield in the field. This method may be economically important compared to the soil fertilization of Fe. The foliar Fe may highly be absorbed through the plant stomata, while soil fertilization may not be efficient since Fe compounds are much subjected to precipitation. Therefore highly reduced uptake especially in calcareous soils (Eichert and Burkhardt, 2001).

This study showed that foliar Fe could significantly increase GY, GP and GL. With regard to the strong and significant correlation observed between GY-GP (0.946,  $P \leq 0.01$ ) and considering SLR results, increase of GP was the main responsible of increase in GY. In fact, application of foliar Fe increased the GP, which led to increase in GY. The increase in GP was due to increase in the amount of leaves chloro-



**Fig. 2. Correlations coefficients between 12 morphological traits in rice under effect of different concentrations of foliar Fe. The strength and direction of the correlations among the different traits are indicated by the color (red indicates positive correlations, while blue indicates negative correlations, and the shading represents the strength of the correlation)**

phyll and thus efficiency of photosynthesis process. The SLR results showed a significant but poor negative effect of PH (-0.020) on GY indicating a competition between vegetative traits (which are enhanced under the effect of foliar Fe) and reproductive functions.

Compared to the control, GY, GP and GL were numerically decreased at 50 mgL<sup>-1</sup> concentration levels followed by increase at 100 and 150 mgL<sup>-1</sup> levels. In this regard, Shenker and Chen (2005) noted that one major problem impairing the success of foliar applications of Fe is slow penetrating Fe through the leaf. High solubility and low molecular weight are key factors governing foliar uptake. Thus, this reduction could be due to slow penetration along with low concentration of Fe. However, the leaf apoplasmic pH might be decreased by foliar Fe application and thereby the leaf symplastic Fe uptake would be advanced (Shenker and Chen, 2005). It seems that higher concentrations overcome to the problem of slow penetration of foliar Fe.

When Fe is deficient, plant chlorophyll content substantially decreases, resulting in the appearance of chlorotic signs on plant leaves. From this viewpoint, it can be thought that Fe spraying on leaves led to raise leaf chlorophyll content as well as leaf area duration. On the other hand, raising these

two traits led to more photosynthesis and thus needed carbohydrate materials for grain filling. Therefore, GY along with GP and GL were increased (Figure 1).

## Conclusion

The results of this study showed that spraying foliar Fe fertilizer raised GY, GP and GL. The maximum raise was observed at 150 mgL<sup>-1</sup> Fe concentrations, while a numerical and non-significant reduction was recorded at 50 mgL<sup>-1</sup> level. According to the results of this study, foliar spray of Fe could be considered as a suitable technique to raise grain yield in rice.

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