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Response of Different Weight Groups of Saffron Corms to Different Food Regimes

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Abstract: *Saffron is the most expensive medicinal crop in the world with several medical and soothing properties. The separate application of organic and inorganic fertilizers has certain effects on plants, but their integrated application is of importance for plants, especially for saffron. The present study focused on the response of different weight groups of saffron corms to four food regimes in Torbat-e Heydarieh University over the 2015-2017 growing seasons in a split-plot experiment based on a randomized complete block design with three replications. The experimental treatments included FR1 as control (no fertilizer application), FR2 (manure, humic acid, micronutrients, and Thiobacillius), FR3 (blood meal, monopotassium phosphate, Fe chelate, humic acid, and Delfard fertilizer), and FR4 (chicken manure, ammonium nitrate, granular humic acid, urea, 20-20-20 fertilizer, Marmarine fertilizer, Dekap fertilizer, and Coda Mix fertilizer) as the main plots and maternal corm weight at three levels (<8 g, 8-12 g, >12 g) as the sub-plots. Saffrons were planted in September 2015. The effect of food regime on the recorded traits in the first year (2016-2017) showed that plants exposed to FR4 produced the highest corm weight, corm diameter, stigma fresh and dry weight, flower fresh weight, and flower number, and the highest levels of these traits were obtained from plants grown from >12 g maternal corms. The interactive effect of the factors in the first year indicated that plants derived from >12 g corms and fertilized with FR4 were related to the highest stigma fresh and dry weight of 38.6 and 7.8 kg ha^{-1}, respectively and the highest corm weight of 1710.88 g m^{-2}. According to the food regime of FR4, it can be concluded that the application of chicken manure, humic acid, macronutrients, and micronutrients such as Fe, Zn, Mn, and so on can play a significant role in improving saffron yield.*

Keywords: *Corm Weight, Fertilizer, Humic Acid, Micronutrients, Stigma Dry weight*

INTRODUCTION

Saffron (*Crocus sativus* L.) is an herbaceous perennial plant from the family Iridaceae with a special place among all known 85 species of the genus Crocus. The edible part of saffron is its crimson stigma (Grilli-Caiola, 2004). As the most expensive agricultural and medicinal crop of the world (Koocheki et al., 2012), saffron has several medical and soothing properties (Ahmadian, 2013). It has a very low yield in Iran versus other producing countries so that its mean yield in Iran is 0.23 kg ha^{-1} in the first year and 1.8 kg ha^{-1} in the second year whereas it is 4-6 kg ha^{-1} in the first year and 10-12 kg ha^{-1} in the second year in Spain as the second leading saffron producer of the world (Hassanzadeh

Aval et al., 2013). Saffron yield in the first year is strongly influenced by the size and reserves of the corms used as seeds. In the first year, the corms form daughter corms, which will be the seeds of the second year. The newly formed corms affect the yield of the subsequent years (Amirshekari et al., 2007).

Organic fertilizers are an important component of agriculture sustainability. Soil organic matter is a key parameter of soil quality as it influences soil physical, chemical and biological properties and processes (Haynes, 1996). Among environmentally-friendly fertilizers, humic acid is an organic fertilizer that is not harmful to the environment and improves soil physical, chemical and biological structure with considerable desirable effects of the quantitative and qualitative traits of crops due to its hormonal compounds (Sabzevari et al., 2010). The application of manures has a long history in agricultural activities and can still play an effective role in sustainable and biological agriculture (Majiian et al., 2008).

Saffron yield responds to N/C ratio of soil positively. Thus, adequate care should be given to the application of nitrogen and organic fertilizers because excessive use of nitrogenous fertilizers may disrupt soil C/N ratio, thereby impairing saffron yield. Calcium-rich soils are proper for saffron growth, and manure is used to satisfy the requirements of this plant species because of its high potassium content (Saeedi-rad and Mokhtarian, 2010).

A study on the effects of applying the biological fertilizer Nitroxin and Delfard on saffron growth and yield reported that the highest number of flowers per unit area and flower and stigma yields were obtained from plants treated with Delfard (Koocheki et al., 2011). Chemical fertilizers and cow manure have better effects on flower and stigma yield of saffron than chicken manure (Rezvani-Moghaddam et al., 2006). The foliar application of Delfard increased corm number and yield per unit area, flower number per unit area, flower fresh and dry yield, and stigma and style yields (Rezvani Moghadam et al., 2013).

Phosphorous is a macronutrient of plants with an essential role in root growth and development, vegetative growth, tillering, plant resistance to winter chilling, dormancy and early-maturity, N uptake, flowering, fruit setting, crop maturity, and plant quality (Leticia et al., 2007).

Materials and Methods

The responses of different saffron corm weight groups to four nutrient management with organic and chemical fertilizers were explored in the University of Torbat-e Heydarieh in Torbat-e Heydarieh (Lat. 35°20' N., Long. 59°133' E., Elevation 1450 m) over 2015-2017 growing season.

Before trial initiation, the soil was sampled and sent to a soil laboratory for the preliminary preparations including drying, beating, and screening. The results of its chemical analysis showed that the soil of the study site was loam-clay with a pH of 8.01 and an electrical conductivity of 2.19 mmos cm^{-1}. Table 1 presents the physical and chemical characteristics of the soil.

Table 1. Physical and chemical characteristics of the soil in the study farm down to the depth of 30 cm

EC (mmos/cm)	pH	Organic C (%)	N (%)	P (ppm)	K (ppm)	Lime (%)	Clay (%)	Silt (%)	Sand (%)	Soil texture
2.19	8.01	0.51	0.049	28	360	17.5	34	53	13	Loam-clay

The study was carried out as a split-plot experiment based on a randomized complete block design with three replications in a rental research farm located in Saffron Research Center of the University of Torbat-e Heydarieh in 2015-2016 and 2016-2017 growing seasons.

The treatments were composed of four food regimes as the main plot and three corm weight groups (<8 g, 8-12 g, >12 g) as the sub-plot. The main plot, i.e. food regime, included the following four levels:

- Food Regime 1 (FR1): control in which no fertilizer was applied.
- Food Regime 2 (FR2): 3400 kg ha^{-1} manure was incorporated with soil before corm planting and then, the farm was fertigated with 5 L ha^{-1} humic acid in October 2015. In December 2015, the fertilizer Roshdineh (8% N, 1% phosphate, 9% K, 3% Fe, 6% Zn, 35% amino acids, bacillus) was applied at the rate of 0.3 kg ha^{-1} and humic acid at the rate of 5 L ha^{-1}. This was repeated in February, April, October, and December 2016 and February 2017.
- Food Regime 3 (FR3): blood meal fertilizer was applied to soil at the rate of 6600 kg ha^{-1} before planting. Then, the field was fertigated with 50 kg ha^{-1} monopotassium phosphate, 2 kg ha^{-1} Fe chelate, and 4 kg ha^{-1} humic acid powder in December 2015. In February 2016, the fertilizer Delfard-8 (3% N, 8% P, 4% K, and Fe, Zn, Mg, and Cu chelates) was applied at the rate of 15 kg ha^{-1} as dissolved in irrigation water. In April 2016, humic acid was applied as fertigation at the rate of 5 L ha^{-1}. In October 2016, the fertilizer 15-5-30 was applied at the rate of 25 kg ha^{-1} and humic acid was applied at the rate of 4 kg ha^{-1} with irrigation, and this was repeated in December 2016 and February 2017.
- Food Regime 4 (FR4): chicken manure was incorporated to the soil at the rate of 3400 kg ha^{-1} before planting in October 2015. After the corms were planted, the farm was fertigated with 400 kg ha^{-1} ammonium nitrate and 300 kg ha^{-1} granular humic acid. In December 2015, it was fertigated with 100 kg ha^{-1} urea, 25 kg ha^{-1} 20-20-20 fertilizer, 1 kg ha^{-1} amino acid, and 200 kg ha^{-1} sulfur. In February 2016, 1 L Marmarine (2% P, 3% K, 250 ppm auxin, gibberellins, and cytokinin) per 1000 L water, 1 L Dekap (42,33,0) per 100 L water, and 1.5 L Coda Mix (0.3 B, 0.38 Cu, 0.4 Mn, 0.8 Mo, 0.64 Zn) per 500 L water were sprayed in two stages. In April 2016, the plots were fertigated with 3 kg ha^{-1} humic acid. The same fertilization regime was repeated in October and December 2016 and February 2017.

After the field was plowed and leveled manually, the plots were prepared at the size of 2.50 m^2 as per the research plan. The plots were spaced 100 cm. Each plot was divided into three parts to be sown with corms based on three weight classes of <8, 8-12, and >12 g. Before planting, the plots were fertilized with the specified food regime. The corms were planted at the depth of 15 cm with an inter-corm spacing of 5 cm on August 27, 2015.

The first irrigation was performed in the first year on October 16, 2016. Flowering initiated on November 5. The flowers were collected on a daily basis and flower fresh weight and stigma fresh and dry weights were recorded. Flowering lasted until December 18, 2016.

Data were statistically analyzed in the SAS 9.2 software package and graphs were drawn using MS-Excel. Means were compared by Duncan's test at the $p < 0.05$ level.

Results and Discussion

Corm weight

This trait was significantly ($p \leq 0.01$) influenced by food regime and corm weight. The interaction of these two factors was also significant ($p \leq 0.05$) for this trait (Table 2).

Table 2. The results of variance analysis (means of squares) for the effect of food regime and corm weight on saffron yield components (2016-2017)

S.O.V.	df	Means of squares					
		Corm weight	Corm diameter	Flower number	Flower fresh weight	Stigma fresh weight	Stigma dry weight
Replication	2	4566.06	0.05	17.02	2.51	0.163	0.004
Food regime (FR)	3	187316.23**	22.4**	3821.66**	524.42**	7.127**	0.29**
Major error	6	1824.99	0.11	27.58	3.98	0.05	0.002
Corm weight (CW)	2	95151.79**	13.32**	3584.19**	272.381**	2.71**	0.124**
FR × CW	6	4385.72*	0.84**	133.19*	6.05*	0.07*	0.002*
Minor error	16	1471.73	0.187	33.98	3.98	0.026	0.0008
C.V. (%)		2.71	6.75	9.21	7.22	6.77	6.13

* and ** show significance at the $p \leq 0.05$ and $p \leq 0.01$ levels, respectively.

Results for the effect of food regime on corm weight showed that the highest and lowest average corm weights of 1565.37 g m^2 and 1239.28 g m^2 were obtained from FR4-treated plants and control plants, respectively. FR2 and FR3 increased corm weight by 5 and 16 percent versus control plants (Table 3). Plants treated with cattle manure exhibited the highest total corm weight, corms with the weight of >8 g, corms with the weight of 4-8 g, and leaf length and width. The highest corm number and the highest number and weight of <1 g corms were obtained from NPK treatment and the highest weight of 1-4 g corms from compost treatment (Teimouri et al., 2013).

Table 3. Means comparison for the effect of food regimes on yield and yield components of saffron (2016-2017)

Food regimes	Corm Weight (g m^{-2})	Corm Diameter (cm)	Flower Number (m^{-2})	Flower fresh weight (g m^{-2})	Stigma fresh weight (g m^{-2})	Stigma dry weight (g m^{-2})
FR1	1239.28 d	4.79 d	39.88 d	11.94 d	1.46 d	0.29 d
FR2	1310.52 c	5.85 c	53.11 c	15.25 c	1.88 c	0.36 c
FR3	1441.49 b	6.42 b	74.55 b	24.45 b	2.97 b	0.59 b
FR4	1565.37 a	8.54 a	85.55 a	28.21 a	3.35 a	1.67 a

Similar letter in each column shows insignificant difference between the means of the treatments according to Duncan's test.

The effect of maternal corm weight on the weight of the sister corm revealed that the highest and lowest average corm weights of 1476.49 and 1298.49 g m^2 were related to the treatment of maternal corms with the weight of >12 g and corms with the weight of <8 g (Table 3).

With respect to the impact of corm weight on saffron yield, it was observed that leaf area and dry matter production over the growth period were increased with corm size and this resulted in the production of more daughter corms at the end of the growth period (Renau-Morata et al., 2012).

Among the interactions, maternal corms of >12 g treated with FR4 produced the highest average sister corm weight of 1710.88 g m^2, and <8 g corms that were not fertilized produced the lowest sister corm weight of 1156.22 g m^2 (Table 5).

The application of higher levels of humic acid and bigger maternal corms improved growth parameters of sister corms partially (Koocheki et al., 2015).

Corm diameter

The simple and interactive effects of food regime and maternal corm weight were significant ($p \leq 0.01$) for this trait (Table 2).

Among the studied food regimes, FR4 was related to the highest average corm diameter of 8.53 cm followed by FR3 (6.42 cm) and FR2 (5.85 cm). The lowest average corm diameter of 4.79 cm was obtained from control plants (Table 3).

The effect of maternal corm weight on corm size showed that 8-12 g corm class produced 20 percent higher corm diameter and >12 g corm class exhibited 39 percent higher corm diameter than control plants (Table 4). It has been reported that flower percentage of saffron was a function of corm diameter and it was recommended to sow corms with a diameter of 3 cm and relative weight of 10 g.

Table 4. Means comparison for the effect of maternal corm weight on yield and yield components of saffron (2016-2017)

Maternal corm weight	Corm weight ($g\ m^{-2}$)	Corm diameter (cm)	Flower number (m^{-2})	Flower fresh weight ($g\ m^{-2}$)	Stigma fresh weight ($g\ m^{-2}$)	Stigma dry weight ($g\ m^{-2}$)
<8 g	1298.49 c	5.34 c	45.41 c	15.06 c	1.95 c	0.38 c
8-12 g	1392.52 b	6.41 b	64.5 b	20.28 b	2.41 b	0.48 b
>12 g	1476.495 a	7.45 a	79.91 a	24.57 a	2.90 a	0.58 a

Similar letter in each column shows insignificant difference between the means of the treatments according to Duncan's test.

The interactive effect of the treatments showed that the highest average corm diameter of 10.31 cm was obtained from >12 g corms treated with FR4 and the lowest (3.51 cm) from non-fertilized <8 g corms (Table 5).

Table 5. Means comparison for the interactive effect of food regime and maternal corm weight on yield and yield components of saffron (2016-2017)

Food regime	Maternal corm weight	Corm weight ($g\ m^{-2}$)	Corm diameter (cm)	Flower number (m^{-2})	Flower fresh weight ($g\ m^{-2}$)	Stigma fresh weight ($g\ m^{-2}$)	Stigma dry weight ($g\ m^{-2}$)
FR1	<8 g	1156.22 h	3.51 h	8.48 g	25.66 h	1.1 i	0.22 g
	8-12 g	1256.42 g	5.06 g	11.43 f	43.66 g	1.35 h	0.28 f
	> 12 g	1305.18 f	5.79 ef	15.91 e	50.33 efg	1.94 f	0.38 e
FR2	<8 g	1219.07 g	5.3 fg	10.42 f	42.33 g	1.56 gh	0.27 f
	8-12 g	1307.46 f	5.98 ef	15.92 e	49 fg	1.71fg	0.34 e
	> 12 g	1405.01 d	6.26 de	19.41 d	68 c	2.38 e	0.48 d
FR3	<8 g	1372.98 e	5.57 ef	18.96 d	54.33	2.32 e	0.46 d
	8-12 g	1466.61 cd	6.26 de	26.19 b	77 ef	3.19 c	0.64 b
	> 12 g	1484.86 b	7.42 c	28.21 b	92.33 c	3.42 b	0.68 b
FR4	<8 g	1445.66 cd	6.97 cd	22.35 c	59.33 d	2.81 d	0.57 c
	8-12 g	1539.56 b	8.33 b	27.56 b	88.33 b	3.38 c	0.66 b
	> 12 g	1710.88 a	10.31 a	34.73 a	109 a	3.86 a	0.78 a

Similar letter(s) in each column shows insignificant difference between the means of the treatments according to Duncan's test.

Flower number

Flower number was significantly influenced by food regime and maternal corm weight ($p \leq 0.01$) and by their interaction ($p \leq 0.05$) (Table 2).

With respect to the effect of food regime on flower number, it was observed that FR4 produced the highest number of flowers per m^2 (85.55) and FR1 produced the lowest number (39.88). FR3 and FR2

exhibited 33 and 86 percent higher flower number than control plants (Table 3). When Thiobacillus was applied to saffron plants, an increase was observed in flower number (Eldin et al., 2001).
The effect of maternal corm weight on flower number showed that the application of >12 g and 8-12 g corms resulted in 47 and 25 percent more flowers per unit area as compared to <8 g corms (Table 4).
The highest flower number per m^2 (109) was obtained from plants grown from >12 g corms and exposed to FR4, and the lowest (25.66) was related to <8 g corms that were not fertilized.

Flower fresh weight

The simple effect of food regime and maternal corm weight was significant on flower fresh weight at the $p \leq 0.01$ level and their interaction was significant for this trait at the $p \leq 0.05$ level (Table 2).
The results for the effect of food regime on flower fresh weight revealed that the highest flower fresh weight of 28.21 g m^{-2} was observed in plants exposed to FR4 and the lowest was 11.94 g m^{-2} observed in plants that were not fertilized.
FR2 and FR3 increased flower fresh weight by 27 and 100 percent versus control, respectively (Table 3). The mixed application of 50% chemical fertilizers, vermicompost, and bacteria was the most effective treatment in improving the quantitative and qualitative yield of saffron so that this treatment was associated with the highest flower number and weight (Rasooli et al., 2015).
With respect to the effect of maternal corm weight, it was observed that the application of >12 g and 8-12 g corms resulted in 34 and 63 percent higher flower fresh weight as compared to <8 g maternal corms (Table 4). As the weight of maternal corms was increased, flower fresh weight was increased considerably so that the highest fresh weight was obtained from plants grown from >12 g maternal corms (Khavari et al., 2016).
About the interactive effect of maternal corm weight and food regime on flower fresh weight, the highest flower fresh weight of 34.73 g m^{-2} was observed in plants grown from >12 g corms and fertilized with FR4 and the lowest fresh weight of 8.48 g m^{-2} was obtained from plants grown from <8 g corms but not exposed to any food regimes (Table 5).

Stigma fresh weight

According to the results of variance analysis (Table 2), stigma fresh weight was significantly influenced by food regime and maternal corm weight at the $p \leq 0.01$ level and by their interaction at the $p \leq 0.05$ level.
With respect to the effect of food regime, it was observed that the highest stigma fresh weight of 3.35 g m^{-2} was obtained from plants exposed to FR4 and the lowest (1.46 g m^{-2}) from the non-fertilized plants. FR3 and FR2 increased stigma fresh weight by 103 and 28 percent versus control plants (Table 3). Various studies have reported the positive effect of complete fertilizer use on saffron yield as compared to other fertilization regimes so that the higher the amount of complete nutrients in the fertilization regime is, the higher the flower yield will be (Jahan and Jahani, 2007).
As far as maternal corm weight was concerned, the application of >12 g and 8-12 g corms improved this trait by 48 and 23 percent as compared to <8 g corms (Table 4). The effect of larger maternal corms has been reported to be positive on saffron flowering (Nasiri Mahallati et al., 2007). The application of corms with a diameter of 3 cm and a weight of >10 g resulted in the highest flower and stigma weight of saffron (Mollafilabi, 2004).
With respect to the interactive effect of the studied factors, it was observed that plants grown from >12 g corms fertilized with FR4 produced the highest stigma fresh weight of 3.86 g m^{-2} and plants grown from <8 g corms but not fertilized with any food regimes produced the lowest one of 1.1 g m^{-2}.
Overall, the increase in maternal corm weight at higher fertilization levels improved stigma fresh weight (Table 5). The interaction of maternal corm weight and different dosages of cattle manure was significant for flower number, flower fresh and dry weight, and stigma dry weight whose highest values were obtained from the treatment of 7-9 g maternal corm weight and 60 t ha^{-1} manure

(Hassanzadeh Aval et al.,2013). A study in Bam and Gonabad counties of Iran in two growing seasons showed that two-time foliar application of K, Zn, and Fe improved leaf length and flower yield versus no foliar application and as the dosage of the solution was increased to 3 L ha^{-1}, stigma yield and qualitative traits of saffron were enhanced.

Stigma dry weight

According to the results of variance analysis in Table 2, stigma dry weight was influenced by the simple effect of food regime and maternal corm weight at the $p \leq 0.01$ level and by their interaction at the $p \leq 0.05$ level.

The study of the simple effect of food regimes showed that the highest stigma dry weight of 0.67 g m^{-2} was obtained from plants exposed to FR4 showing significant differences with other treatments. This trait was 100 and 24 percent higher in plants exposed to FR 3 and FR2 as compared to control plants, respectively (Table 3). The effect of chemical and biological fertilizers on saffron yield revealed that the maximum yield of stigma and style was related to the application of 150 kg ha^{-1} N in a study in which the effective role of nutrition was reported in enhancing leaf number, leaf length, stigma length, and stigma fresh and dry yield in urea-treated saffron plants (Omidi et al., 2009).

The effect of maternal corm weight on stigma dry weight revealed that the application of >12 g and 8-12 g corms increased this trait by 52 and 26 percent as compared to <8 g corms, respectively (Table 4).

The interaction of these two factors showed that plants grown from >12 g corms and fertilized with FR4 produced the highest stigma dry weight of 0.78 g m^{-2} and those grown from <8 g corms and not fertilized produced the lowest stigma dry weight of 0.22 g m^{-2}. According to the results, in all food regimes, the increase in corm weight from <8 to >12 g improved saffron yield (Table 5). A study on the impact of different levels of humic acid and maternal corm weight on saffron indicated that in the first year, the interactive effect of these two factors was negligible, but in the second year, as the rate of humic acid application and the weight of maternal corms were increased, all saffron yield traits including stigma fresh and dry weight and other traits were improved significantly (Koocheki et al., 2015).

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