Pak. J. Bot., 55(SI): 97-102, 2023. DOI: http://dx.doi.org/10.30848/PJB2023-SI(11)

INVESTIGATING THE IMPACT OF MICROBIAL PLANT ACTIVATOR TREATMENT AT DIFFERENT DOSAGES ON THE QUALITY CHARACTERISTICS OF SAFFRON GROWTH

HASAN ASİL

Hatay Mustafa Kemal University, Altınözü Vocational School of Agricultural Sciences, Medicinal and Aromatic Plants Program, Hatay, Turkey
*Correspondening email: hasan.asil@hotmail.com

Abstract

Activators are products applied to plants or soils to enhance plant productivity and regulate physiological processes. Recently, activators have been used for sustainable crop management. However, few studies have investigated the use of activators for medicinal and aromatic plants and saffron cultivation. Therefore, this study aims to examinexamine the impact of various amounts of Albit, microbial plant activator, on corm and leaf quality focusing on sustainable saffron cultivation. Two separate experiments were conducted to apply activators to corm and leaves, with four different doses (0, 2, 4, 8 mL) used in each application. The weight of the harvested corm indicated that the activator application to the corm was the most effective, resulting in 331.90 g. The highest yield was obtained with the corm x 8 mL treatment, which produced 342.97 g, and the application x treatment interaction, which made346.20 g. When assessing the effect of activator application on saffron pharmacological agents, the highest average amount of safranal was obtained in the corm application with a concentration of 717.68 ppm.

Furthermore, further research is being conducted to develop additional methods for increasing plant resilience to drought stress, which is one of the significant impacts of global climate change. This study on microbial plant activators has revealed the performance components of saffron in dry conditions. In summary, the results of this study suggest that Albit application to corm and leaves has a positive effect on saffron growth and pharmacological properties, highlighting the potential of microbial plant activators for sustainable saffron cultivation. Further research is necessary to optimize the use of activators in different growth conditions and to explore their potential use in other medicinal and aromatic plants.

Key words: Activators, Saffron, Crops; Dose, Dry condition, Microbial plant activators.

Introduction

Saffron (*Crocus sativus* L.) is a sterile triploid plant that can only be propagated vegetatively through its corms (Sameer *et al.*, 2012; Koocheki & Seyyedi, 2015). Therefore, selecting productive corms is critical for saffron production, as studies have shown that corm size positively influences saffron flowering. Although traditional methods are still in use, various studies are underway to increase the number of corms that allow for vegetative propagation and improve quality. These studies aim to replace traditional methods with modern production methods (Ebrahimzadeh *et al.*, 2000; Maggio *et al.*, 2006; Mollafilabi & Shoorideh, 2009; Cinar & Onder, 2019).

In many parts of the world, mineral overload, industrial waste, fertilizers, and pesticides are significant sources of pollution. They contribute to the increased concentration of various heavy metals such as lead, copper, nickel, and other heavy metals in the air, soil, and water (Briffa *et al.*, 2020). These heavy metals can negatively impact the entire ecosystem and have been reported to have harmful health effects on all living organisms (Rostami *et al.*, 2015).

In recent years, efforts have been made to find non-toxic and technological alternatives that reduce the use of chemical pesticides. One of these alternatives is immune inducers, which are agents that can be broken down into non-biologically and biologically active molecules. Immune inducers can induce systemic acquired immune resistance in plants. The biologically active molecules are recognized by receptors on the surface of plant cells, generatcausing plant immunity and triggering the plant defense mechanism. This is expected to reduce pesticide residues in agricultural products and develop high-quality and healthy products (Dewen *et al.*, 2017).

Plant immune attractants promote plant growth, activate the autoimmune system, and improve disease resistance. They align with the requirements of sustainable agricultural development and have become one of the most popular areas for pesticide development (Asil, 2018; Hucheng *et al.,* 2021). These solutions offer promising alternatives to traditional chemical pesticides, potentially improving crop quality and health while reducing the environmental impact.

At the beginning of today's research, topics are microbial activators with multiple effects such as bacterial isolates, active use of microelements, hormone applications, biocontrol against various plant diseases and pests, and resistance to climatic factors and drought stress. Studies suggest that microbial activators can be used as biofertilizer alternatives to chemical fertilizers and pesticides (Olanrewaju *et al.,* 2017; Asil & Ayanoglu, 2018; Strobel, 2018; Backer *et al.,* 2018). Plant activators, including disease defense inducers, are increasingly used in agriculture. A plant activator is the application of a substance or microorganism to plants to increase nutritional efficiency, abiotic stress tolerance, and plant quality traits, regardless of nutrient content (La Spada *et al.*, 2021).

In recent years, activators have gained attention for their potential in sustainable crop management. However, there is a lack of research on the use of activators for medicinal and aromatic plants, including saffron. This study aimed to investigate the effect of different doses of Albit, a microbial plant activator, on corm and leaf quality and stigma quality in saffron cultivation. The goal was to explore the potential of Albit as a tool for sustainable saffron production.

Materials and Methods

Obtaining the samples: This study was conducted between October 2019 and May 2021 in the Hassa district of Hatay province. The saffron corms used in the experimental area had an average weight of 6 g (between 5-7 g) and 60 g per plot. The microbial plant activator used was the Albit® brand with the following technical characteristics: Albit®, a plant growth regulator and bio fungicide, reportedly improved seedling germination and viability, vegetative growth, and accelerated maturation and yield of cereals and vegetables (Albit Biotechnology LLC, 2008). Albit® TPS is a hydrolysate of microorganisms (*Bacillus megatherium*, *Pseudomonas putrefacient*), microelements, growth agents (poly-beta-hydroxybutyrate (6.2 g PHB L^{-1}), and terpenes extracted from plant shoots (Zlotnikov, 2006). Albit® was developed as an alternative to less environmentally friendly synthetic fertilizers and fungicides. Safranal ≥90% stabilized (W338907-Sample-K), crocetin dialdehyde (18804-10 MG), and crocin (17304-1G) were purchased from Sigma Aldrich and were used as received.

Plant material and its growing: Two separate experiments were conducted where the microbial plant activator was applied to the corm and leaves separately. Each application involved using four different doses (0, 2, 4, and 8 mL) of the microbial plant activator. In the corm application, each of the four doses was added to 500 mL of water, and the corms were placed in the water for half an hour. For the foliar application, once the plants reached a leaf length of 10 cm, each of the four doses was added to 500 mL of water and sprayed on the leaves. The experiment was laid out in a randomized plot design with three replicates, two applications (corm and foliar), and four different doses. In each plot, 10 corms were planted at a depth of 10 cm and were supplied with water after planting. Under field conditions, no further irrigation was applied, and weed control was done by hand. No pesticides or fertilizers were used in the experiment.

Preparation of standard substances and extraction procedure: Standard solutions of safranal, crocin, and crocetin were prepared in ethanol with a concentration of 0.40 mg/mL, which were then diluted to concentrations ranging from 10 to 2,000 ng/mL and stored at four °C. An ultrasound-assisted solvent extraction method was used to remove the saffron stigma samples. 40 mg of the stigma was ground and mixed with 10 mL of ethanol in a flask. The mixture was sonicated in an ultrasonic bath for 15 minutes and then centrifuged at 5000 rpm for 3 minutes. This process was repeated three times, and the supernatant was collected in another flask. The solvent mixture was evaporated to a final volume of 1 mL, and the extracts were stored at +4°C in the dark for GC-MS FID analysis (Asil, 2021a).

Gas chromatography-mass spectroscopy (GC-MS FID) analysis: GC-MS analysis was carried out using Hewlett-Packard 6890 series GC-MS analyzer in Hatay Mustafa Kemal University Central Research Laboratory. The column of the device is HP-88 fused silica column (100 m 0.25 mm i.d. film thickness 0.25 µm), and the detector is Hewlett-Packard mass selective detector 6890. GC-MS analysis was performed according to the procedure specified in the literature. The oven is heated to 90°C and waited 1 minute at this temperature. Then, the temperature is increased by 15 degrees per minute to 175ºC and kept for 15 minutes, then the temperature increased by 5 degrees per minute to 225ºC and waited for 5 minutes. Finally, the temperature increased by 10 degrees per minute to 255ºC, and we waited 10 minutes. For flame formation, 60 mL/min H_2 (UHP grade), 400 mL/min air (zero degree), and Helium (99.99%) as a carrier gas, and 10 mL/min flow rate were used as a gas mixture. The injector temperature was 200ºC (Asil, 2021a).

Experimental sites and meteorological conditions: The soil properties of Hassa are slightly alkaline (pH 7.75), salt-free (0.032%), low organic matter (0.78%), medium content of phosphorus (66.05 kg ha^{-1}), low content of potassium (25.8 kg ha^{-1}). The pH of the soil is slightly acidic, and the organic matter is insufficient. The soil contains high amounts of iron, phosphorus, magnesium, and sodium and enough copper, zinc, calcium, manganese, and potassium. The soil structure is loamy and desirable for agricultural activities, soil water, and air balance.

Hatay receives an average of 890 mm of precipitation annually. The average total rainfall in the first year of the study is 1120 mm, and 569 mm in the second year. The first year was a semi-arid climate with abundant precipitation. The average relative humidity was slightly lower in the experimental and autumn months, while increased humidity was observed in the summer and winter months. The relative humidity ranged from 55 to 95%.

Data analysis

Statistical analyzes were made according to the transformed data. The variance analysis of the mean values of the characters obtained from the experiment was createdcreated with the MSTAT-C statistical package program. The Duncan test determined the differences between the standards (Asil *et al.,* 2023).

Results and Discussion

The analysis of variance (ANOVA) of the parameters related to the effects of the applications of activator in saffron (*Crocus sativus* L) cultivation is shown in Table 1, and the characteristics studied (application (A) F values, treatment (B) and application x treatment (A×B)).

Table 1. Analysis of variance (ANOVA) of the parameters of the effects of activator application on the corm quality of saffron.

Parameters	F Value (Replication)	F Value (Application)	F Value (Treatment)	F Value (Application x Treatment)	Coefficient of variation
Leaf length (1st Year) (cm)	16.33	101.77 *	1.06	1.96	13.87
Leaf length (2nd Year) (cm)	2.45	37.00**	0.63	6.65*	6.37
Number of leaves (1st Year)	8.59	2.80	2.59	2.94	29.87
Number of leaves (2nd Year)	1.67	0.09	2.14	0.57	35.61
Total number of flowers (pieces parcel^{-1})	10.13	1.11	7085.51**	2.47	19.46
Dried stigma yield (g/block)	5.54	8.22	5.83**	2.64	42.31
Safranal ratio (%)	0.00	0.00	13630.99*	8690.84*	0.56
Safranal amount (ppm)	6.29	5777091.44*	75406.65*	37715.15*	0.38
Crocin amount (ppm	2.76	26307.13 *	2309.64*	4333.14*	1.79
Crocetin amount (ppm)	0.62	10.80	251.45*	102.93*	8.54
Total corm weight (g parcel^{-1})	13.29	2.05	0.95	0.08	12.37
Corm weight < 5 g (g parcel^{-1})	740.42	34.31**	4.22**	1.95	24.66
Corm weight between 5-10 g (g parcel^{-1})	31.63	119.48*	13.13*	1.74	19.35
Corm weight > 10 g (g parcel^{-1})	2.59	9.68	2.36	0.87	33.10
Total number of corms (pieces parcel^{-1})	3.25	19.14**	0.63	3.50**	8.72
Number of corms < 5 g (pieces parcel^{-1})	1.33	8.15	1.89	10.03*	7.26
Number of corms between 5-10 g (pieces parcel^{-1})	31.00	123.93*	12.34*	2.02	20.83
Number of corms > 10 g (pieces parcel^{-1})	1.75	8.26	3.77**	0.92	27.56
Harvested unit corm weight (g)	2.90	8.92	0.80	1.44	11.44
Daughter corm ratio (%)	3.25	19.14**	0.63	3.50 **	8.72

*. **. ^{ns}. It is significant at 0.01 and 0.05 levels. Respectively. And there is no statistical difference between the averages shown with the same letter

Effects on corm quality: The effects of the trial on the corm character and statistical groupings are shown in Table 2. In examining the of the problem on the weight of the harvested corns, it was found that the application to the corm was significant at the 5% level. Treatment applications were found to be lycrucial at the 5% level, and the interaction between application x treatment dose was not statistically significant. Considering the weight of the harvested corm weights, applying the activator to the corm is the highest at 331.90 g. The highest results were obtained for the dose treatment with corm x 8 mL treatment with 342.97 g and the application x treatment interaction with 346.20 g. When the harvested corms are classified by size, the highest value of 115.67 in corms is less than 5 g in the 0-dose treatment. The highest results were obtained with an application of 176.40 and a treatment dose of 8 mL in 5 to 10 g and 136.27 g with a treatment dose in corm weight more significant than 10 g (Table 2). When the harvested plot was evaluated concerning the total number of corms, the application method was statistically significant at the 5% significance level. Treatment doses were statistically significant at the 5% significance level, and the interaction of application x treatment dose was not statistically significant (Table 2).

The highest average values regarding the total number of corms were obtained when 72.75 corms were applied, 70.00 units at a treatment dose of 8 ml, and 78.33 units when corm and a treatment dose of 2 ml were used. When the number of corms harvested according to their corm lengths is evaluated. TThe highest average of 45.33 corm applications was obtained in corms less than 5 g. It was 42.67 for a treatment dose of 0 mL and 49.33 for a treatment dose of 4 mL x 2 mL. The highest results were obtained in 5 to 10 g and 20.50 corm applications. They were obtained with a treatment dose of 22.00 to 8 mL and with an application of 27.00 x 8 mL treatment dose. On average, the highest results were obtained for corms over 10 g with 8.83 foliar applications, 9.33 at 0 mL treatment dose, and 9.33 at foliar applications x 4 mL treatment dose (Table 2).

When the average weight of harvested corms was examined, this averaged 5.29 g for foliar applications. The highest values were obtained with 5.14 g at 0 treatment dose and 5.74 g at 2 mL treatment dose and application on foliar. Looking at the values for the daughter corm, they were 727.50% when applied to the corm. The highest average values were obtained at the 8-mL treatment dose with 700% and the 2-mL treatment x corm application with 783.33% (Table 2).

In the one-year study to investigate the effect of different activators, the plant activator corm treatment yielded 308.3 g, and the foliar treatment yielded 2013.0 g of corms. Regarding the number of corms, 60.4 were obtained in the corm treatment and 46.7 packages in the foliar treatment (Asil, 2021b). A study on the saffron growing cycle reported that it is more valuableand beneficial to harvest corm every two years (Cardone *et al.*, 2021). In the ssurvey of the effects of soil physical and chemical properties on saffron cultivation, it was reported that the corm yield ranged from 21.4 to 35.0 g per plant. The average corm weight ranged from 6.3 to 14.4 g (Cardone *et al.*, 2020). Their survey on the impact of four plant growth regulators on saffron performance reported that the effects on corm yield ranged from 1.89 to 6.1 g m^{-2} (Heidari *et al.*, 2022). When literature studies are compared, there are not many studies of microbial activators in plants. The results obtained are promising concerning the various saffron studies.

Table 2. The effect of activator application on the corm quality of saffron and Duncan groups.

Measured characters	Application	Dozes (mL)				Average
		0	2	4	8	
Total corm weight (g parcel^{-1})	Leaf	353.90 ns	328.91 ns	302.47 ns	339.17 ns	**331.11 ns**
	Corm	353.90 ns	342.03 ns	326.53 ns	346.20 ns	**342.16 ns**
	Average	**353.90 ns**	**335.47 ns**	**314.50 ns**	**342.68 ns**	
Corm weight < 5 g (g parcel^{-1})	Leaf	138.73 ns nss	101.46 ns	70.27 ns	109.30 ns	**104.94 B**
	Corm	138.73 ns	94.87 ns	113.57 ns	81.37 ns	**107.13 A**
	Average	**138.73 a**	**98.16 b**	**91.92 b**	**95.33 b**	
Corm weight between 5-10 g (g parcel^{-1})	Leaf	69.60 ns	112.14 ns	105.27 ns	119.33 ns	**101.59 B**
	Corm	69.60 ns	146.47 ns	151.67 ns	176.40 ns	**136.03 A**
	Average	**69.60 b**	**129.30 a**	**128.47 a**	**147.87 a**	
Corm weight > 10 g (g parcel^{-1})	Leaf	145.57 ns	115.30 ns	126.93 ns	110.53 ns	**124.58 ns**
	Corm	145.57 ns	100.70 ns	61.30 ns	88.43 ns	**99.00 ns**
	Average	**145.57 ns**	**108.00 ns**	**94.12 ns**	**99.48 ns**	
Total number of corms (pieces parcel^{-1})	Leaf	65.33 b	57.33 c	60.00 c	64.67 b	**61.83 B**
	Corm	65.33 b	78.33 a	76.33 a	75.33 a	**73.83 A**
	Average	**65.33**	**67.83**	**68.17**	**70.00**	
Number of corms < 5 g (pieces parcel^{-1})	Leaf	44.33 b	33.33 d	34.67 d	39.33 c	**37.92 ns**
	Corm	44.33 b	49.33 a	47.67 a	41.67 bc	**45.75 ns**
	Average	**44.33**	**41.33**	**41.18**	**40.50**	
Number of corms between 5-10 g (pieces parcel^{-1})	Leaf	10.00 ns	15.67 ns	16.00 ns	17.00 ns	**14.68 B**
	Corm	10.00 ns	22.33 ns	23.67 ns	27.00 ns	**20.75 A**
	Average	**10.00 b**	**19.00 a**	**19.83 a**	**22.00 a**	
Number of corms > 10 g (pieces parcel^{-1})	Leaf	11.00 ns	8.33 ns	9.33 ns	8.33 ns	**9.25 ns**
	Corm	11.00 ns	6.67 ns	5.00 ns	6.67 ns	**7.33 ns**
	Average	**11.00 a**	**7.50 b**	**7.17 b**	**7.50 b**	
Harvested unit corm weight (g)	Leaf	5.14 ns	5.73 ns	5.03 ns	5.25 ns	**5.28 ns**
	Corm	5.14 ns	4.37 ns	4.30 ns	4.60 ns	**4.60 ns**
	Average	**5.14 ns**	**5.05 ns**	**4.66 ns**	**4.92 ns**	
Daughter corm ratio (%)	Leaf	653.33 b	573.33 c	600.00 c	646.67 b	**618.33 B**
	Corm	653.33 b	783.33 a	763.33 a	753.33 a	**738.33 A**
	Average	**653.33**	**678.33**	**681.67**	**700.00**	

Effect on stigma quality: Saffron is an essential, and expensive spice plant. The most commonly used part of saffron in commerce is its stigmas. In the case of the stigmas, both the potency and the pharmacologically active ingredients are at the forefront of the quality criteria. TThis section studied the stigma yield and quality criteria of saffron (Table 3).

The effect on the corm and leaf is investigated for studying the impact of microbial activators on the stigma. Regarding the site of application, the highest number of flowers was obtained when applied on the corm with an average of 17.83 and the highest average of 22.00- and 4-mL treatments according to the treatments used at different doses. In evaluating stigma efficiency, the highest average value of 0.514 mg was obtained in the application of corm, and the highest average value of 0.647 mg was obtained in the treatment method with 4 mL (Table 3).

When investigating the effect of saffron on the pharmacological agents of the study, the highest average amount of safranal, 717.68 ppm, was obtained when the cormorant was applied. Depending on the treatment doses applied. 879.13 ppm and 4 g treatment doses were obtained depending on the treatment doses applied. The highest average value of 1036.17 mg in the bilateral interaction was obtained with a treatment dose of 4 ml and a foliar application. As for crocin content, the highest average value of 69.59 ppm was obtained with corm application, and the highest average value of 75.57 ppm was obtained with the 8 g treatment method. As for the amount of crocetin, the highest average value of 4.48 ppm was obtained in the foliar application. According to the applied treatment doses, the highest value of 7.71 ppm was obtained in the 8-g treatment (Table 3).

In the study on the effects of soil's physical and chemical properties on saffron, the dry weight of stigmas ranged from 0.0039 g to 0.0071 g (Cardone *et al.*, 2020). In the study on the effect of four different plant growth regulators on saffron performance, the average number of flowers ranged from 7.0 to 42.0. In addition, when the effects on stigma weight were studied, the average yield ranged from 0.120 to 0.760 g m^{-2} dry weight (Heidari *et al.*, 2022). The results of the study are consistent with the literature. It has been shown that the efficiency of the scars can be increased if the activator is used in an appropriate dosage.

Effect on leaf characteristics: When examining the effect of activator application on leaf length, it was found that the corm application had a higher leaf length of 42.67

cm in the first year of the study, 42.33 cm with 8 mL gr activator treatment in terms of treatment doses, and 38.33 and 37.17 cm with 8 mL in the highest leaf treatment in the second year obtained from the dose therapy. Considering the number of leaves per plant, the highest number of leaves was 44.25 in the second year of the study, and in terms of treatment doses, the highest treatment dose of 2 mL was 54.67 units (Table 4).

In the literature, the leaf length of saffron is reported to be between 21-24 cm (Asil & Ayanoglu, 2018). In studies on saffron in Hatay conditions, the number of leaves varied between 1.67 and 7.33 pieces/plant according to different corm sizes and planting depth (Yıldırım *et al.*, 2017), and the number of leaves per plant varied between 3.9 and 7.33 pieces. have reported. 24.4 pieces/plant at different onion lengths and storage temperatures (Hajyzadeh *et al.*, 2017). In the study on the effect of soil's physical and chemical properties on saffron, it was found that leaf length ranged from 26.9 to 49.8 cm, and the number of leaves ranged from 20.7 to 27.6 (Cardone *et al.*, 2020). In their study on the effect of four different plant growth regulators on saffron performance, they reported that when the impact on leaf length was investigated, the leaf length ranged from 16.0 to 38.0 cm, and the number of leaves ranged from 4.7 to 10.7 (Heidari *et al.*, 2022). Compared to the studies in the literature, better results were obtained for leaf length and the number of leaves using different dosages of activators.

Table 3. Effect of activator application on stigma quality of saffron and Duncan groups.

Measured characters	Application	Dozes (mL)				Average
		0	2	4	8	
Total number of flowers (pieces parcel^{-1})	Leaf	14.00 ns	17.67 ns	21.00 ns	24.67 ns	**19.33 ns**
	Corm	14.00 ns	19.33 ns	23.00 ns	16.67 ns	**18.25 ns**
	Average	**14.00 c**	**18.50 b**	**22.00 a**	**20.67 ab**	
Stigma yield (mg parcel^{-1})	Leaf	0.217 ns	0.265 ns	0.424 ns	0.614 ns	**0.381 ns**
	Corm	0.217 ns	0.523 ns	0.870 ns	0.489 ns	**0.525 ns**
	Average	**0.217 d**	**0.395 c**	**0.648 a**	**0.552 b**	
Safranal amount (ppm)	Leaf	295.83 f	354.13 e	1.036.17 a	532.00 d	**554.53 B**
	Corm	295.83 f	876.13 c	722.10 d	978.30 b	**718.09 A**
	Average	**295.83 d**	**615.13 c**	**879.13 a**	**755.15 b**	
Crocin amount (ppm)	Leaf	35.70 e	5.13 g	20.07 f	86.03 b	**36.73 B**
	Corm	35.70 e	75.07 c	103.17 a	65.10 d	**69.76 A**
	Average	**35.70 d**	**40.10 c**	**61.62 b**	**75.57 a**	
Crocetin amount (ppm)	Leaf	3.00 e	5.01 c	1.25 f	8.71 a	**4.94 A**
	Corm	3.00 e	1.44 f	4.61 d	6.7 b1	**3.94 B**
	Average	**3.00 b**	**3.22 b**	**2.93 b**	**7.71 a**	

Table 4. Effect of activator application on leaf properties of saffron and Duncan groups.

Measured characters	Application	Dozes (mL)				Average
		0	2	4	8	
Leaf length (cm) (1st year)	Leaf	40.00 ns	34.33 ns	36.67 ns	38.67 ns	**37.42 B**
	Corm	40.00 ns	47.67 ns	37.00 ns	46.00 ns	**42.67 A**
	Average	**40.00 ns**	**41.00 ns**	**36.83 ns**	**42.33 ns**	
Leaf length (cm) (2nd year)	Leaf	36.67 c	36.33 c	39.67 ab	40.67 a	**38.33 A**
	Corm	36.67 c	37.67 bc	35.67 c	31.00 d	**35.25 B**
	Average	**36.67 ns**	**37.00 ns**	**37.17 ns**	**35.83 ns**	
Number of leaves per plant (piece) (1st year)	Leaf	44.33 ns	34.67 ns	29.67 ns	32.00 ns	**35.16 ns**
	Corm	44.33 ns	20.00 ns	52.00 ns	41.67 ns	**39.50 ns**
	Average	**44.33 ns**	**27.33 ns**	**40.83 ns**	**36.83 ns**	
Number of leaves per plant (piece) (2nd year)	Leaf	28.00 ns	62.33 ns	49.67 ns	37.00 ns	**44.25 ns**
	Corm	28.00 ns	47.00 ns	50.67 ns	45.00 ns	**42.67 ns**
	Average	**28.00 ns**	**54.67 ns**	**50.17 ns**	**41.00 ns**	

Conclusion

In this study, stigma yield and safranal content are significant for the spice value of saffron. The best results were obtained for stigma yield and safranal value, and the best results were obtained with the application in the corm and a treatment dose of 4 ml. Further applications and methods are being developed to increase plant resilience to drought stress, one of the jorsignificant impacts of global climate change. There is little research on saffron and bulbous plants on this topic. It has been concluded that activators will support the plant under stress conditions. In addition, this study on the use of activators in bulbous plants is preliminary. It is thought that it will guide the researchers and contribute to creating an alternative model.

References

Asil, H. 2021a. Evaluation of the effects of different storage times on pharmacological agents of Saffron (*Crocus sativus* L.) (safranal. crocin and crocetin) and their quality characteristics. *Celal Bayar Üniv. Fen Bilim. Derg.,* 8(2): 263-269.

Asil, H. 2021b. The effect of different biostimulants applications on corm characters of saffron (*Crocus Sativus* L.). Academic Reseach in Life Sciences for Sustainibility. Publisher: Published by Artikel Akademi: Istanbul, Turkey, 123-135

Asil, H. and F. Ayanoglu. 2018. The effects of different gibberellic acid doses and corm cutting methods on saffron (*Crocus sativus* L.) yield components in Turkey. *Fresenius Environ. Bull.*, 27(12 A): 9222-9229.

Asil, H., F. Celik, S. Tasgin, M. Celik and I. Uremis. 2023. Effects of some weed control methods on stigma in saffron (*Crocus sativus* L.) cultivation. *JAST*, 25(1): 115-124.

Asil. H. 2018. GC-MS analysis of volatile components of Safranbolu and Kirikhan saffron (*Crocus sativus* L.) prepared by ultrasonic extraction. *Fresenius Environ. Bull.*, 27(12 B): 9557-9563

Backer, R., J.S. Rokem, G. Ilangumaran, J. Lamont, D. Praslickova, E. Ricci and D.L. Smith. 2018. Plant growth-promoting rhizobacteria: context, mechanisms of action, and roadmap to commercialization of biostimulants for sustainable agriculture. *Front. Plant Sci.*, 1473.

Briffa, J., E. Sinagra and R. Blundell. 2020. Heavy metal pollution in the environment and their toxicological effects on humans. *Heliyon*, 6(9): e04691.

Cardone, L., D. Castronuovo, M. Perniola, L. Scrano, N. Cicco and V. Candido. 2020. The influence of soil physical and chemical properties on saffron (*Crocus sativus* L.) growth, yield and quality. *Agronomy*, 10(8): 1154.

Cardone, L., D. Castronuovo, M. Perniola, N. Cicco and V. Candido. 2020. Saffron (*Crocus sativus* L.), the king of spices: *An overview. Sci. Hort*., 272: 109560. https://doi.org/10.1016/j.scienta.2020.109560

Cardone, L., V. Candido, D. Castronuovo, M. Perniola and N. Cicco. 2021. Comparing annual and biennial crop cycle on the growth, yield and quality of saffron using three corm dimensions. *Sci. Hort*., 288: 110393.

Cinar, A.S. and A. Onder. 2019. Anadolu'nun kültürel mirası: *Crocus sativus* L. (Safran). *J. Pharm. Sci.*, 44(1): 79-88.

Dewen, Q., D. Yijie, Z. Yi, L. Shupeng and S. Fachao. 2017. Plant immunity inducer development and application. *Mol. Plant Microb. Interac.*, 30(5): 355-360.

Ebrahimzadeh, H., T. Radjabian and R. Karamian. 2000. In vitro production of floral buds and stigma-like structures on floral organs of *Crocus sativus* L. *Pak. J. Bot.,* 32(1): 141-150.

Hajyzadeh, M., H. Asil, M.U. Yildirim, E.O. Sarihan, F. Ayanoglu and K.M. Khawar. 2017. Evaluating effects of corm circumference and storage temperatures on yield and yield components of saffron at different elevations. *Acta Hort*., 1184: 39-46.

Heidari, F., F. Shekari, B. Andalibi, J. Saba, D. Uberti and A. Mastinu. 2022. Comparative effects of four plant growth regulators on yield and field performance of *Crocus sativus* L. *Hort.,* 8(9): 799.

Hucheng, D., F. Meiyan, Z. Wen, Z. Liying, X. Xinhui, L. Yilin and W. Dailun. 2021. The pores evolution of lacustrine shale induced by smectite-to-illite conversion and hydrocarbon generation: upper Triassic Yanchang Formation, Ordos Basin, China. *J. Petrol Sci. Eng.,* 202: 108460.

Koocheki, A. and S.M. Seyyedi. 2015. Relationship between nitrogen and phosphorus use efficiency in saffron (*Crocus sativus* L.) as affected by mother corm size and fertilization. *Ind. Crops Prod.,* 71: 128-137.

La Spada, F., F. Aloi, M. Coniglione, A. Pane and S.O. Cacciola. 2021. Natural biostimulants elicit plant immune system in an integrated management strategy of the postharvest green mold of orange fruits incited by *Penicillium digitatum*. *Front Plant Sci.*, 12: 1149.

Maggio, A., G. Raimondi, A. Martino and S. De Pascale. 2006. Soilless cultivation of saffron in Mediterranean environment. In: III International Symposium on Models for Plant Growth, Environmental Control and Farm Management in Protected Cultivation, 718 pp. 515-522.

Mollafilabi, A., M.H. Aslami and H. Shoorideh. 2009. Replacement of Saffron (*Crocus sativus* L.) with Poppy (*Papaver somniferum* L.) and its Socio-economic Results in Afghanistan. In: III International Symposium on Saffron: Forthcoming Challenges in Cultivation, Research and Economics, 850 pp. 299-302.

Olanrewaju, O.S., B.R. Glick and O.O. Babalola. 2017. Mechanisms of action of plant growth promoting bacteria. *World J. Microbiol. Biotechnol.*, 33(11): 1-16.

Rostami, M., R. Karamian and Z. Joulaei. 2015. Effect of different heavy metals on physiological traits of saffron (*Crocus sativus* L.). *Saffron Agron. Technol.*, 3(2): 83-96.

Sameer, S.S., S. Bashir, F.A. Nehvi, A.M. Iqbal, S. Naseer, S.A. Nagoo and N.A. Dar. 2012. Effect of biofertilizers, biological control agents and soil amendments on the control of saffron corm rot (*Crocus sativus* L.). In: *IV International Symposium on Saffron Biology and Technology*, 1200 pp. 121-124.

Strobel, G. 2018. The emergence of endophytic microbes and their biological promise. *J. Fungi.*, 4(2): 57.

Yildirim, M.U., H. Asil, M. Hajyzadeh, E.O. Sarihan and K.M. Khawar. 2017. Effect of changes in planting depths of saffron (*Crocus sativus* L.) corms and determining their agronomic characteristics under warm and temperate (Csa) climatic conditions of Turkish province of Hatay. *Acta Hort*., 1184: 47-54.

(Received for publication 27 August 2022)