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Original Article

The Phytochemical and Morpho-Physiological Response of Saffron (*Crocus sativus*) to Different Summer Irrigation Regimes

Hamed Eslamia[1], Hossein Salehi-Arjmand[1]*, Reza Shahhoseinia[1], Morteza Akramian[1] and Iman Mirmazloum[2]

[1]*Department of Medicinal Plants, Arak University, Arak, Iran*
[2]*Department of Plant Physiology and Ecology, Hungarian University of Life Sciences, Budapest, Hungary*

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***Corresponding author**
Salehiarjmand66@gmail.com

ABSTRACT

This study aimed to investigate the effect of summer irrigation on the phytochemical and morpho-physiological traits of Iranian saffron. In this paper, first, four irrigation regimes, including control (without summer irrigation), irrigation in early July, irrigation in early August, and irrigation in early September, were conducted using a randomized complete block design with four replications in a research farm in Khomein County, Markazi Province, Iran (2020-2021). Based on this, the data encompasses the information collection concerning flowering and leafing time, stigma yield, vegetative growth rate, and phytochemical compounds of stigma, specifically crocin, safranal, and picrocrocin. The results demonstrated that summer irrigation had significant effects on assessed characteristics. Additionally, summer irrigation enhanced saffron growth rate and stigma yield ($p\leq 0.05$). Furthermore, the irrigation in early August accelerated flowering time and prolonged the flowering period from 13.5 to 16 days. The highest dry weight of saffron stigmas and the best stigma yield was 12.08 g/kg of flower fresh weight and 7.08 kg/h, respectively, and the optimal levels of crocin, picrocrocin, and safranal content obtained from irrigated plants in early August. Therefore, irrigation in early September resulted in earlier leaf occurrence before flowering, with the highest leaf dry weight (274 g m2) and leaf length (39 cm). The findings showed that the applied summer irrigation, specifically in early August, was an effective and economically viable practice to enhance saffron yield and stigma quality. This irrigation treatment is easily applicable and would be recommended to producers for its potential economic benefits.

INTRODUCTION

Saffron (*Crocus sativus* L.) is an herbaceous flowering plant of the Iridaceae family, typically cultivated in Iran, Afghanistan, India, Spain, and Italy [1]. The high value of saffron plants lies in the dried red stigmas of its flowers, known as saffron, which is considered the world's most expensive spice and is widely used in the pharmaceutical and food industries due to its distinctive color and fragrance [2]. The remarkable health-promoting properties of saffron are attributed to its unique phytochemical composition within the dried stigmas. The quality of saffron stigma is linked to singular and marker secondary metabolites including crocin (water-soluble crocetin esters), picrocrocin (a monoterpene glycoside and a precursor of safranal), and safranal (a monoterpenoid) that determines the color, taste, and odor of saffron, respectively [3]. Recent studies suggest incorporating saffron into the human diet for the prevention of chronic diseases and to lower the risk of cancer [4, 5]. Saffron is a triploid and sterile plant that does not produce seeds and propagates through corms [6]. Due to its unique life cycle and distinctive biological and morphological features, such as low water requirement and thick leaves, saffron plants can adapt and thrive in arid and semi-arid climates [7, 8]. The saffron lifecycle initiates with fall flowering, during which one to seven deep-purple-colored flowers appear from each corm. The flowers bear three stigmas, each 25-30 mm long, which droop over the petals. Additionally, the flowers contain three yellow stamens with phytochemical compounds of no considerable significance. The vegetative organs of saffron develop during the autumn, and the growing season concludes in spring when producing new corms. Although saffron flowers in autumn, flower induction occurs during the warm and arid summer conditions when the corms exit the dormant phase [9]. Many efforts have been made to improve the yield and quality of saffron stigma by applying different agronomic practices, especially fertilization [10- 12] and irrigation [13, 14]. Saffron plants typically require irrigation at the end of summer, often coinciding with seasonal rainfall that compensates for the necessary irrigation [15]. Recommendations for improving saffron production management include irrigation at six distinct stages: I) summer irrigation, II) early October, III) in November, IV) late December-early January, V) in early March, and VI) in early April [16]. A significant part of the saffron growing season is known as the dormant phase (May-October), characterized by the absence of vegetative growth, while flowering is induced during this period. Soil moisture becomes a critical factor in flower induction and its efficiency during the summer [17]. Flowering induction, and consequently the development and quality of saffron flowers, can be influenced by environmental conditions and cultivation management [18, 19]. Earlier reports have indicated the positive impact of summer irrigation on saffron stigma yield and quality [20-23]. However, there are inconsistencies among the reported results, particularly concerning the recommended time for summer irrigation. Concerns have been raised about the risks of increased corm rot, typically caused by *Rhizoglyphus robini* mites or pathogenic fungi due to excessive moisture accumulation in the rhizosphere [24]. Therefore, in determining the appropriate strategy for summer irrigation, several factors, such as geographical and climatic conditions, pathogenicity rates in the region, and soil characteristics should be carefully considered when recommending irrigation schedules. The present study was conducted to evaluate Summer Irrigation Regimes on flowering time, vegetative growth, yield, and phytochemical compounds of produced saffron stigma, compared with without irrigated saffron plants.

MATERIALS AND METHODS

The Climatic Condition of the Experiment Site

The study was conducted in four-year-old saffron fields over two growing seasons of 2020 and 2021 in Khomein County, Markazi province, Iran. Khomein is located at 37° 37' N, 50° 05' E, and an altitude of 1834.6 m above sea level, with experiencing cold winters and moderate to hot summers. The annual average rainfall and the temperature in the experiment site were 306.8 mm and 13.6 °C, with a yearly average humidity of 51% (2020) and 50% (2021). The soil physicochemical properties of the experimental farm are presented in Table 1.

The Applied Treatments and Experiment Design

The experiment was designed as a randomized complete block design (RCBD) with four replications. Each block had four (4 ×1.5m) plots. The summer irrigation regimes included a control (no summer irrigation), irrigation in early July, early August, and early September. In each irrigation treatment, 400 m^3/ha water was applied using the basin irrigation method, with a flow rate of 1.26 m^3/s (Fig. 1). The field preparation and irrigation treatments are presented in Table 1.

The Assayed Traits

Flowering and Leafing Time

During our daily visits in autumn, we recorded the time of flowering and leafing for each treatment plot. We measured the number of emerged flowers before the leaves’ appearance and recorded the daily leaf growth rate in centimeters until the conclusion of the experiment in November. At the end of the flowering period (when no new flowers appeared for 3-4 consecutive days), we calculated the length of the flowering period for each treatment plot.

Saffron Stigma Yield

Upon full blooming, the flowers of each plot were hand-picked every morning, and the stigma and style were separated from the flowers when the stigmas were detached from the style. The fresh stigmas of each plot were spread out to dry on white cotton cloth in the shade at room temperature. After four days, the semi-dried stigmas were grouped in bunches and kept under the same conditions for another three days. The dry weight of stigmas collected from each treatment plot was measured before the samples were packed in aluminum boxes and transferred to the laboratory for phytochemical analysis.

Leaf Dry Weight and Leaf Length

At the end of the experimental period, the leaves of six individual plants from each treatment plot were harvested to measure leaf length using a ruler, and the results were presented in cm. The collected leave samples were placed in paper bags labeled for each treatment plot and dried in an oven at a temperature of 70 °C for three days to obtain the dry weight of the leaves. The weight was measured on a laboratory scale and expressed in g/m^2.

Phytochemical Analysis

The biomarker compounds of color (crocin), flavor (picrocrocin), and fragrance (safranal) in dried stigma were assayed according to the ISO method (ISO3632). In this procedure, 900 mL of distilled water was combined with 500 mg of dried stigma homogenous powder and stirred at 1000 rpm for one hour using a magnetic stirrer under dark, room temperature conditions. The resulting solution was then adjusted to a final volume of 1000 mL with distilled water. Subsequently, 20 mL sample solutions were transferred to volumetric flasks and diluted to 1000 mL with distilled water while shaking moderately. The solution was quickly filtered using filter paper in the absence of light to obtain a clear solution. Finally, the absorbance of filtrate samples was measured by an 1800 UV–VIS spectrophotometer (Shimadzu Inc., Kyoto, Japan) at the designated wavelengths of 257, 330, and 440 nm for picrocrocin, safranal, and crocin, respectively. The maximum absorbance of each compound in 1% aqueous solution of dried stigma was calculated based on the equation suggested by Sereshti *et al*. 2018 [25].

Statistical Analysis

The variance analysis of the recorded data was conducted using the statistical analysis program (SPSS ver.16.0). The normality of the residuals was confirmed by the Shapiro-Wilk's test ($p>0.05$). Subsequently, the data were analyzed using one-way ANOVA and Duncan's multiple range test at ($p\leq0.05$). Charts were prepared using MS Excel 2017 for presentation purposes.

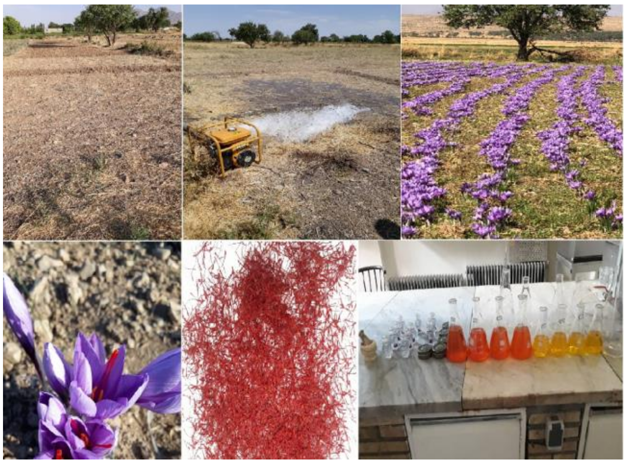

Fig. 1 Pictures related to some stages of research implementation

RESULTS

Analysis of Variance

The results of variance analysis showed that the effect of summer irrigation on the number of flowering days before leafing, the number of leafing days before the flowering, and the length of a flowering period were significant at $p\leq0.05$ (Table 2).

Table 1 The properties of soil and the applied irrigation water (ISP-OES)

Soil properties	Unit	Values	Water analysis	Unit	Values
Sand	%	41	EC	Mmhos/cm	718
Clay	%	21	T.D.S	Mg/l	396.1
Silt	%	38	pH		7.9
EC	Ms/m	3.38	Cl^-	ppm	81
pH		7.8	NO_{3-}	ppm	135
OC	%	1.17	Mg^{++}	ppm	146
N_{total}	%	2.1	Ca^{++}	ppm	244
P_{ava}	ppm	15.9	$MgCl_2$	ppm	565
K_{ava}	ppm	138	$CaCl_2$	ppm	634
Fe	ppm	9.11	Na_2SO_4	ppm	249
B	ppm	6.23	$MgSO_4$	ppm	214
Mg	ppm	7.04	$CaSO_4$	ppm	782
Cu	ppm	2.47	-	-	-

Flowering and Leafing Time

Summer irrigation accelerated the flowering time of saffron compared to non-irrigated plants. Only in irrigated saffron in early August did flowering start before leafing, occurring five days before the appearance of the leaves. In early July, early September, and non-irrigated (control) conditions, leaf growth was observed at 4.25, 8.5, and 3.5 days before flowering, respectively (Table 3). The results indicate that the most extended flowering period (16 days) occurred with the irrigation treatment in early August, significantly higher ($p \leq 0.05$) than the other irrigation dates. Conversely, the shortest flowering duration (11.25 days) was observed in the plants irrigated in early September (Table 3).

Stigma Yield

There were significant differences in the case of dry weight and yield of stigma (Table 2). The stigma dry weight ranged between 12.08 and 10.85 g/kg flower fresh weight. The collected stigma from the irrigated saffron plants in early August showed the highest dry weight was 15.32% higher than the non-irrigated plants. However, the irrigation in early September decreased the stigma dry weight of saffron plants by 2.25% compared with without irrigation treatment (Table 4). Also, the highest (7.08 kg/h) and lowest (5.45 kg/h) stigma yield was obtained in the early August and early September irrigation, respectively (Table 4).

Leaf Dry Weight and Leaf Length

According to the statistical analysis, summer irrigation significantly influenced the leaf dry weight and leaf length of saffron plants (Table 2). The highest leaf dry weight (274 g.m-2) was recorded in irrigated plants in early September, significantly higher than the other irrigation treatments. Irrigated plants in early August had the lowest leaf dry weight (240 g.m-2), which did not differ significantly from the control treatment (without irrigation). The leaf length of saffron plants varied between 39 and 30.75 cm, with irrigated plants in early September and early July producing the larger leaves (Table 4).

Phytochemical Compounds

In the present study, the content of crocin, picrocrocin, and safranal of saffron dried stigma was remarkably influenced by summer irrigation (Table 5). The highest quantity of crocin (264.6 A1%/440) was detected in saffron plants irrigated in early August, significantly higher than plants under the other irrigation treatments. The lowest amount of crocin (258.6 A1%/440) was recorded in the control treatment without irrigation (Table 6).

Table 2 The mean squares of summer irrigation affect flowering traits, and stigma yield and leave the growth of the saffron plant

Sources	df	MS						
		Days of leaf growth before the flowering	Days of flowering before leaf growth	Days of Flowering period	Stigma yield	Stigma yield	Leaves yield	Leaves length
Replication	3	0.229 ns	0.167	0.500	0.011	0.006	1.560	1.890
irrigation	3	48.700 **	25.00 **	15.170 **	1.12 **	1.96 **	1109.9 **	62.56 **
Error	9	0.229	0.167	0.556	0.010	0.010	26.560	2.78

** Significant at 1% probability levels

Table 3 The effect of different summer irrigation regimes on flowering and leafing time of saffron plant

Summer irrigation	Days of leaf growth before flowering	Days of flowering before leaf growth	Days of flowering period
Non-irrigated	3.50±0.58 b	-	13.50±0.58 b
Early July	4.25±0.50 b	-	13.25±0.50 b
Early August	5.00±0.82 a	-	16.00±0.82 a
Early September	8.50±0.58 a	-	11.25±0.96 c

Data are mean ± standard deviations (*n*=6). Mean values with different letters in a column are significantly different at a 5% level probability according to the LSD test ($p < 0.05$)

Table 4 The effect of different summer irrigation regimes on stigma yield and leaf growth of saffron plant

Summer irrigation	Dry weight of stigma (g/kg FW)	Stigma yield (kg/h)	Leaf dry weight (g/m^2)	Leaf length (cm)
Non-irrigated	11.10±0.08 c	5.80±0.08 c	242.50±2.89 c	33.00±2.45 b
Early July	11.40±0.08 b	6.00±0.08 b	265.00±5.77 b	38.00±1.41 a
Early August	12.08±0.10 a	7.08±0.10 a	240.00±4.08 c	30.75±0.96 b
Early September	10.85±0.13 d	5.45±0.13 d	273.75±4.79 a	39.00±1.15 a

Data are mean ± standard deviations (*n*=6). Mean values with different letters in a column are significantly different at a 5% level probability according to the LSD test ($p < 0.05$)

Table 5 The mean squares of summer irrigation effects on crocin, picrocrocin, and safranal content of saffron-dried stigma

Source of variation	df	MS		
		Crocin	Picrocrocin	Safranal
Replication	3	1.750	0.920	0.500
Summer irrigation	3	29.080 **	12.420 **	1.500 **
Error	9	0.806	0.639	0.229

ns and ** are for non-significant and significant differences at 1% probability ($p < 0.01$), respectively

Table 6 The effect of different summer irrigation regimes on the content of crocin, picrocrocin, and safranal of saffron-dried stigma

Summer irrigation	Crocin (264.6 A1%/440)	Picrocrocin (129.2 A1%/257)	Safranal (53.5 A1%/330)
Non-irrigated	258.60±3.72 c	125.60±2.21 b	51.50±0.45 b
Early July	261.40±4.18 b	125.50±2.17 b	52.25±0.37 b
Early August	264.60±4.51 a	129.20±2.31 a	53.50±0.48 a
Early September	260.30±4.63 b	125.30±2.09 b	51.50±0.43 b

Data are mean ± standard deviations (*n*=6). Mean values with different letters in a column are significantly different at a 5% level probability according to the LSD test ($p < 0.05$)

Irrigated plants in early August produced the highest quantity of picrocrocin (129.2 A1%/257) and safranal (53.5 A1%/330). There was no significant difference between irrigated plants in early July and early September compared to the non-irrigated plants concerning the flavor and fragrance characteristics of saffron stigma.

DISCUSSION

The flower induction of saffron occurs during the corm dormancy stage, and as reported by Wang *et al*. 2021 [18] and Zhou *et al*. 2022 [19], the initiation and emergence of saffron flowers depend on environmental conditions. The present study was conducted to assess the effect of summer irrigation on flowering-associated traits of saffron. The results demonstrated that summer irrigation significantly affected the vegetative growth, quantity, and quality of the produced saffron stigmas. Specifically, in the saffron corms irrigated in early August, flowering occurred earlier than leafing and displayed the longest flowering period in this treatment. In the other summer irrigation times (early July, early September) and without irrigation, leafing appeared before flowering. The flowering process is regulated by a complex and sensitive molecular network connected with different environmental signals such as humidity, temperature, and photoperiod [17].

A decrease in soil temperature due to summer irrigation leads to the required low humidity for saffron corms, thereby enhancing flower induction [13, 14]. This primes conditions for saffron flowering, which leads to the production of more and larger flowers, ultimately leading to a higher stigma yield [11]. According to the data obtained from the current experiment, the stigma yield and quality significantly varied due to different irrigation times during the summer season. The highest dry weight and yield of stigmas observed in the saffron plant irrigated in early August. However, plants receiving irrigation in early September exhibited an earlier leafing time compared to other treatments, leading to the lowest flowering time and the highest vegetative growth, as evidenced by the dry weight and length of the leaves.

Our findings are in line with Bicharanloo *et al*. 2020 [21] and Behdani *et al*. 2008 [26], who reported that the yield of dried stigma in summer-irrigated saffron corms was higher than that of the non-irrigated ones. Similarly, the results of Naderi *et al.* 2017 [20] also confirmed that summer irrigation of saffron in August significantly increased the stigma yield and improved the weight and number of produced corms, while summer irrigation in July did not have significant effects on saffron growth and stigma yield. Feizi *et al*. 2015 [23] investigated the summer irrigation effect on flowering traits of saffron and reported an increased number of flowers and stigma yield in plants irrigated in August. Gresta *et al*. 2008 [27] reported that saffron irrigation at the beginning of September resulted in an earlier flowering time, while the stigma yield in the irrigated corms in late August was significantly higher than that in non-irrigated plants.

In contrast to the findings of the majority of research, Seghatoleslami and Sabzekar 2017 [28] stated that the negative effect of spring and summer irrigation on saffron tuber decay was more significant than its positive effect on stigma yield. These results underscore the importance of conducting experiments on summer irrigation to determine the optimal timing, which is contingent upon specific geographical and climatic factors, to enhance both the quantity and quality of saffron overall.

The saffron life cycle begins in early autumn with the emergence of leaves, flowers, or both, depending on soil moisture and temperature [9]. The development and differentiation process in saffron dormant corms results from mitosis in the apical meristem of the dormant corms, coinciding with decreasing humidity and increasing summer temperature [8]. Humidity is one of the most critical factors in decomposition and mineralization associated with soil microbial activity [10]. Summer irrigation for saffron provides favorable moisture and appropriate temperature for the decomposition of soil organic matters that affect the development and differentiation process of saffron vegetative and reproductive organs [29]. It seems that the lower yield and dry weight of stigma in the irrigated saffron in early July was related to the co-occurrence of water supply and differentiation of vegetative organs. However, irrigation in early August occurred at the reproductive organs differentiation phase leading to a subsequent higher stigma yield. The vegetative and reproductive organs of saffron were thought to be differentiated until September so that irrigation in early September did not affect the stigma production. According to the results of Pirasteh-Anosheh *et al*. 2022 [29] and Aghhavani Shajari *et al*. 2020 [30] soil moisture at flower initiation time is the main factor that determines the induced flower quantity and quality from the saffron corms. In a recent study, Pirasteh-Anosheh and coworkers 2023 [31] investigated an extensive collection of factors infusing the saffron yield in different geographical locations. Although they could find a positive correlation between the precipitation rate at the early growing season (before flowering) and the higher yield, a decreased stigma yield was observed in an area with a higher precipitation rate. The authors assumed that the constant precipitation without a long enough drought interval could harm the final yield [31].

The phytochemical analysis of saffron stigma showed a significant effect of summer irrigation on crocin, picrocrocin, and safranal content (Table 6). The highest quality saffron stigma was harvested from irrigated plants in early August. The obtained stigmas from the other treatments did not have considerable differences compared with the non-irrigated plants concerning color, flavor, and fragrance attributes. The effect of summer irrigation on saffron quality was previously reported by Koocheki and Seyyedi 2016 [22]. Aghhavani Shajari *et al*. (2022) have highlighted the significant impact of environmental conditions on the phytochemical composition of saffron. Their research emphasizes that effective irrigation management plays a crucial role in enhancing the quality of saffron stigmas. Environmental conditions such as day and night temperatures and water availability during the flower development process can influence the expression of responsible genes and the activity of involved enzymes in biosynthesis pathways of crocin, picrocrocin, and safranal changes the saffron quality [15].

CONCLUSION

The production of saffron is subject to various climatic factors, with field management playing a significant role in its cultivation. The findings indicate that summer irrigation significantly impacts the vegetative growth, flowering time, stigma yield, and phytochemical properties of saffron plants and their stigmas. Timing summer irrigation correctly is crucial, as it can enhance both the quantitative and qualitative characteristics of the produced stigmas.

Early August irrigation notably accelerated the flowering time and prolonged the flowering period. Saffron plants irrigated in early August yielded the highest quality stigmas with the highest crocin, picrocrocin, and safranal content. Furthermore, early September irrigation led to the highest leaf dry weight and length, initiating leaf growth before flowering. Implementing early August irrigation resulted in a significant increase in stigma yield by over one kilogram per hectare. Therefore, it is recommended for saffron farmers to enhance efficiency and productivity, leading to increased yield and income.

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REFERENCES

1. Cardone L., Castronuovo D., Perniola M., Cicco N., Candidob V. Saffron (*Crocus sativus* L.), the king of spices: An overview. Sci Hortic. 2020;272:109560.
2. Ahmad Bakshi, R., Singh Sodhi N., Ahmed Wani I., Showkat Khan Z., Dhillon B., Gani A. Bioactive constituents of saffron plant: Extraction, encapsulation and their food and pharmaceutical applications. Appl. Food Res. 2022; 2:100076.
3. Avila-Sosa R., Nevarez-Moorillon G.V., Ochoa-Velasco C.E., Navarro-Cruz A.R., Hernandez-Carranza P., Cid-Perez T.S. Detection of saffron's main bioactive compounds and their relationship with commercial quality. Foods. 2022; 11: 3245.
4. Lahmass I., Ouahhoud S., Elmansuri M., Sabouni A., Elyoubi M., Benabbas R., Choukri M., Saalaoui E. Determination of antioxidant properties of six by-products of *Crocus sativus* L. (saffron) plant products. Waste Biomass Valorization. 2018; 9:1349-1357.
5. Moratalla-Lopez N., Bagur M.J., Lorenzo C., Martinez-Navarro M.E., Salinas M.R., Alonso G.L. Bioactivity and bioavailability of the major metabolites of *Crocus sativus* L. flower. Molecules. 2019;24:2827.
6. Nemati Z., Harpke D., Gemicioglu A., Kerndorff H., Blattner F.R. Saffron (*Crocus sativus*) is an autotriploid that evolved in Attica (Greece) from wild *Crocus cartwrightianus*. Mol. Phylogenet. Evol. 2019;136:14-20.
7. Hajihashemi S. Characterization of exogenous nitric oxide effect on *Crocus sativus* response to different irrigation regimes. J. Plant Growth Regul. 2020;40:1510-1520.
8. Dewir Y.H., Alsadon A., Ibrahim A., El-Mahrouk M. Effects of growing substrate, mode of nutrient supply, and saffron corm size on flowering, growth, photosynthetic competence, and cormlet formation in hydroponics. Hort Technol. 2022; 32:234-240. doi:10.21273/HORTTECH04980-21
9. Askari-Khorasgani O., Pessarakli M. Shifting saffron (*Crocus sativus* L.) culture from traditional farmland to controlled environment (greenhouse) condition to avoid the negative impact of climate changes and increase its productivity. J. Plant Nutr. 2019; 42:2642-2665. doi:10.1080/01904167.2019.1659348
10. Fallahi H.R., Mahmoodi S. Impact of water availability and fertilization management on saffron (*Crocus sativus* L.) biomass allocation. J. Hortic Postharvest Res. 2018; 1:131-146. doi:10.22077/jhpr.2018.1487.1017
11. Ghanbari J., Khajoei-Nejad G.M., Van Ruth S., Aghighi S. The possibility for improvement of flowering, corm properties, bioactive compounds, and antioxidant activity in saffron (*Crocus sativus* L.) by different nutritional regimes. Ind Crops Prod. 2019;135:301-310.
12. Hashemi S.E., Madahhosseini S., Pirasteh-Anosheh H., Sedaghati E., Race M. The role of nitrogen in inducing salt stress tolerance in *crocus sativus* L.: Assessment based on plant growth and ions distribution in leaves. Sustainability. 2022;15:567.
13. Aghhavani Shajari M., Rezvani Moghaddam P., Ghorbani R., Koocheki A. Does nutrient and irrigation managements alter the quality and yield of saffron (*Crocus sativus* L.)? Agric. Water Manage. 2022; 267:107629.
14. Dastranj M., Sepaskhah A.R. Saffron response to irrigation regime, salinity and planting method. Sci Hortic. 2019; 251:215-224.
15. Kothari D., Thakur M., Joshi R., Kumar A., Kumar R. Agro-climatic suitability evaluation for saffron production in areas of Western Himalaya. Front. Plant Sci. 2021; 12:657819.
16. Dastranj M., Sepaskhah A.R. Effect of irrigation water salinity and deficit irrigation on soil ions variation and uptake by saffron (*Crocus sativus* L.) under two planting methods. J. Plant Growth Regul. 2020; 41:282-299.
17. Chen J., Zhou G., Dong Y., Qian X., Li J., Xu X., Huang H., Xu L., Li L. Screening of key proteins affecting floral initiation of saffron under cold stress using iTRAQ-based proteomics. Front. Plant Sci. 2021;12:1-17.
18. Wang Z., Li X., Xu J., Yang Z., and Zhang Y. Effects of ambient temperature on flower initiation and flowering in saffron (*Crocus sativus* L.). Sci Hortic. 2021; 279:109859.
19. Zhou T., Qiu X., Zhao L., Yang W., Wen F., Wu Q., Yan J., Xu B., Chen J., Ma Y., Pei J. Optimal light intensity and quality increased the saffron daughter corm yield by inhibiting the degradation of reserves in mother corms during the reproductive stage. Ind Crops Prod. 2022; 176:114396.
20. Naderi M., Jalalizand A., Javanmard H. Effects of summer irrigation and weed control methods on morphological characteristics and yield of saffron in Iran's climate. Res Crops. 2017; 18:541-545.

21. Bicharanloo, B., Moghaddam P.R., Asadi G. Does summer irrigation alter nitrogen uptake and utilization efficiency of saffron (*Crocus sativus* L.) for different organic and chemical fertilizers? Arch. Agron. Soil Sci. 2020; 67:1754-1769.
22. Koocheki A., Seyyedi S.M. Effects of different water supply and corm planting density on crocin, picrocrocin and safranal, nitrogen uptake and water use efficiency of saffron grown in semi-arid region. Not. Sci. Biol. 2016; 8:334-341.
23. Feizi H., Mollafilabi A., Sahabi H., Ahmadian A. Effect of summer irrigation and conservation tillage on flower yield and qualitative characteristics of saffron (*Crocus sativus* L.). Saffron Agron. Technol. 2015; 2:255-263.
24. Rahimi H., Mokhtarian A., Bazoobandi M., Kiani M., Behdad M. Effects of sowing depth and summer irrigation on *Rhizoglyphus robini* (Acari: Acaridae) population in Gonabad. Appl. Entomol. Phytopathol. 2008; 76:1-14.
25. Sereshti H., Ataolahi S., Aliakbarzadeh G., Zarre S., Poursorkh Z. Evaluation of storage time effect on saffron chemical profile using gas chromatography and spectrophotometry techniques coupled with chemometrics. J. Food Science. Technol. 2018; 55:1350-1359.
26. Behdani M.A., Nassiri Mahallati M., Koocheki A. Evaluation of irrigation management of saffron at agro-ecosystem scale in dry region of Iran. Asian J. Plant Sci. 2008; 21:133-141.
27. Gresta F., Lombardo G.M., Ruberto G., Siracusa L. Saffron, an alternative crop for sustainable agricultural systems: a review. Agron. Sustainable Dev. 2008; 28:95-112.
28. Seghatoleslami M., Sabzekar E. Saffron responses to summer irrigation and nano zinc oxide. Acta Hortic. 2017; 1184:115-118.
29. Pirasteh-Anosheh, H., Hashemi E., Del Borghi A., Spasiano D., Rad M., Race M. Feasibility study of saffron cultivation using a semi-saline water by managing planting date, a new statement. Environ. Res. 2022; 203:111853.
30. Aghhavani Shajari M., Rezvani Moghaddam P., Ghorbani R., Koocheki A. The possibility of improving saffron (*Crocus sativus* L.) flower and corm yield through the irrigation and soil texture managements. Sci Hortic. 2020; 271:109485.
31. PirastehAnosheh H., BabaieZarch M.J., Nasrabadi M., Parnian A., AlaviSiney S.M, Beyrami H., Kaveh H., Hashemi S.E, Durrer K., McDonald K., Race M. Climate and management factors influence saffron yield in different environments. Agrosyst. Geosci. Environ. 2023; e20418.