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RESEARCH REPORT

Light spectrum modulation of corm development and starch biosynthesis in saffron (*Crocus sativus* L.): insights into *CsSUS1*, *CsPhol*, and *SBE* gene expression

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Abstract

Saffron (*Crocus sativus* L.), prized for its culinary, cosmetic, and medicinal uses, faces challenges such as climate change and resource-intensive farming. Controlled environment agriculture (CEA) offers a solution by optimizing growing conditions, particularly for producing large daughter corms essential for yield. This study investigated the impact of various light spectra—blue (B), red (R), and far-red (FR) combinations (1B:1R, 1B:1FR, 1B:2R:1FR, and 1R:1FR)—on saffron growth, photosynthesis, and carbohydrate biosynthesis genes (i.e., *CsSUS1*, *CsPhol*, *SBE*). Results showed that R and FR light enhanced leaf growth and photosynthesis but reduced underground biomass, while a balanced R and FR light increased daughter corm production. Molecular analysis indicated that FR light elevated soluble carbohydrates but decreased carbohydrate storage. The highest *CsSUS1* expression occurred under 1R:1FR light, whereas *CsPhol* and *SBE*, associated with starch biosynthesis, were upregulated under 1B:1R light. A strong correlation was observed between *CsSUS1* expression and soluble carbohydrate content. In summary, FR light promotes vegetative growth while reducing starch accumulation, resulting in smaller daughter corms. In contrast, R and B light enhance starch-related gene expression, increasing daughter corm size. These findings underscore the potential of light spectrum manipulation in CEA to optimize saffron corm production with minimal resource input.

Keywords Carbohydrate metabolism · Chlorophyll fluorescence · Controlled environment agriculture · Gene expression · Light quality · Starch branching

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1 Introduction

Saffron, often called 'red gold', is highly valued for its antioxidant-rich stigmas, which are rich in flavonoids and apocarotenoids. These compounds underpin its significance across the pharmaceutical, cosmetic, food, and health industries (Mzabri et al. 2019). Saffron's propagation relies on asexual reproduction through corms, an underground storage structure, which makes the production of robust, high-weight corms a critical factor for sustaining yield and quality over time (Poggi et al. 2017). Previous studies have highlighted the significance of corm weight and size, as daughter corms derive approximately 40% of their carbohydrates from mother corms (Negbi et al. 1989; Renau-Morata et al. 2012). The production of saffron faces numerous challenges attributed to climate change, diminishing soil and fertile land, water scarcity, and the prevalence of diverse corm diseases (Molina et al. 2004, 2010; Moradi et al. 2022).

Controlled environment agriculture (CEA) has emerged as a solution to these challenges, focusing on healthy crop production, reduced pest and disease infections, increased production per unit area, and efficient resource use (Kozai et al. 2020; Moosavi-Nezhad et al. 2022b). Recent studies have explored CEA as a potential solution for saffron cultivation challenges (Poggi et al. 2017; Moradi et al. 2021, 2022; Moradi 2022; Zhu et al. 2022a; Gao et al. 2023). CEA systems like vertical farms allow tighter environmental control, addressing limitations of traditional greenhouses. These limitations can restrict production and increase susceptibility to pests and diseases. Some studies have focused on improving the quality and quantity of saffron through greenhouse cultivation. Some studies have focused on enhancing the quality and quantity of saffron through greenhouse cultivation (Maggio et al. 2006; Molina et al. 2010; Molina et al., 2005a; Souret and Weathers 2000; Qaryouti et al., 2025). While these efforts have improved stigma quality, producing suitable corms remains challenging in greenhouse environments (Renau-Morata et al. 2012). To address this, greenhuse-field saffron cultivation has been proposed, where flower production occurs in greenhouses, and corm production occurs in the field (Poggi et al. 2017).

Environmental factors play crucial roles in corm production (Negbi et al. 1989). In particular, light is an indispensable environmental factor influencing plant growth and development. It serves as an energy source for photosynthesis and acts as a key determinant for processes such as photomorphogenesis, flowering, secondary metabolite production, and biomass partitioning or distribution of carbohydrates among different plant organs (Hosseinzadeh et al. 2021; Kozai et al. 2016; Moosavi-Nezhad et al. 2022a; Moradi et al. 2021; Vaštakaitė et al. 2018; Aliniaeifard et al., 2025). Despite the substantial research on the effects of temperature and other environmental factors on corm production, the influence of lighting conditions on corm production remains less explored.

Recent studies have highlighted the role of light in saffron (*Crocus sativus* L.) flowering and photosynthesis. Red light has been shown to enhance photosynthetic activity, promote vegetative growth, and produce larger leaves (Zhu et al. 2022b, c). Monochromatic red light also advanced flowering, increased stigma yield, and improved phytohormone levels and enzymatic activity, particularly in larger corms (Zhu et al. 2022a). Light conditions significantly influence saffron quality. Moradi et al. (2022),evaluated the influence of different blue, red, and white light combinations on saffron's physiological, biochemical, and molecular responses during the flowering stage, reporting distinct outcomes. Blue light improved flower morphology and markedly increased crocin content by upregulating its biosynthetic genes, whereas red light enhanced the levels of safranal and picrocrocin.

There is no research on the impact of light spectra on the genes of the carbohydrate biosynthesis pathway in saffron. However, recent studies on soybean and potato have confirmed the influence of light spectra on the regulation of gene expression and the production of various enzymes in the carbohydrate pathway. These studies suggest that certain light spectra, at specific stages of growth, regulate the expression levels of carbohydrate-related genes, facilitating transport, storage, and the shift between sink and source tissues (He et al. 2022; Wang et al. 2023). Li et al. 2017a) demonstrated that specific light spectra, such as combinations of red and blue (3R:1B), significantly influence carbohydrate metabolism by enhancing the activities of enzymes such as sucrose synthase (SS), acid invertase (AI), and starch accumulation in tomatoes. Previously, we investigated the effects of varying blue (B) and red (R) light ratios on daughter corm production. While B light promoted the formation of larger daughter corms, it was associated with a lower total number of corms. This outcome aligns with findings suggesting that B light induces apical dominance in corm buds, suppressing lateral bud growth (Moradi et al. 2021). More recently, a study on greenhouse-field saffron cultivation found that combining R and B light (3R:2B ratio) improved stigma quality and daughter corm development compared to monochromatic light. Additionally, a photosynthetic photon flux density (PPFD) of 200 µmol m^{-2} s^{-1} was identified as optimal for daughter corm production (Zhou et al. 2022).

Far-red (FR) light plays a pivotal role in photomorphogenesis, influencing key growth processes such as seed germination, stem elongation, leaf expansion, flowering induction, and regulating developmental and physiological responses. The effects of FR light are highly context-dependent, varying with plant species, developmental stage,

and environmental conditions (Lercari 1982). In crops like onion and potato, FR light has been shown to stimulate or accelerate carbohydrate translocation from leaves to underground storage organs (Plantenga et al. 2016). This process is closely tied to the activity of starch synthase and starch-branching enzymes, which are critical for plant starch biosynthesis (Tetlow and Emes 2014). Although extensive research has explored the effects of far-red (FR) light on various plant species, its role in daughter corm production and growth in saffron remains underexplored. A study investigating the influence of the red (R) to far-red (FR) light ratio on saffron growth and crocin production during daughter corm development reported that while the R/FR ratio did not significantly affect corm or stigma size, a low R/FR ratio (1.8) substantially increased crocin levels. This effect was attributed to enhanced translocation of photosynthetic products to the corms, mediated by altered phytochrome activity (Kajikawa et al. 2018). Moreover, understanding the expression of genes involved in carbohydrate biosynthesis—particularly those governing starch, the primary storage carbohydrate, and sucrose, the key soluble carbohydrate—is critical to elucidate the effects of light on these metabolic pathways. (Kajikawa et al. 2018)

Here, we investigate how varying light spectra influence daughter corm production and carbohydrate accumulation in saffron. We hypothesize that specific light wavelengths modulate both physiological responses and gene expression patterns involved in carbohydrate biosynthesis during the vegetative stage. The investigation included biochemical and molecular analyses conducted during the vegetative stage of saffron and focused on the developmental aspects of daughter corms within this stage. By exploring the effects of various light spectra, we aimed to gain insights into the physiological and molecular mechanisms underlying daughter corm production and carbohydrate accumulation during the vegetative growth phase.

2 Materials and methods

2.1 Plant materials and growth conditions

Saffron (*Crocus sativus* L.) corms were obtained from a traditional field in Nishabur, Iran, during the dormancy period in late June. The corms weighing 13 and 15 g were treated with a 0.1% prochloraz fungicide solution and air-dried for two hours. Following a floral initiation protocol adapted from Molina et al. (2005b), the corms were stored at 25 °C and 85 ± 2% relative humidity in a dark room for 90 days (Molina et al. 2005b). The corms were then planted in 15 cm pots filled with perlite and transferred to a 10 m², 2.5 m high LED-equipped growth chamber, where they were exposed to varying ratios of blue (B; 400–500 nm), red (R; 600–700 nm), and far-red (FR; 700–750 nm) light to induce flower emergence at 17 °C.

Upon floral initiation, a biweekly fertigation protocol was followed, using modified Hoagland and Arnon nutrient solutions at half the standard concentration (Hoagland and Arnon 1938).

The light treatments for floral induction included monochromatic and composite combinations of R (600–700 nm) and B (400–500 nm) wavelengths, with or without FR (700–750 nm). The specific light treatments included B (100% B light), R (100% R light), 1B:1R (50% B:50% R), 1B:1FR (50% B:50% FR), 1B:2R:1FR (25% B:50% R:25% FR), and 1R:1FR (50% R:50% FR) configurations. These were provided using 24 W OPPLE LED modules, maintaining a uniform photosynthetic photon flux density (PPFD) of 250 ± 10 µmol m^{-2} s^{-1}. Light treatments were designed based on previous findings in saffron and related crops, suggesting that specific light spectra, particularly combinations of R and B, can significantly affect physiological and molecular responses and their known effects on photosynthesis, growth, and carbohydrate metabolism in plants, which are essential for optimizing saffron corm development and stigma quality (Moradi et al. 2021, 2022; Zhou et al. 2022; Zhu et al. 2022a; Gao et al. 2023). The selected ratios were structured to cover a broad range of spectral conditions, from pure R and B to mixed treatments with varying R to B proportions. This allowed us to evaluate how shifts in light composition influence daughter corm development and gene expression, and to distinguish spectral effects across a continuum of increasing or decreasing R and B light. The photoperiod was set to 11 h per day, and light parameters, including intensity and spectrum, were verified using a PAR-Fluorpen FP 100-MAX and a Sekonic C-7000 light meter. The detailed light spectra for each treatment are shown in Fig. 1. The total exposure to the different light spectra lasted 134 days.

For the vegetative growth phase, temperature settings were maintained at 15/6°C (day/night) cycles, following prior research guidelines. The 11-hour photoperiod was continued to optimize conditions for daughter corm proliferation (Kajikawa et al. 2018; Moradi et al. 2021, 2022).

2.2 Morphological properties during the vegetative stage

Various growth and morphological characteristics were evaluated during the vegetative growth stage and daughter corm production. These included the number of leaves per plant, leaf length, fresh weight (FW), dry weight (DW), root length, and FW and DW of the roots, assessed 60 days after flowering. After the leaves desiccated (120 days after

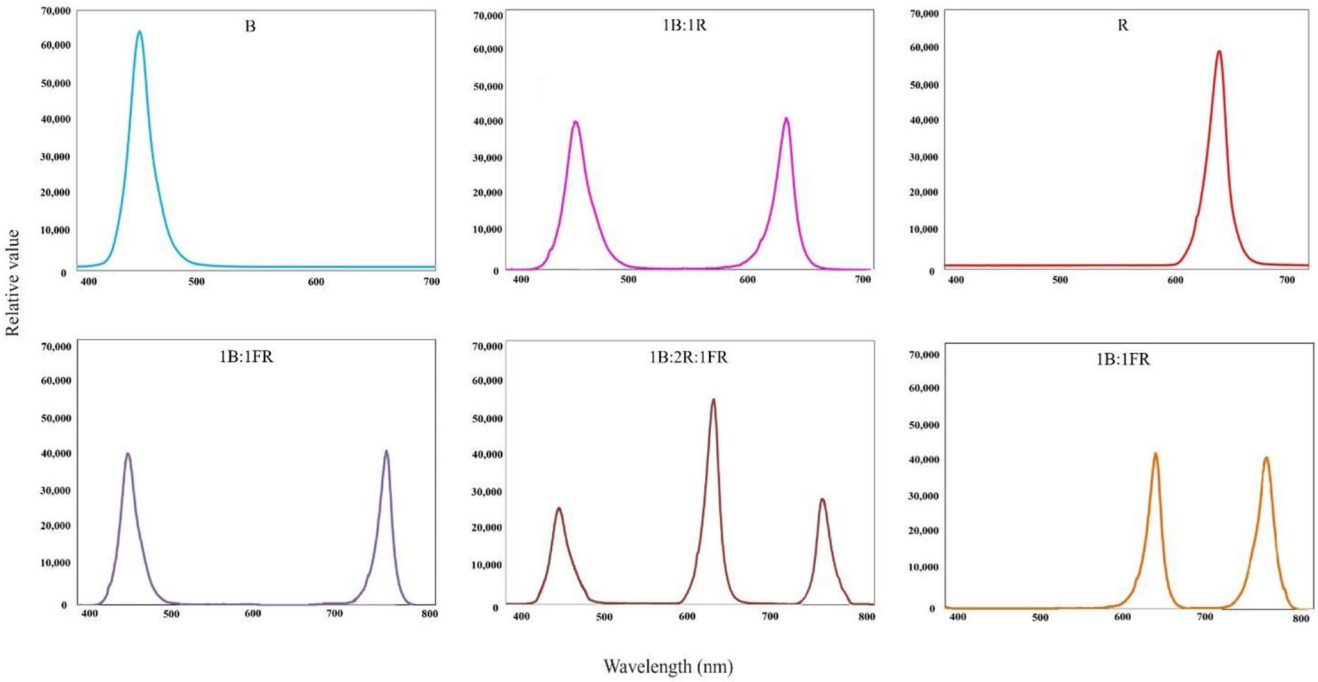


Fig. 1 Light treatments for saffron plant growth in this study. Each treatment represents different light spectral proportions: blue (monochromatic B), red (monochromatic R), 1B:1R (50% B:50%R), 1B:1FR (50%B:50% FR), 1B:2R:1FR (25%B:50%R:25%FR), and 1R:1FR (50%R:50%FR)

flowering, 134 days after light exposure), characteristics related to the daughter corms were measured, including the number of daughter corms per mother corm, as well as the FW and DW of the daughter corms. To determine DW, FW was first recorded at harvest and then reweighed after drying in an oven at 80 °C for 72 h.

2.3 Analysis of photosynthetic functionality

Sixty days after flowering, fully developed leaves of saffron plants were dark-adapted for 20 min by turning off the LED lights. Chlorophyll fluorescence imaging was done using a FluorCam FC 1000-H (Photon Systems Instruments, Drásov, Czech Republic, equipped with a high-resolution CCD camera and four LED panels for precise illumination. The saturation light intensity used for the measurements was set to 3900 µmol photons m^{-2} s^{-1} to ensure complete closure of PSII reaction centers.

Moreover, the polyphasic chlorophyll fluorescence transients (OJIP curves) were recorded using a PAR-fluorPen FP 100-MAX (Photon Systems Instruments, Drásov, Czech Republic) following dark adaptation (≥20 min), capturing fluorescence rise over the first second of illumination. The OJIP transients were plotted on a logarithmic time scale, including phases O to J, J to I, and I to P. The minimal fluorescence yield (F0) was recorded at 50 µs. It represented the open state of PSII, while subsequent points, such as Fj (2 ms) and FI (30 ms), corresponded to intermediate stages of electron transport. The maximum fluorescence yield (Fm) was determined under saturating light and reflects the redox state of QA, the primary quinone electron acceptor of PSII (Aliniaeifard et al. 2014). Nine replicate plants were measured per treatment.

The derived parameters and detailed analysis of the OJIP transients were conducted by methodologies outlined in previous studies (Estaji et al. 2019; Moradi et al. 2021, 2022; Strasser et al. 2000), which provided insights into the in vivo adaptive behavior of the photosynthetic apparatus in response to different growth environments.

2.4 Determination of carbohydrate content

The carbohydrate concentration in the daughter corms was measured using a spectrophotometer (PowerWave™ XS Microplate Reader, BioTek Instruments, Inc., Vermont, USA) following the method outlined by Van Doorn 2012). Daughter corms were collected 120 days after flowering, frozen in liquid nitrogen, and ground into a fine powder. A 0.2 g sample of the powdered tissue was used for analysis. Total soluble carbohydrate content was measured using the anthrone reagent, and absorbance was recorded at 625 nm. Starch quantification was conducted using the sugar–anthrone–sulfuric acid reaction, following the protocol described by McCready et al. 1950).

2.5 Total RNA extraction and rt–qpcr analysis

Daughter corms of saffron plants were collected 120 days after flowering, following leaf senescence. The samples were immediately frozen in liquid nitrogen and stored at -80 °C for later gene expression analysis. The frozen corms were ground into a fine powder and then lyophilized (Beta 1–8 LSC, CHRIST Co., Osterode am Harz, Germany). Total RNA was isolated using TRIzol reagent, following the method described by Wang et al. (2012). RNA quality was assessed via electrophoresis on a 1.5% agarose gel, while RNA purity and concentration were determined using the NanoDrop method (Thermo, Nanodrop 2000, Wilmington, USA).

For cDNA synthesis, 1 µg of total RNA was reverse transcribed using the ProtoScript® II First Strand cDNA Synthesis Kit #E6560 (New England Biolabs, Hitchin, UK) with oligo dT primers. The reverse transcription reaction was conducted at 42 °C for 60 min, followed by enzyme inactivation at 70 °C for 5 min.

To investigate the effects of light spectra on carbohydrate metabolism at the molecular level, we analyzed the expression of key genes in the saffron carbohydrate biosynthesis pathway. Primers for sucrose synthase (*CsSUS1*), starch phosphorylase (*CsPho1*), and starch branching enzyme (*SBE*) genes were designed using Primer3plus software (Untergasser et al. 2012). As the starch branching enzyme-encoding gene has not been sequenced in saffron, we identified conserved domains in monocot species and designed primers to examine the SBE gene in saffron for the first time (Tetlow and Emes 2014). The primers used for gene quantification by quantitative real-time PCR (qRT-PCR) are listed in Table 1.

2.5.1 Reference gene selection and validation

Three candidate reference genes, 18 S rRNA, ubiquitin, and tubulin, were selected based on prior studies that employed gene expression analysis in saffron to ensure accurate normalization of gene expression data (Álvarez-Ortí et al. 2004; Sharma et al. 2018; Ahrazem et al. 2019). Each candidate was evaluated for expression stability using the BestKeeper algorithm, which identified tubulin as the most stable gene under the experimental light conditions. Furthermore, melt curve analysis confirmed the specificity of amplification for all target and reference genes, and PCR efficiency was calculated for each primer pair using standard curve analysis to ensure reliable quantification.2.5.2 RT-qPCR Conditions and Validation.

Quantitative real-time PCR (qRT-PCR) was performed using a LightCycler® system (Roche, USA). The reaction mixture was prepared in a final volume of 20 µl, containing HOT FIREPol® EvaGreen® qPCR Mix Plus (Solis BioDyne, Tartu, Estonia), 2 µl of 1:10 diluted cDNA, 0.5 µM of each primer, and nuclease-free water. The thermal cycling conditions consisted of an initial denaturation step at 95 °C for 12 min, followed by 40 cycles of denaturation at 95 °C for 15 s, annealing at 60 °C for 20 s, and extension at 72 °C for 20 s.

2.5.2 Data extraction

Data were extracted using LightCycler® 96 software. To control for factors such as primer consumption and device efficiency, we used the threshold cycle (Ct) for relative analysis between samples, as Ct is less affected by these variables. Biological and technical replicates were performed with three and two replicates per sample. The relative expression levels of target genes were calculated using the 2^(-ΔΔCt) method (Livak and Schmittgen 2001). The ΔΔCt method was used for data normalization and analysis, with mean comparisons performed using SAS software version 9.4.2. The calculation for ΔΔCt is as follows:

$$\Delta C_t = C_t\ (Target) - C_t\ (Reference)$$

$$2^{-\Delta\Delta Ct} = 2^{-(\Delta Ct\, Target - \Delta Ct\, Reference)}$$

Where Ct Target refers to the threshold cycle of the target sample, while Ct Reference represents the threshold cycle of the reference gene sample. ΔCt Target is the mean Ct for the target gene, and ΔCt Reference is the mean Ct for the reference gene.

2.6 Statistical analysis

This study employed a completely randomized design with six light spectral treatments and twenty individual plants as replicates under each light spectrum. Morphological, biochemical, and photosynthetic parameters were analyzed in six representative plants per treatment, while molecular analyses included three biological replicates with two technical repetitions each. Sample sizes were selected based

Table 1 Sequences of primers used for starch branching (*SBE*), starch sucrose-synthase *(CsSUS1*), phosphorylase (*CsPhol)*, *and tubulin* (reference gene) gene expression analysis in the saffron carbohydrate metabolism pathway

Gene Name	NCBI Reference sequence	Forward primer sequence	Reverse primer sequence
SBE	-	F 5' CTTCGGAAATTGCACCTGCC 3'	R 3' CCCTCGACAACTCTTACGCA 5'
CsPho1	BM005653.1	F 5' ACTATTGCATTCTGGTTGATGGA 3'	R 3' GGATGCCCAAACTCATTTCCCAT 5'
CsSUSl	BM005654.1	F 5' GTCTCGAACCTCGACCGTAG 3'	R 3' TCACTCCATCAACAGCCAAG 5'
Tubulin	CAD33261.1	5' GAATCTGGTGAGGGTGAGGA 3'	3' CAGCCACATCTTCAAACCAA 5'

on prior saffron studies and were considered sufficient to detect meaningful biological and molecular differences with acceptable statistical power.

Data normality and variance homogeneity were confirmed using the Shapiro–Wilk and Levene's tests, respectively, ensuring compliance with ANOVA assumptions. One-way ANOVA identified significant differences among treatment means, followed by Duncan's multiple range test ($\alpha = 0.01$) for mean separation.

To examine associations between starch and sucrose content and the expression of the target genes, Spearman correlation coefficients were calculated using the CORR procedure in SAS. This nonparametric method was chosen for its robustness with ranked data and non-linear relationships. The analysis highlighted monotonic relationships between gene expression and metabolite accumulation under different light treatments. All statistical analyses were conducted in SAS (version 9.4).

3 Results

3.1 Morphological characteristics of the saffron vegetative stage

The morphological characteristics of leaves and roots were measured 60 days after flowering and 80 days after initiating light treatments. Light spectra treatments caused clear, statistically significant variation in growth responses (Table 2).

The 1R:1FR light spectrum yielded the highest number of leaves per corm (23.63), significantly exceeding the 13.43 observed under B light ($P < 0.0001$), marking a 76% increase. This highlights the promoting role of R and FR light in leaf proliferation.

Leaf biomass, both FW and DW, was significantly enhanced under the 1R:1FR treatment, reaching 7.47 mg and 2.08 mg, respectively. These were roughly 3.1-fold and 3.25-fold higher than the values observed under B light (2.37 mg and 0.64 mg). This indicates a pronounced effect of R and FR light in promoting leaf biomass accumulation.

On the other hand, the root growth parameters showed an opposite trend. The root FW and DW were highest in plants exposed to monochromatic B light, averaging 6.12 mg for FW and 2.08 mg for DW. These values were significantly greater ($P < 0.0001$) than those recorded under the 1R:1FR treatment, which presented the lowest root FW (1.66 mg) and DW (0.65 mg), indicating a 73% decrease in FW and a 69% decrease in DW in the 1R:1FR treatment compared with those in the B light treatment.

The root length followed a similar pattern, with the longest roots measured at 4.12 cm in the B light treatment being significantly longer ($P < 0.0001$) than the shortest roots measured at 1.46 cm in the 1R:1FR treatment.

These results indicate that the R and FR light components significantly enhanced leaf growth while inhibiting root extension compared with the B light. The variance analysis for all measured parameters revealed high statistical significance ($P < 0.0001$), with coefficients of variation ranging from 4.78 to 17.42%, indicating the reliability of the observed trends.

The production of large daughter corms is significant, ensuring a promising saffron yield in the following year. The properties of the daughter corms were recorded immediately after the leaves withered (120 days after flowering) in the present study. The impact of light quality on the reproduction of *Crocus sativus* L. was quantified based on production and biomass accumulation in daughter corms (Fig. 2). Statistical analysis revealed significant differences ($P < 0.0001$) in the number of daughter corms per corm across the various lighting treatments (Table 3). Treatments including R, 1R:1FR, and 1B:2R:1FR did not exhibit significant differences when compared to each other. However, these treatments, characterized by a high ratio of red light or the combination of R and FR light, showed a notable increase in the number of daughter corms compared to the other treatments.

Table 2 Leaf and root properties of saffron plants grown under different ratios of red (R), blue (B), and far-red (FR) light. The treatments included B, 1B:1R, R, 1B:1FR, 1B:2R:1FR, and 1R:1FR (spectra are shown in Fig. 1), with a photosynthetic photon flux density (PPFD) of 250 ± 10 µmol m^{-2} s^{-1}

Treatment	No. of leaves per corm	Leaf FW (mg)	Leaf DW (mg)	Leaf length (cm)	Root FW (mg)	Root DW (mg)	Root length (cm)
B	13.43^{f}	2.37^{d}	0.64^{c}	33.11^{f}	6.12^{a}	2.08^{a}	4.12^{a}
1B:1R	15.61^{d}	5.35^{bc}	0.85^{c}	40.08^{e}	4.75^{b}	1.16^{b}	3.75^{b}
R	16.8^{c}	5.95^{b}	1.04^{b}	47.53^{c}	2.06^{d}	0.95^{c}	2.25^{d}
1B:1FR	14.46^{e}	4.63^{c}	0.95^{c}	43.47^{d}	2.8^{c}	0.88^{d}	2.87^{c}
1B:2R:1FR	18.53^{b}	6.32^{b}	1.53^{b}	55.47^{b}	2.61^{cd}	0.96^{c}	1.86^{e}
1R:1FR	23.63^{a}	7.47^{a}	2.08^{a}	62.1^{a}	1.66^{e}	0.65^{e}	1.46^{e}
P	< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001
CV %	7.61	17.42	8.07	5.07	4.78	4.79	8.72

According to Duncan's multiple range test, means within a column followed by the same letter are not significantly different at $p \leq 0.01$. CV percentage indicates the percentage of the coefficient of variation among the treatments. Fresh weight, FW; Dry weight, DW

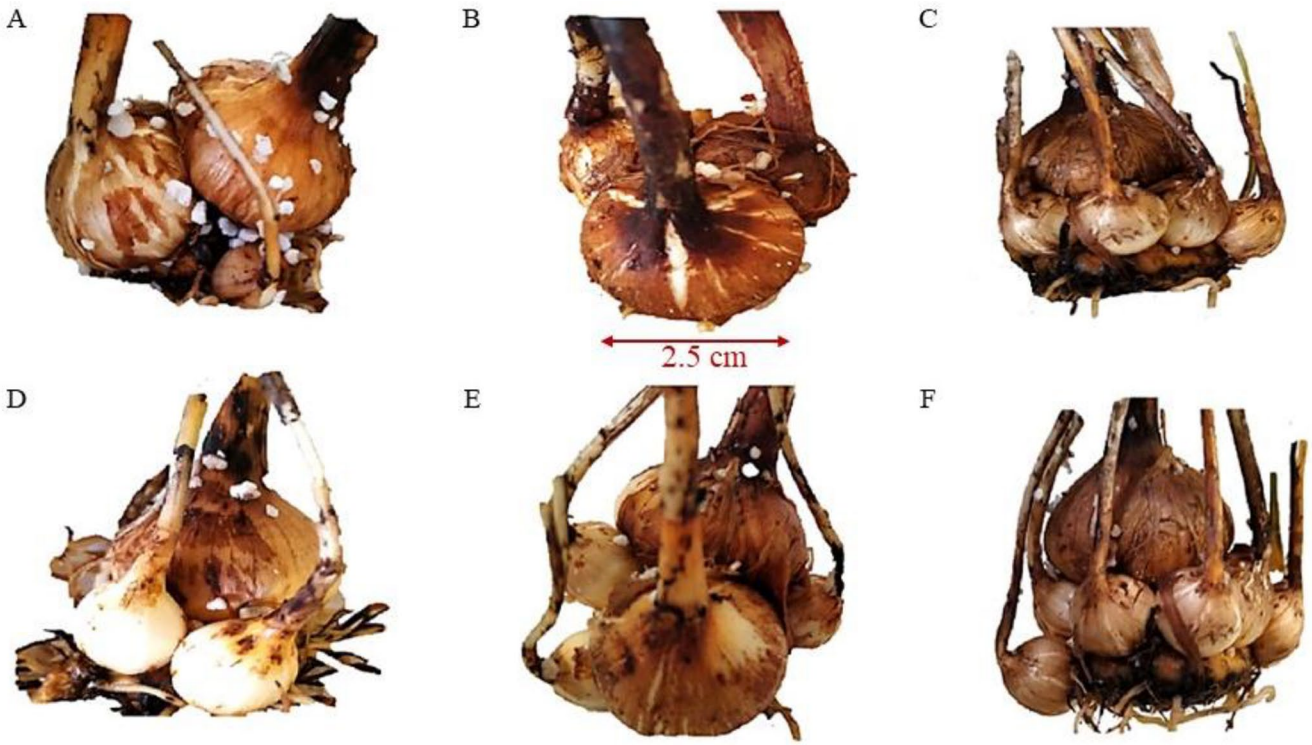


Fig. 2 Representative photos of saffron daughter corms grown under different ratios of red (R), blue (B), and far-red (FR) light. The treatments included B (**A**), 1B:1R (**B**), R (**C**), 1B:1FR (**D**), 1B:2R:1FR (**E**), and 1R:1FR (**F**), with a photosynthetic photon flux density (PPFD) of 250 ± 10 µmol m^{-2} s^{-1}

Table 3 Daughter corm properties of saffron plants grown under different ratios of red (R), blue (B), and far-red (FR) light. The treatments included the following: B, 1B:1R, R, 1B:1FR, 1B:2R:1FR, and 1R:1FR (spectra are shown in Fig. 1), with a photosynthetic photon flux density (PPFD) of 250 ± 10 µmol m^{-2} s^{-1}

Treatment	Number of daughter corms per mother corm	Daughter corm FW (g)	Daughter corm DW (g)	FW of the biggest daughter corm (g)
B	2.03^{c}	12.87^{a}	6.65^{a}	10.74^{a}
1B:1R	2.23^{bc}	12.22^{a}	6.51^{a}	9.53^{b}
R	4^{a}	6.18^{bc}	2.34^{b}	4.88^{d}
1B:1FR	3.05^{b}	6.64^{b}	2.34^{b}	5.94^{c}
1B:2R:1FR	4.45^{a}	5.35^{c}	2.51^{b}	2.93^{e}
1R:1FR	4.84^{a}	4^{d}	1.82^{b}	2.02^{f}
P	<0.0001	<0.0001	<0.0001	<0.0001
CV %	13.45	12.79	21.68	12.36

According to Duncan's multiple range test, means within a column followed by the same letter are not significantly different at $p \leq 0.01$. CV percentage indicates the percentage of the coefficient of variation among the treatments. Fresh weight, FW; Dry weight, DW

Under 1R:1FR light, there was a marked increase, with an average of 4.84 daughter corms per corm, approximately 2.38 times greater than the number produced under monochromatic B light (2.03 daughter corms per corm).

The daughter corm FW and DW also varied significantly with light treatment. The highest FW of the daughter corms was observed under B light (12.87 g), which was statistically equivalent to that under the 1B:1R treatment (12.22 g) but significantly greater than that under the 1R:1FR treatment (Fig. 2. F), which presented the lowest FW (4.00 g). For DW, the B and 1B:1R light treatments (Fig. 2. A, B) resulted in the highest weights (6.65 g and 6.51 g, respectively), which were nearly threefold greater than those recorded under the 1R:1FR treatment (1.82 g).

The FW of the largest daughter corm followed a similar trend, with the heaviest corm at 10.74 g under B light, which was significantly greater ($P < 0.0001$) than that under the 1R:1FR light treatment, where the FW was the lowest at 2.02 g. This represents a substantial decrease of approximately 81% in the FW of the largest daughter corm under the 1R:1FR light compared with that under the B light.

3.2 Photosynthetic efficiency in response to light quality

The maximum quantum yield of PSII (Fv/Fm) varied significantly across light treatments (Fig. 3A). The 1B:2R:1FR treatment recorded the highest value (0.831), suggesting an optimal red-to-far-red ratio for enhancing PSII efficiency in saffron. Compared to the lowest value (0.61) under monochromatic R light, this marks a 36% improvement (Fig. 4).

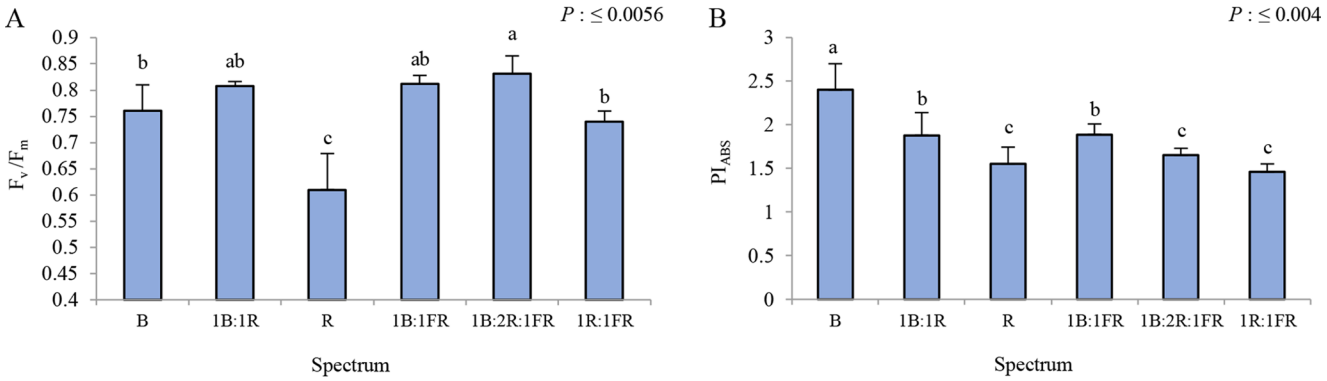


Fig. 3 Maximum quantum yield of PSII (**A**; F_v/F_m) and performance index based on absorption (**B**; PI_{ABS}) of saffron plants grown under different ratios of red (R), blue (B), and far-red (FR) light. The treatments included B, 1B:1R, R, 1B:1FR, 1B:2R:1FR, and 1R:1FR (spectra are shown in Fig. 1), with a photosynthetic photon flux density (PPFD) of 250 ± 10 µmol m^{-2} s^{-1}. Different letters indicate that values significantly differ at $p \leq 0.01$ according to Duncan's multiple range test. The bars represent the mean value ± standard error ($p \leq 0.01$)

These findings imply that excessive red light alone may compromise photochemical performance, while combining it with blue and FR light improves efficiency.

The 1B:1R, 1B:1FR, and 1B:2R:1FR treatments didn't show significant differences, and they represented efficacy in optimizing the F_v/F_m parameter. The intermediate F_v/F_m values of 0.76 and 0,74, recorded for both the B and 1R:1FR treatments, did not differ significantly, yet they were distinct compared to the treatments at the spectrum's extremes.

Similarly, the performance index on absorption basis (PI_{ABS}) varied significantly with light treatment, reflecting the overall photosynthetic performance and energy conservation from photons absorbed by PSII to the reduction in intersystem electron acceptors (Fig. 3B). The highest PI_{ABS} value was recorded for control treatment (B), at 2.4, approximately 46% greater than the lowest value observed in the 1R:1FR treatment (1.46). The plants in the 1B:1R and 1B:2R:1FR treatments presented a PI_{ABS} value of 1,88 and 1,65, respectively, suggesting a possible threshold effect where the addition of far-red light did not further increase the performance index when red light was present.

3.3 Carbohydrate contents of daughter corms

Both soluble sugars and starch content were quantified to evaluate how the light spectrum affects carbohydrate accumulation in daughter corms. Soluble carbohydrate levels significantly increased with R and FR-enriched light, rising from 60.17 mg/g FW in B light to 100.06 mg/g FW under 1R:1FR (Fig. 5A). This 66% increase highlights the stimulatory effect of FR light in the presence of red light on sugar translocation and synthesis.

Conversely, the storage carbohydrate content decreased under the same light treatments (Fig. 5B). The highest storage carbohydrate content was found in the plants in the B treatment (697.95 mg/g DW), which was more than twofold higher than that in the plants in the 1R:1FR treatment (260.06 mg/g DW). Notably, the 1B:2R:1FR treatment resulted in a storage carbohydrate content of 350,01 mg/g DW, which was not significantly different from that of the 1B:1FR treatment at 390,92 mg/g DW. These results suggest that while B light maximizes carbohydrate storage, adding FR light significantly reduces carbohydrate accumulation in daughter corms.

3.4 Gene expression results

3.4.1 Major genes in sucrose biosynthesis

To gain insight into the influence of growing light spectra on the expression of the genes involved in sucrose biosynthesis (Cs*SUS1*), the transcript level of the Cs*SUS1 gene* was analyzed in the saffron corm. The relative gene expression of Cs*SUS1* significantly differed among the treatments (Fig. 6. A). High expression of Cs*SUS1* was observed in daughter corms under the 1R:1FR treatment compared with the other treatments, whose expression was almost two times greater than that in daughter corms grown under monochromatic B light. The results revealed an increasing trend in Cs*SUS1* expression with increasing R and FR light proportions.

3.4.2 Expression of critical genes involved in starch biosynthesis

The influence of light spectra on the expression levels of genes that play key roles in the induction of starch biosynthesis, known as starch phosphorylases (*Cs Pho1)*, was analyzed. The results indicated that the expression of *Cs Pho1* was downregulated by increasing the R and FR light proportions (Fig. 6. B). High transcript levels were detected in daughter corms grown under the 1B:1R light treatment, whose transcript level was almost two times greater than

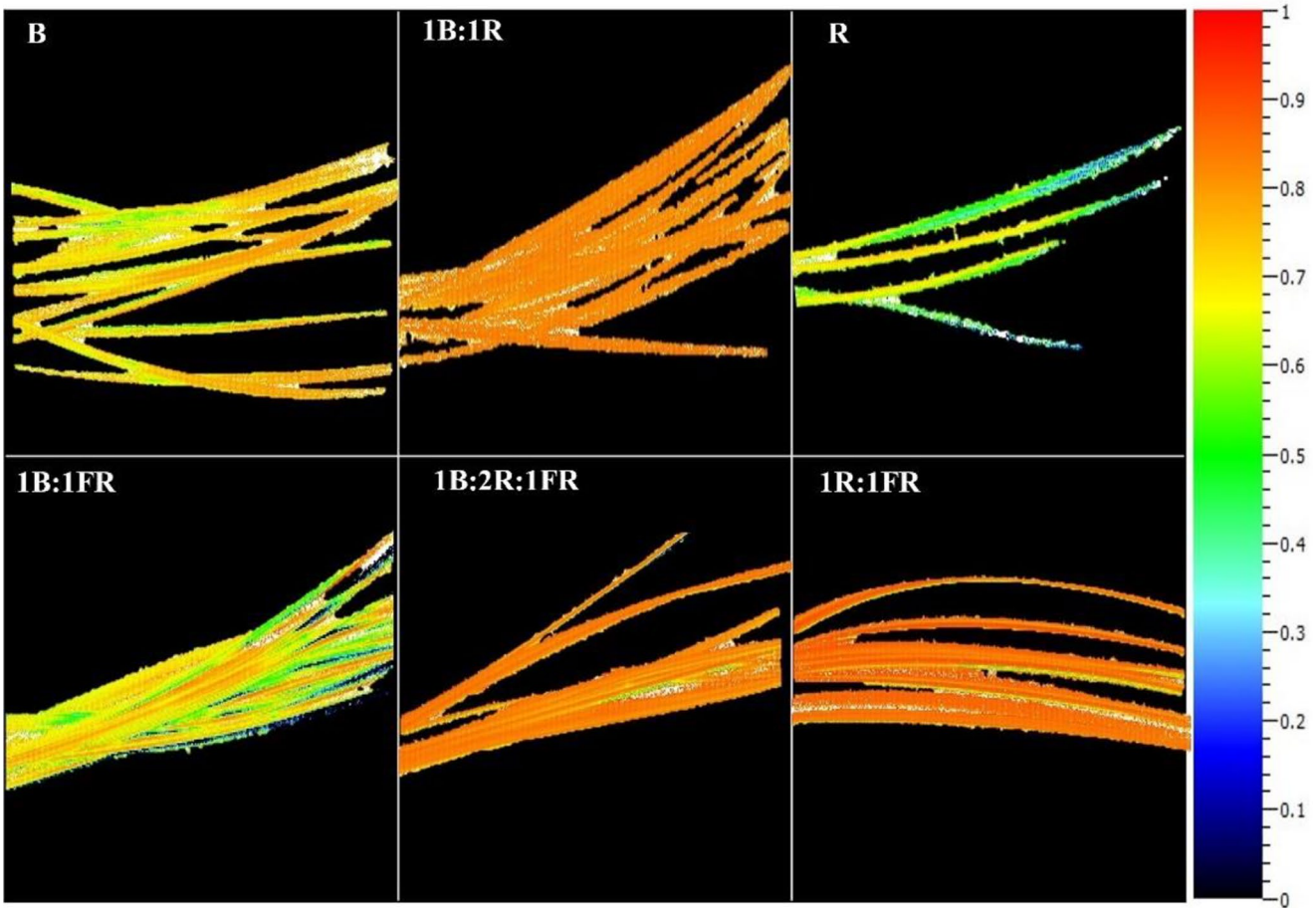
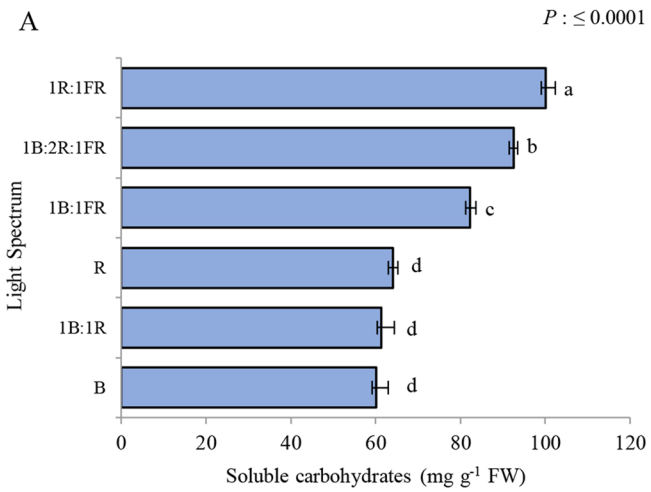


Fig. 4 Chlorophyll *a* fluorescence imaging of saffron plants grown under different ratios of red (R), blue (B), and far-red (FR) light. The treatments included B, 1B:1R, R, 1B:1FR, 1B:2R:1FR, and 1R:1FR (spectra are shown in Fig. 1), with a photosynthetic photon flux density (PPFD) of 250 ± 10 µmol m^{-2} s^{-1}



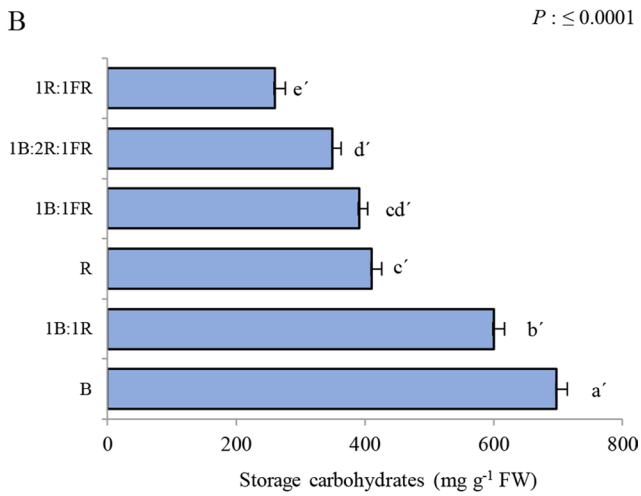


Fig. 5 The contents of soluble carbohydrates (**A**) and storage carbohydrates (**B**) in the daughter corms of plants grown under different ratios of red (R), blue (B), and far-red (FR) light. The treatments included B, 1B:1R, R, 1B:1FR, 1B:2R:1FR, and 1R:1FR (spectra are shown in Fig. 1), with a photosynthetic photon flux density (PPFD) of 250 ± 10 µmol m^{-2} s^{-1}. The bars represent the mean value ± standard error ($p \leq 0.01$)

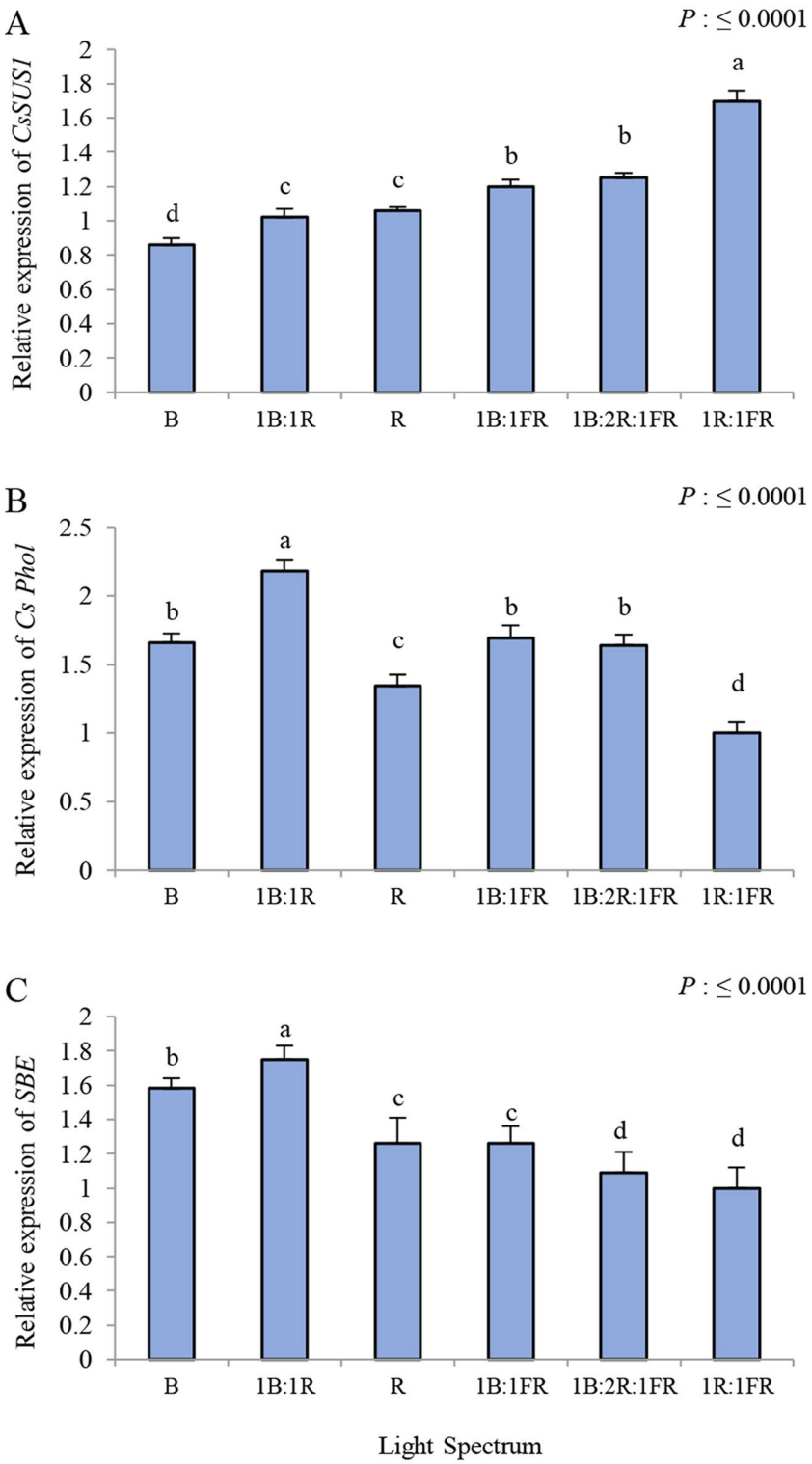


Fig. 6 Relative gene expression levels of significant genes involved in sucrose biosynthesis, sucrose synthase (*Cs SUS1*; **A**), starch biosynthesis starch phosphorylase (*CsPho1*; **B**) and starch barcoding (*SBE*; **C**), in the daughter corms of plants grown under different ratios of red (R), blue (B), and far-red (FR) light. The treatments included B, 1B:1R, R, 1B:1FR, 1B:2R:1FR, and 1R:1FR (spectra are shown in Fig. 1), with a photosynthetic photon flux density (PPFD) of 250 ± 10 µmol m^{-2} s^{-1}. Different letters indicate that values significantly differ at $p \leq 0.01$ according to Duncan's multiple range test. The bars represent the mean value ± standard error ($p \leq 0.01$). The line and R-squared represent linear trends among the treatments and high correlations

that in the plants grown under the 1R:1FR light treatment. The gene expression of daughter corms grown under B, 1B:2R:1FR, or 1B:1FR did not significantly differ. The relative expression of the *SBE* gene, an essential gene in the last step of the starch biosynthesis pathway, under different ratios of B, R, and FR was analyzed. A decreasing trend in the expression of *SBE* was observed with increasing proportions of R and FR light (Fig. 6. C). The highest expression of *SBE* was recorded in the daughter corm grown under the 1B:1R light treatment.

The analysis revealed distinct interactions between enzyme activities and carbohydrate content in saffron plants

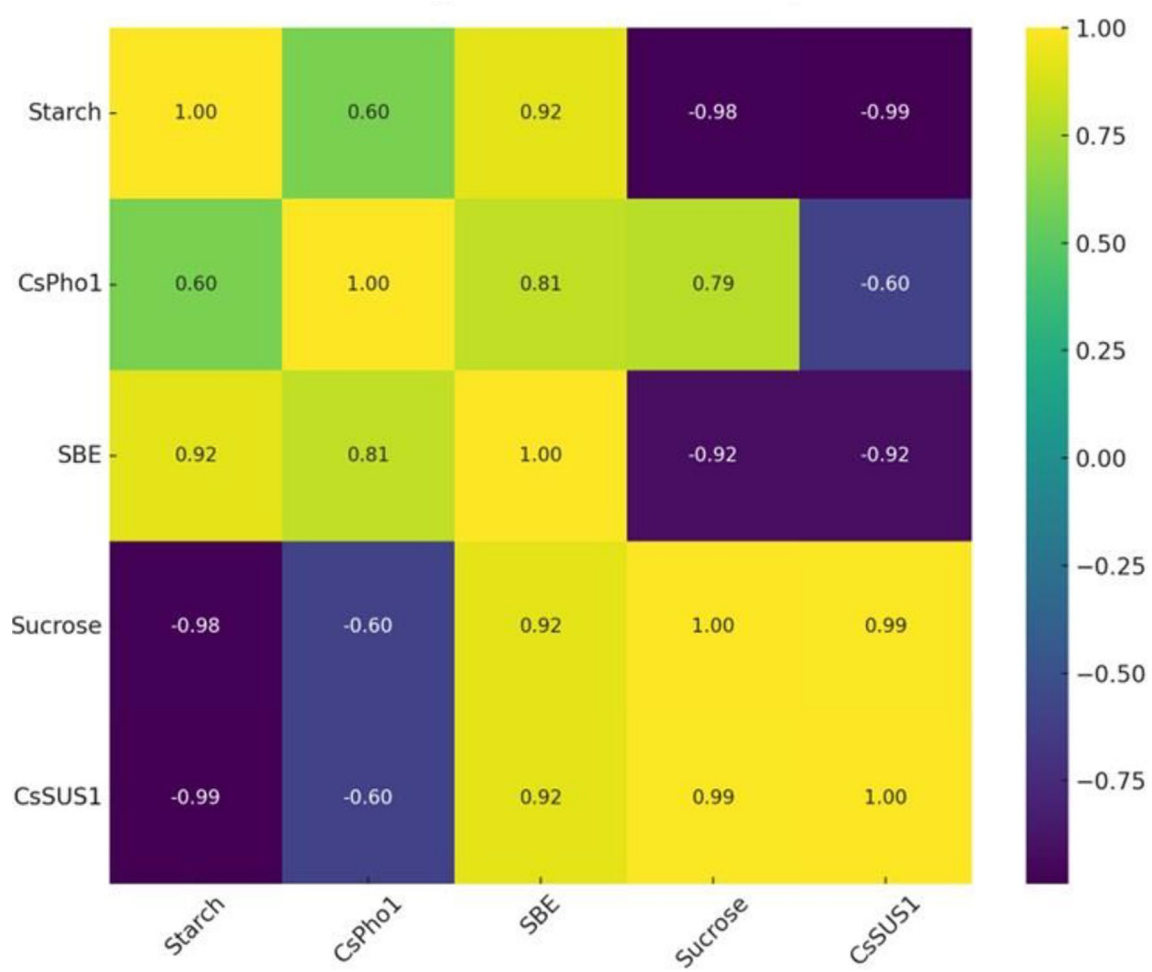


Fig. 7 Correlation among the relative gene expression of carbohydrate metabolism genes and sucrose (as the main soluble carbohydrate) and starch contents in the daughter corms of plants grown under different ratios of red (R), blue (B), and far-red (FR) light. The treatments included B, 1B:1R, R, 1B:1FR, 1B:2R:1FR, and 1R:1FR (spectra are shown in Fig. 1), with a photosynthetic photon flux density (PPFD) of 250 ± 10 µmol m^{-2} s^{-1}

(Fig. 7). Correlation analysis reveals that starch content was closely linked to the activity of starch branching enzyme (*SBE*) (R^2 = 0.92) while showing an inverse relationship with sucrose levels (R^2 = -0.98), suggesting a competitive dynamic between these two pathways. Sucrose synthase 1 (*Cs SUS1*) correlated inversely with starch (R^2 = -0.99) but aligned positively with sucrose (R^2 = 0.99), highlighting its pivotal role in balancing carbohydrate synthesis. Meanwhile, cytosolic phosphorylase 1 (*Cs Pho1*) exhibited moderate correlations with both starch (R^2 = 0.60) and sucrose (R^2 = 0.79), indicating a versatile involvement across these metabolic routes.

4 Discussion

4.1 Far-red light combined with blue and red light promoted aerial organ production in saffron

Light acts as a fundamental environmental signal, orchestrating various developmental processes through photomorphogenic responses regulated by specialized photoreceptors (Gruda et al. 2025). Among these, blue (B) light is perceived by cryptochromes (CRY) and phototropins (PHTO), while red (R) and far-red (FR) light are sensed by phytochromes (PHYs), enabling plants to tune their growth in response to environmental cues finely (Yu et al. 2010). These light receptors collectively mediate photomorphogenesis, facilitating adaptive interactions with the environment (Kong and Zheng 2022). This study highlights the critical role of light quality in shaping plant morphology. R and FR light enhanced leaf expansion and increased foliar number,

promoting vegetative growth. In contrast, elevated levels of B light led to more compact growth with enhanced root development, potentially through CRY1-mediated modulation of auxin dynamics (Casal 2013). Leaf FW and DW parameters are also responsive to R and FR light, with FR light eliciting a shade avoidance syndrome characterized by slender stems and elongated leaves (Demotes-Mainard et al. 2016). Previous studies on lettuce, rose, chrysanthemum, and campanula plants have reported the positive effects of R light on vegetative growth and leaf production (Li et al. 2021; Ouzounis et al. 2014). It has been suggested that R and FR, by modulating phytochrome photoreceptors, R and FR improve plants' elongation and vegetative growth (Ouzounis et al. 2014; Li et al. 2021).

The positive effects of B light on root properties were recorded in this study. B light receptors induce root development by controlling auxin biosynthesis and accumulation (Dou et al. 2017). Similarly, the positive effect of B light on the improvement of root FW and DW has been reported in potato plants (Aksenova et al. 1994).

The production of heavy daughter corms is the turning point in successful saffron production. The properties of the daughter corms confirmed the influence of R and FR light on the number of daughter corms produced by the mother corm. In contrast, B light reduced the number of corms and increased the FW and DW of the daughter corm. Therefore, a high proportion of FR light affects corms' axillary buds and induces their growth. Phytochrome photoreceptors, which are sensitive to R and FR, induce gibberellin (GA) biosynthesis and improve bud outgrowth in plants (Demotes-Mainard et al. 2016; Dierck et al. 2017; Xie et al. 2017). In the present study, the highest numbers of daughter corms, FW and DW, were recorded in the B- and 1B:1R-grown plants. Similar results were reported in a study on lily plants, which reported the largest size of lily bulblets in the plants grown under a 50% B: 50% R light treatment (Lian et al. 2002).

4.2 The combination of blue and far-red light improves photosynthetic performance

The OJIP test, a fast and precise method, was used to study the photosynthetic performance of saffron in response to different light spectra. The highest F_v/F_m was recorded in the leaves of plants that were grown under an equal combination of R and B light with or without FR (1B:1R, 1B:2R:1FR), and the lowest F_v/F_m was recorded under monochromatic R light. The results of the present study revealed high photosynthetic efficiency in plants grown under high proportions of FR and B light, indicating the positive effects of FR and B light on PSII proficiency in saffron plants. It has been suggested that FR improves photosynthetic efficiency by increasing the degree of quenching in photosystem II (Lercari 1982; Li et al. 2021). The findings of the present study are consistent with those of previous studies reporting the positive influence of B light on increasing the efficiency of photosynthesis by facilitating electron transport in the electron transport chain in chrysanthemum and saffron (Aliniaeifard & van Meeteren 2016; Moradi et al. 2021). The performance index for absorption (i.e., PI_{ABS}) is a parameter used to evaluate the overall performance of the photosynthetic apparatus of a plant. The OJIP test and PI_{ABS} calculation are valuable tools for monitoring plant stress, as changes in the OJIP transient curve and PI_{ABS} can provide early indications of photosynthetic damage caused by environmental stresses such as drought, heat, and nutrient deficiencies. PI_{ABS} provides a sensitive indicator of the photosynthetic performance of a plant under different environmental conditions or stressors (Mathur et al. 2013; Bayat et al. 2018; Estaji et al. 2019). In the present study, the highest value of the PI_{ABS} was detected in plants grown under monochromatic B light treatment, and plants grown under treatment with a high proportion of R presented a low PI_{ABS}. The difference in trends between F_v/F_m and PI_{ABS} can be attributed to the distinct ways these parameters capture photosynthetic responses to stress and efficiency. F_v/F_m is sensitive to photodamage and the direct impact of stress on PSII, indicating that high red light treatments may inhibit PSII efficiency due to potential overexcitation or ineffective energy transfer to chlorophyll a. In contrast, PI_{ABS} reflects the broader functionality and resilience of the photosynthetic apparatus, showing that blue light supports photosynthetic efficiency even when stress indicators appear in F_v/F_m. These parameters provide complementary insights: F_v/F_m highlights photodamage and stress. At the same time, PI_{ABS} reveals the overall energy conversion capacity, underlining the importance of both metrics for a comprehensive understanding of light-induced responses in photosynthetic performance.

The finding that the highest PI_{ABS} was observed in plants grown under monochromatic B light highlights light quality's crucial role in regulating photosystem II's efficiency (PSII). B light is known to have a significant effect on the function of PSII, as the B light photoreceptor cryptochrome absorbs it and influences a range of processes, including photomorphogenesis, chloroplast movement, and stomatal opening (Sipos et al. 2021). A relatively high PI_{ABS} was detected in plants grown under monochromatic B light, suggesting that B light is particularly effective in promoting optimal PSII functionality, possibly by enhancing energy transfer efficiency between antenna pigments and/or the primary photochemistry of PSII. These findings are consistent with previous studies that reported the positive influence of B light on photosynthetic efficiency in a range of plant

species, including tomato, lettuce, and basil (Li and Kubota 2009; Bantis et al. 2016; Sipos et al. 2021).

The finding that plants grown under treatment with 1R:1FR presented a low PI_{ABS} suggests that R light in combination with FR light may have a less favorable effect on photosystem II (PSII) efficiency than B light. The positive impact of FR light on photosynthetic performance indicates that FR light enhances the photosynthetic efficiency of short-wavelength light that overexcites PSII, which is consistent with the results of the present study (Zhen and van Iersel 2017). This may be due to several factors, including differences in the absorption spectra of chlorophyll a and b pigments, the efficiency of energy transfer between antenna pigments, and the primary photochemistry of PSII. Red light is primarily absorbed by chlorophyll a, whereas chlorophyll a and b absorb B light. The lower PI_{ABS} recorded in plants grown under a high proportion of R light may therefore reflect a lower efficiency of energy transfer from accessory pigments to chlorophyll a, leading to reduced photochemical activity of PSII (Zhen and van Iersel 2017).

4.3 A high proportion of R and FR light reduced carbohydrate storage in the daughter corm

The influence of different ratios of B, R, and FR on the carbohydrate content of the daughter corm was measured at the daughter corm development stage (120 days after flowering). Light quality significantly influenced the distribution of carbohydrates in saffron daughter corms. Higher proportions of R and FR light increased soluble carbohydrate accumulation, with the 1R:1FR treatment resulting in the highest concentrations. Conversely, starch content decreased under these treatments, suggesting a trade-off between transient sugar production and long-term carbon storage. In contrast, B light increased starch deposition, highlighting its role in directing assimilated carbon toward structural storage in underground tissues.

These findings suggest that R and FR light enhance the mobilization and accumulation of soluble sugars, potentially stimulating vegetative growth through increased source–sink activity. However, their effect on starch deposition appears limited, possibly due to the preferential allocation of carbon toward transitory pools. This interpretation aligns with previous studies showing that FR light did not improve daughter corm biomass in saffron, despite enhancing sugar levels (Kajikawa et al. 2018) while positively influencing sugar storage in onion roots (Lercari 1982). In contrast, B light significantly improved starch accumulation in saffron daughter corms, indicating a spectral regulation of carbon storage pathways. This is consistent with studies in crops such as cucumber, tomato, and sweet pepper, where B light promoted below-ground biomass and starch reserves(Samuolienė et al. 2010). In contrast, some reports have shown that monochromatic R light promotes sugar and starch accumulation in cotton, tomato, and strawberry (Choi et al. 2009; Li et al. 2010, 2017b).

The effects of light spectra on carbohydrate accumulation differ among plant species. The accumulation of carbohydrates in underground organs under different light spectra depends on whether this process is affected by phytochromes and cryptochromes. For example, the formation of underground onion organs may be influenced by the phytochrome receptor, which is sensitive to R and FR light (Lercari 1982).

4.4 Blue light upregulates the expression of genes involved in starch metabolism

To investigate how light spectra influence the expression of genes associated with carbohydrate metabolism, we assessed transcript levels of CsSUS1, CsPho1, and SBE in saffron daughter corms. CsSUS1, which catalyzes sucrose breakdown into UDP-glucose and fructose, showed significantly increased expression under red and far-red light. This corresponded with higher soluble carbohydrate content, suggesting that light quality regulates sucrose metabolism through phytochrome-mediated signaling. A strong correlation (R^2 = 0.99) between CsSUS1 expression and soluble sugar levels reinforces this link. These observations align with findings in lettuce, where red-light-enriched spectra upregulated SS activity and promoted sugar accumulation(Liu and Liu 2022). The results suggest that specific wavelengths can modulate sucrose metabolism at both transcriptional and enzymatic levels, shaping carbon allocation strategies in response to light.

Plastidyl starch phosphorylase (*CsPho1*) has a role in stimulating and inducing starch biosynthesis. Another study suggested that there may be a positive interplay between starch phosphorylase and starch branching enzymes in rice, which may promote the biosynthesis of maltodextrins (MDs). This positive interaction increases the branching of MDs and initiates the biosynthesis of amylopectin molecules, which are the main building blocks of starch (Nakamura et al. 2010). SBE is responsible for introducing α-1,6-glycosidic linkages in amylopectin, creating the branched structure of starch. This branching is essential for starch solubility and functionality. The activity of SBE directly influences the amylose-to-amylopectin ratio, affecting the gelatinization, digestibility, and other properties of starch. During the filling stage of storage organs, *SBE* activity is crucial for efficient starch deposition and optimized energy storage (Tetlow and Emes 2014)(Tetlow and Emes 2014). The highest expression level of the *SBE* gene was observed under an equal ratio of red and blue light.

Additionally, it was found that increasing the proportion of red or far-red light decreased gene expression. These results clearly demonstrate how light influences the physiological pathways of various carbohydrates, aiding the plant in energy storage.

The present study's results agree with those of the present study, which revealed a strong correlation between the *CsPho1* gene and the *SBE* gene ($R^2 = 0.81$). CsPho1 and SBE presented increased expression levels with a high proportion of B light. The highest expression of *CsPho1* and *SBE* was recorded with the combination of B and R light (1B:1R). These results indicated that B light significantly influences plant carbohydrate metabolism and structural development. The positive effect of B light on the modulation of genes in the carbohydrate biosynthesis pathway was also reported in previous studies. They reported that blue light enhances sucrose transport to sink tissues and boosts starch production in potato tubers. Together, these findings underscore the critical role of blue light in orchestrating carbohydrate dynamics and structural adaptations in diverse plant species (He et al. 2022). Similarly, in soybean hypocotyls, it regulates enzymes and metabolites involved in the synthesis of cellulose, hemicellulose, and pectin, leading to altered cell wall composition, reduced elongation, and increased stem thickness (Wang et al. 2023). This finding further supports the hypothesis that light spectra play a pivotal role in coordinating starch metabolism through the regulation of gene networks. Such modulation allows plants to adapt their energy storage and utilization strategies to varying light environments.

The results revealed a positive relationship between the starch content as a storage carbohydrate and the gene expression of *CsPho1* and *SBE*, with $R^2 = 0.6$ and $R^2 = 0.92$, respectively. In agreement with these results, a study on the effect of light intensity on the gene expression of alfalfa plants revealed a strong correlation between the expression of the *Pho1* and *SBE genes* and carbohydrate storage. It has also been reported that light affects the expression of these genes (Tang et al. 2022). This study's results contribute to the growing body of evidence that the light spectrum affects plant morphology and plays a crucial role in regulating the genetic pathways that govern carbohydrate metabolism. By elucidating these relationships, the study highlights light spectra's intricate and multifaceted regulatory role in shaping metabolic processes at the genetic and physiological levels. The varied responses observed under different light conditions highlight the nuanced and complex nature of light-mediated metabolic regulation in plants.

These findings highlight that saffron's carbohydrate metabolism is shaped by light quality and intricate interactions within light-responsive pathways involving phytochromes (sensitive to red and FR light) and cryptochromes (sensitive to B light). The gene expression patterns suggest that red and far-red light pathways activate processes that support soluble carbohydrate synthesis, while blue light favors pathways that promote starch storage. Adjusting light spectra in controlled environments could optimize how carbohydrates are allocated in saffron, balancing between vegetative growth and storage depending on production objectives.

For commercial saffron cultivation, these findings highlight opportunities to enhance corm storage carbohydrate content through strategic light management. For instance, applying blue light during specific growth stages can boost starch accumulation in underground biomass, supporting the development of robust daughter corms and potentially increasing future yields. However, the higher energy demand of blue light than red and far-red light must be weighed against production costs. While red and far-red light promote soluble carbohydrate accumulation and vegetative growth, they may limit storage carbohydrates, necessitating a balanced approach to optimize yields.

A tailored light strategy based on energy efficiency and developmental needs could address these challenges. During the early vegetative phase (approximately the first month after flowering), a high red spectrum could stimulate vegetative growth and sucrose production. In subsequent months, a balanced 50% blue and 50% red light composition could be introduced, transitioning to blue light during the final two months of vegetative growth. This phase, critical for daughter corm development, would benefit from blue light to maximize starch accumulation and ensure robust storage reserves.

Optimizing daughter corm production in controlled environments also holds promise for shortening the growth cycle and enabling multiple harvests of daughter corms within a single year. Future studies could explore dynamic light scheduling systems tailored to saffron's unique developmental timeline to further increase production efficiency.

5 Conclusion

The crucial part of saffron cultivation in greenhouses is the production of large daughter corms to guarantee the yield of the subsequent year. The results of the present study were indicative of the negative effects of R and FR light on daughter corm production. Both R and FR light spectra promoted the growth of leaves and led plants to continue vegetative and above-ground growth. The highest levels of carbohydrate storage and transcripts of genes related to the starch pathway were recorded in plants grown under a high proportion of B light. The combination of B and R lights (1B:1R) promoted the production of large daughter corms

and increased the starch content, resulting in the upregulation of important genes in the starch biosynthesis pathway (*CsPho1* and *SBE*). These findings suggest that daughter corm production in saffron is dependent on B light in the growing environment. To further elucidate the mechanism underlying this phenomenon, measuring the enzymatic activity associated with carbohydrate metabolism could provide valuable information. These investigations may shed light on the specific enzymatic processes stimulated under various light conditions, thereby enhancing our understanding of the biochemical pathways that govern corm development in saffron.

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Data availability The original contributions presented in the study are included in the article, further inquiries can be directed to the corresponding authors.

Declarations

Conflict of interest The author declares that there are no conflicts of interest.

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References

Ahrazem O, Diretto G, Argandoña Picazo J, Fiore A, Rubio-Moraga Á, Rial C, Varela RM, Macías FA, Castillo R, Romano E, Gómez-Gómez L (2019) The specialized roles in carotenogenesis and apocarotenogenesis of the phytoene synthase gene family in saffron. Front Plant Sci 0:249. https://doi.org/10.3389/FPLS.2019.00249

Aksenova NP, Konstantinova TN, Sergeeva LI, Macháčková I, Golyanovskaya SA (1994) Morphogenesis of potato plants in vitro. I. Effect of light quality and hormones. J Plant Growth Regul 13:143–146. https://doi.org/10.1007/BF00196378

Aliniaeifard S, Van Meeteren U (2016) Stomatal characteristics and desiccation response of leaves of cut chrysanthemum (Chrysanthemum morifolium) flowers grown at high air humidity. Sci Hortic 205:84–89. https://doi.org/10.1016/J.SCIENTA.2016.04.025

Aliniaeifard S, Azizi S, Zarbakhsh S, Esmaeili S, Baghalian K, Gruda NS (2025) Light in controlled environment agriculture. Int J Veg Sci 1–7. https://doi.org/10.1080/19315260.2025.2503228

Aliniaeifard S, Malcolm Matamoros P, van Meeteren U (2014) Stomatal malfunctioning under low VPD conditions: induced by alterations in stomatal morphology and leaf anatomy or in the ABA. signaling? Physiol Plant 152:688–699. https://doi.org/10.1111/PPL.12216/FULLPDF

Álvarez-Ortí M, Gómez-Gómez L, Rubio Á, Escribano J, Pardo J, Jiménez F, Fernández JA (2004) Development and gene expression in saffron corms. Acta Hortic 650:141–153. https://doi.org/10.17660/ACTAHORTIC.2004.650.14

Bantis F, Ouzounis T, Radoglou K (2016) Artificial LED lighting enhances growth characteristics and total phenolic content of Ocimum basilicum, but variably affects transplant success. Sci Hortic 198:277–283. https://doi.org/10.1016/j.scienta.2015.11.014

Bayat L, Arab M, Aliniaeifard S, Seif M, Lastochkina O, Li T (2018) Effects of growth under different light spectra on the subsequent high light tolerance in Rose plants. AoB Plants 10. https://doi.org/10.1093/AOBPLA/PLY052

Casal JJ (2013) Photoreceptor signaling networks in plant responses to shade. Annu Rev Plant Biol 64:403–427. https://doi.org/10.1146/ANNUREV-ARPLANT-050312-120221

Choi HG, Moon BY, Kang NJ (2009) Effects of LED light on the production of strawberry during cultivation in a plastic greenhouse and in a growth chamber. Sci Hortic 189:22–31. https://doi.org/10.1016/J.SCIENTA.2015.03.022

Demotes-Mainard S, Péron T, Corot A, Bertheloot J, Le Gourrierec J, Pelleschi-Travier S, Crespel L, Morel P, Huché-Thélier L, Boumaza R, Vian A, Guérin V, Leduc N, Sakr S (2016) Plant responses to red and far-red lights, applications in horticulture. Environ Exp Bot 121:4–21. https://doi.org/10.1016/J.ENVEXPBOT.2015.05.010

Dierck R, Dhooghe E, Van Huylenbroeck J, Van Der Straeten D, De Keyser E (2017) Light quality regulates plant architecture in different genotypes of Chrysanthemum morifolium Ramat. Sci Hortic 218:177–186. https://doi.org/10.1016/J.SCIENTA.2017.02.016

Dou H, Niu G, Gu M, Masabni JG (2017) Effects of light quality on growth and phytonutrient accumulation of herbs under controlled environments. Horticulturae 3. https://doi.org/10.3390/horticulturae3020036

Estaji A, Kalaji HM, Karimi HR, Roosta HR, Moosavi-Nezhad SM (2019) How glycine betaine induces tolerance of cucumber plants to salinity stress? Photosynthetica 57:753–761. https://doi.org/10.32615/ps.2019.053

Gao D, Ji X, Yuan Q, Pei W, Zhang X, Li F, Han Q, Zhang S (2023) Effects of total daily light integral from blue and broad-band red leds on flowering of saffron (Crocus sativus L). Sci Rep 13. https://doi.org/10.1038/S41598-023-34424-0

Gruda NS, Samuolienė G, Dong J, Li X (2025) Environmental conditions and nutritional quality of vegetables in protected cultivation.

Compr Rev Food Sci Food Saf 24. https://doi.org/10.1111/1541-4337.70139
He W, Pu M, Li J, Xu ZG, Gan L (2022) Potato tuber growth and yield under red and blue leds in plant factories. J Plant Growth Regul 41:40–51. https://doi.org/10.1007/S00344-020-10277-Z/METRICS
Hoagland DR, Arnon DI (1938) The water-culture method for growing plants without soil: university of California. Circular Calif Agricultural Exp Stn 347:32
Hosseinzadeh M, Aliniaeifard S, Shomali A, Didaran F (2021) Interaction of light intensity and CO2Concentration alters biomass partitioning in Chrysanthemum. J Hortic Res 29:45–56. https://doi.org/10.2478/JOHR-2021-0015
Kajikawa N, Uno Y, Kuroki S, Miyagawa S, Yamashita Y, Hamaguchi Y, Ueda Y, Kobayashi M, Kaji K, Itoh H (2018) Effect of far-red light on saffron (crocus sativus L.) growth and Crocin yields. Environ Control Biol 56:51–57. https://doi.org/10.2525/ecb.56.51
Kong Y, Zheng Y (2022) Low-activity Cryptochrome 1 plays a role in promoting stem elongation and flower initiation of mature Arabidopsis under blue light associated with low phytochrome activity. https://doi.org/10.1139/CJPS-2021-0122.
Kozai T, Fujiwara K, Runkle ES (2016) LED lighting for Urban agriculture. LED Lighting for Urban Agriculture 1–454. https://doi.org/10.1007/978-981-10-1848-0
Kozai T, Niu G, Takagaki M (2020) Plant factory. Elsevier
Lercari B (1982) The effect of far-red light on the photoperiodic regulation of carbohydrate accumulation in Allium cepa L. Physiol Plant 54:475–479. https://doi.org/10.1111/J.1399-3054.1982.TB00711.X
Li Q, Kubota C (2009) Effects of supplemental light quality on growth and phytochemicals of baby leaf lettuce. Environ Exp Bot 67:59–64. https://doi.org/10.1016/j.envexpbot.2009.06.011
Li H, Xu Z, Tang C (2010) Effect of light-emitting diodes on growth and morphogenesis of upland cotton (Gossypium hirsutum L.) plantlets in vitro. Plant Cell Tissue Organ Cult 103:155–163. https://doi.org/10.1007/S11240-010-9763-Z
Li Y, Xin G, Wei M, Shi Q, Yang F, Wang X (2017a) Carbohydrate accumulation and sucrose metabolism responses in tomato seedling leaves when subjected to different light qualities. Sci Hortic 225:490–497. https://doi.org/10.1016/J.SCIENTA.2017.07.053
Li Y, Xin G, Wei M, Shi Q, Yang F, Wang X (2017b) Carbohydrate accumulation and sucrose metabolism responses in tomato seedling leaves when subjected to different light qualities. Sci Hortic 225:490–497. https://doi.org/10.1016/J.SCIENTA.2017.07.053
Li Y, Wu L, Jiang H, He R, Song S, Su W, Liu H (2021) Supplementary Far-Red and Blue Lights Influence the Biomass and Phytochemical Profiles of Two Lettuce Cultivars in Plant Factory. Molecules 2021, Vol 26, Page 7405 26:7405. https://doi.org/10.3390/MOLECULES26237405
Lian ML, Murthy HN, Paek KY (2002) Effects of light emitting diodes (LEDs) on the in vitro induction and growth of bulblets of Lilium Oriental hybrid Pesaro. Sci Hortic 94:365–370. https://doi.org/10.1016/S0304-4238(01)00385-5
Liu J, Liu W (2022) Regulation of accumulation and metabolism circadian rhythms of starch and sucrose in two leaf-color lettuces by red:blue ratios of LED continuous light. Environ Exp Bot 196:104811. https://doi.org/10.1016/J.ENVEXPBOT.2022.104811
Livak KJ, Schmittgen TD (2001) Analysis of relative gene expression data using real-time quantitative PCR and the 2(-Delta Delta C(T)) method. Methods 25:402–408. https://doi.org/10.1006/METH.2001.1262
Maggio A, Raimondi G, Martino A, De Pascale S (2006) Soilless cultivation of saffron in mediterranean environment. Acta horticulturae. International Society for Horticultural Science, pp 515–521
Mathur S, Mehta P, Jajoo A (2013) Effects of dual stress (high salt and high temperature) on the photochemical efficiency of wheat leaves (Triticum aestivum). Physiol Mol Biology Plants 19:179–188. https://doi.org/10.1007/s12298-012-0151-5
McCready RM, Guggolz J, Silviera V, Owens HS (1950) Determination of starch and amylose in vegetables. Anal Chem 22:1156–1158. https://doi.org/10.1021/ac60045a016
Molina RV, Valero M, Navarro Y, García-Luis A, Guardiola JL (2004) The effect of time of corm lifting and duration of incubation at inductive temperature on flowering in the saffron plant (Crocus sativus L). Sci Hortic 103:79–91. https://doi.org/10.1016/j.scienta.2004.04.008
Molina RV, Valero M, Navarro Y, García-Luis A, Guardiola JL (2005a) Low temperature storage of corms extends the flowering season of saffron (Crocus sativus L). J Hortic Sci Biotechnol 80:319–326. https://doi.org/10.1080/14620316.2005.11511937
Molina RV, Valero M, Navarro Y, Guardiola JL, García-Luis A (2005b) Temperature effects on flower formation in saffron (Crocus sativus L). Sci Hortic 103:361–379. https://doi.org/10.1016/j.scienta.2004.06.005
Molina RV, Renau-Morata B, Nebauer SG, García-Luis A, Guardiola JL (2010) Greenhouse saffron culture - temperature effects on flower emergence and vegetative growth of the plants. Acta Hortic 850:91–94. https://doi.org/10.17660/actahortic.2010.850.12
Moosavi-Nezhad M, Alibeigi B, Estaji A, Gruda NS, Aliniaeifard S (2022a) Growth, biomass partitioning, and photosynthetic performance of Chrysanthemum cuttings in response to different light spectra. Plants (Basel) 11. https://doi.org/10.3390/plants11233337
Moosavi-Nezhad M, Salehi R, Aliniaeifard S, Winans KS, Nabavi-Pelesaraei A (2022b) An analysis of energy use and economic and environmental impacts in conventional tunnel and LED-equipped vertical systems in healing and acclimatization of grafted watermelon seedlings. J Clean Prod 361. https://doi.org/10.1016/J.JCLEPRO.2022.132069
Moradi S (2022) Saffron production under controlled conditions. 221–235. https://doi.org/10.1007/978-3-031-10000-0_13
Moradi S, Kafi M, Aliniaeifard S, Salami SA, Shokrpour M, Pedersen C, Moosavi-Nezhad M, Wróbel J, Kalaji HM (2021) Blue Light Improves Photosynthetic Performance and Biomass Partitioning toward Harvestable Organs in Saffron (Crocus sativus L.). Cells 2021, Vol 10, Page 1994 10:1994. https://doi.org/10.3390/CELLS10081994
Moradi S, Kafi M, Aliniaeifard S, Moosavi-Nezhad M, Pedersen C, Gruda NS, Salami SA (2022) Monochromatic blue light enhances Crocin and Picrocrocin content by upregulating the expression of underlying biosynthetic pathway genes in saffron (Crocus sativus L). Front Hortic 0:1. https://doi.org/10.3389/FHORT.2022.960423
Mzabri I, Addi M, Berrichi A (2019) Traditional and Modern Uses of Saffron (Crocus Sativus). Cosmetics 2019, Vol 6, Page 63 6:63. https://doi.org/10.3390/COSMETICS6040063
Nakamura Y, Utsumi Y, Sawada T, Aihara S, Utsumi C, Yoshida M, Kitamura S (2010) Characterization of the reactions of starch branching enzymes from rice endosperm. Plant Cell Physiol 51:776–794. https://doi.org/10.1093/PCP/PCQ035
Negbi M, Dagan B, Dror A, Basker D (1989) Growth, flowering, vegetative reproduction, and dormancy in the saffron crocus (Crocus sativus l). Isr J Bot 38:95–113. https://doi.org/10.1080/0021213X.1989.10677116
Ouzounis T, Fretté X, Rosenqvist E, Ottosen CO (2014) Spectral effects of supplementary lighting on the secondary metabolites in roses, chrysanthemums, and Campanulas. J Plant Physiol 171:1491–1499. https://doi.org/10.1016/j.jplph.2014.06.012
Plantenga FDM, Siakou M, Bergonzi S, Heuvelink E, Bachem CWB, Visser RGF, Marcelis LFM (2016) Regulating flower and tuber

formation in potato with light spectrum and day length. Acta Hortic 1134:267–275. https://doi.org/10.17660/ACTAHORTIC.2016.1134.36

Poggi LM, Portela JA, Pontın MA, Moratalla-López N, Alonso GL (2017) New proposal for handling of saffron flowering (Crocus sativus L.) under environmentally controlled conditions. Acta Hortic 1184:245–252. https://doi.org/10.17660/ActaHortic.2017.1184.34

Qaryouti M, Al-Soqeer A, ABDELAZIZ ME, Gruda NS, AlSahly S, Alrasheed W, Althobiti S, Babiker O, Sharafeldin M, Voogt W (2025) Cutting-Edge greenhouse practices for better stigma yield and corm quality of saffron (Crocus sativus L). Front Plant Sci 16:1612791. https://doi.org/10.3389/FPLS.2025.1612791

Renau-Morata B, Nebauer SG, Sánchez M, Molina RV (2012) Effect of corm size, water stress and cultivation conditions on photosynthesis and biomass partitioning during the vegetative growth of saffron (Crocus sativus L). Ind Crops Prod 39:40–46. https://doi.org/10.1016/j.indcrop.2012.02.009

Samuolienė G, Brazaitytė A, Urbonavičiu¯tė A, Šabajevienė G, Duchovskis P (2010) The effect of red and blue light component on the growth and development of frigo strawberries. undefined

Sharma M, Kaul S, Dhar MK (2018) Transcript profiling of carotenoid/apocarotenoid biosynthesis genes during corm development of saffron (Crocus sativus L). Protoplasma 2018 256(1):249–260. https://doi.org/10.1007/S00709-018-1296-Z

Sipos L, Balázs L, Székely G, Jung A, Sárosi S, Radácsi P, Csambalik L (2021) Optimization of Basil (Ocimum Basilicum L.) production in LED light environments– a review. Sci Hortic 289. https://doi.org/10.1016/j.scienta.2021.110486

Souret FF, Weathers PJ (2000) The growth of saffron (Crocusï¿¿sativus L.) in aeroponics and hydroponics. J Herbs Spices Med Plants 7:25–35. https://doi.org/10.1300/J044v07n03_04

Strasser RJ, Srivastava A, Tsimilli-Michael M (2000) The fluorescence transient as a tool to characterize and screen photosynthetic samples

Tang W, Guo H, Baskin CC, Xiong W, Yang C, Li Z, Song H, Wang T, Yin J, Wu X, Miao F, Zhong S, Tao Q, Zhao Y, Sun J (2022) Effect of Light Intensity on Morphology, Photosynthesis and Carbon Metabolism of Alfalfa (Medicago sativa) Seedlings. Plants 2022, Vol 11, Page 1688 11:1688. https://doi.org/10.3390/PLANTS11131688

Tetlow IJ, Emes MJ (2014) A review of starch-branching enzymes and their role in amylopectin biosynthesis. IUBMB Life 66:546–558. https://doi.org/10.1002/IUB.1297

Untergasser A, Cutcutache I, Koressaar T, Ye J, Faircloth BC, Remm M, Rozen SG (2012) Primer3—new capabilities and interfaces. Nucleic Acids Res 40:e115. https://doi.org/10.1093/NAR/GKS596

Van Doorn WG (2012) Water relations of cut flowers: an update. Hortic Rev (Am Soc Hortic Sci) 40:55–106. https://doi.org/10.1002/9781118351871.ch2

Vaštakaitė V, Viršilė A, Brazaitytė A, Samuolienė G, Miliauskienė J, Jankauskienė J, Duchovskis P (2018) Pulsed LED light increases the phytochemical level of Basil microgreens. Acta Hortic 1227:579–584. https://doi.org/10.17660/ACTAHORTIC.2018.1227.73

Wang G, Wang G, Zhang X, Wang F, Song R (2012) Isolation of high quality RNA from cereal seeds containing high levels of starch. Phytochem Anal 23:159–163. https://doi.org/10.1002/PCA.1337

Wang C, Chen Y, Cui C, Shan F, Zhang R, Lyu X, Lyu L, Chang H, Yan C, Ma C (2023) Blue light regulates cell wall structure and carbohydrate metabolism of soybean hypocotyl. Int J Mol Sci 24:1017. https://doi.org/10.3390/IJMS24021017/S1

Xie Y, Liu Y, Wang H, Ma X, Wang B, Wu G, Wang H (2017) Phytochrome-interacting factors directly suppress MIR156 expression to enhance shade-avoidance syndrome in Arabidopsis. Nat Commun 2017 8(1 8):1–11. https://doi.org/10.1038/s41467-017-00404-y

Yu X, Liu H, Klejnot J, Lin C (2010) The Cryptochrome blue light receptors. Arabidopsis Book 8:e0135. https://doi.org/10.1199/tab.0135

Zhen S, van Iersel MW (2017) Far-red light is needed for efficient photochemistry and photosynthesis. J Plant Physiol 209:115–122. https://doi.org/10.1016/J.JPLPH.2016.12.004

Zhou T, Qiu X, Zhao L, Yang W, Wen F, Wu Q, Yan J, Xu B, Chen J, Ma Y, Pei J (2022) 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 176:114396. https://doi.org/10.1016/J.INDCROP.2021.114396

Zhu J, Zhang Y, Zhou L, Yang L (2022a) Growth and flowering of saffron (Crocus sativus L.) with three corm weights under different LED light qualities. https://doi.org/10.1016/j.scienta.2022.111202. Sci Hortic 303

Zhu J, Zhang Y, Zhou L, Yang L (2022b) Growth and flowering of saffron (Crocus sativus L.) with three corm weights under different LED light qualities. Sci Hortic 303:111202. https://doi.org/10.1016/J.SCIENTA.2022.111202

Zhu J, Zhang YC, Yang LY, Zhou L (2022c) Effect of different LED-lighting quality conditions on growth and photosynthetic characteristics of saffron plants (Crocus sativus L. Photosynthetica 60:497–507. https://doi.org/10.32615/PS.2022.038

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