



Effects of Supplemental Light Quality and Foliar Application with Calcium on Photosynthetic Parameters and Flower Stem Strength of Cut Gerbera (*Gerbera jamesonii* 'Bayadere')

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ABSTRACT

Photosynthetic parameters and stem strength of *Gerbera jamesonii* 'Bayadere' plants were studied after foliar application with calcium chloride (0, 0.5, 1, and 1.5 g L⁻¹) under natural light (NL), red light LEDs (R), blue light LEDs (B) and red + blue light LEDs (RB). Chlorophyll content increased under LED lights and foliar application with calcium chloride. The maximal quantum yield of PSII efficiency (F_v/F_m) and performance index (PI) reached the lowest value under NL conditions. The highest values of photosynthesis rate, stomatal conductance, and transpiration rate (E) were achieved by RB lighting. Under the RB light and calcium chloride spraying, the plants showed the lowest leaf nitrate content, compared to NL conditions. This resulted in the highest leaf nitrate content. The higher the nitrate content, the lower the stem strength. However, the leaf calcium content showed an opposite trend. Sprays of 1.5 g L⁻¹ calcium chloride in combination with RB lighting significantly increased the flower stem strength and reduced stem bending, compared to non-treated plants. In summary, the RB lighting and foliar application with calcium chloride enhanced the growth and flower stem firmness of cut gerbera.

Introduction

Gerbera (*Gerbera jamesonii* H. bolus ex Hook. f.) is an ornamental flower native to South Africa and belongs to the Asteraceae family. Due to its wide range of flower colors and shapes, the popularity of this flower is international. Based on the statistics of FloraHolland (2016), gerbera is placed among the top 10 cut flowers at Dutch flower auctions. In addition to its ornamental value, gerbera is also a model plant for studying the flower formation process (Bhattarai et al., 2021).

Healthy petals and the flowering stem (scape) are vital parts of this plant. Stem length and strength are highly important in terms of quality indices for gerbera cut flowers. Bending of the flowering stem (bent neck) is a major post-harvest disorder of gerbera which reduces its quality and vase life

(Cheng et al., 2020). Bacterial blockage of scape and subsequent shortage of water uptake at the cut surface of the stem is the major cause of bending in gerbera (Perik et al., 2014). However, this bending might also be related to other factors. Lack of mechanical support in the scape is a postulate cause. Perik et al. (2012) reported a correlation between bending and formation of a cylinder of sclerenchyma in stems. Moreover, bending correlated with levels of stem lignification. They concluded that stem bending occurs mainly due to water loss and low mechanical strength in the upper part of the stems. There are some cultural and biotechnological strategies to overcome this bending (Jaberian Hamedan et al., 2019). Management of the light spectrum during plant growth is a useful approach to enhance

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photosynthesis, resulting in better development of lignin tissues (Pawłowska et al., 2018). Moreover, pre-harvest application with calcium is very useful to avoid bent neck and loss of visual quality of gerbera flowers (Aghdam et al., 2019). Light intensity, duration, and spectral composition regulate plant growth and development via different molecular and physiological pathways. Light is used as an energy source for carbon fixation in photosynthesis. Enhancements in photosynthesis lead to the production of more photo-assimilates that are involved in cell wall lignification. Cell wall polymers are mostly energy-rich linked sugars that form the major structural components in plant cell walls, particularly in the thick secondary cell walls that characterize certain tissues. Moreover, many physiological processes during plant growth and development are activated and regulated by light signals (Paradiso and Proietti, 2021). For optimal growth, each plant requires a defined amount of photosynthetic photon flux density (PPFD). In addition to light quantity, light color (spectral quality) affects plant photosynthesis and morphogenesis. For instance, red and blue light are the most important spectra in orchestrating photosynthesis (Paradiso et al., 2011). The development of photosynthetic apparatus is largely affected by red light. Blue light affects stomatal opening and chlorophyll biosynthesis in plants. Plant height and stem elongation are affected by blue and red/far red light. Far red light has a dramatic effect on the flowering of long day plants (Zheng et al., 2019). Alifar et al. (2020a) reported that blue LED lighting prolonged the vase life of carnation cut flowers by improving their antioxidant defense system in petals and enhancing photosynthetic performance in the leaves.

Artificial light sources can change the quantity or quality of light for plants. Depending on the purpose of lighting, different lamp types have applications for plant production. High-intensity discharge (HID) lamps function in greenhouses and plant growth chambers. They have low efficiency and produce high radiant heat, which makes them difficult to use in summer (Magar et al., 2019). In contrast, light-emitting diodes (LEDs) exhibit higher energy efficiency with low thermal emissions when compared to traditional light sources (Cocetta et al., 2017). They are eco-friendly light sources with high durability and long operating lifetime producing light at specific wavelengths (colors) (Rabara et al., 2017). In addition to the production area, LEDs can function during the postharvest of cut flowers. Alifar et al. (2020b) showed that the exposure of

carnation-cut flowers to blue LED light postponed their senescence. This beneficial effect was due to the down-regulation of ethylene biosynthetic genes, with the up-regulation of ABA biosynthetic genes under blue light.

Supplementary lighting is recommended for greenhouse production of cut flowers in regions with low light intensity (Llewellyn et al., 2020). In addition to light energy, lamps with different colors (wavelengths) can drastically affect the growth and postharvest quality of cut-flowers (Alifar et al., 2020a and b). The rate of flower bud initiation and development in gerbera is negatively affected by high temperatures. Thus, growing gerberas at lower temperatures produced by LED lamps were recommended. In research on gerbera, red (85%) + blue (15%) light was substituted for high-pressure sodium vapor lamps with the same photosynthetic active radiation. Flower diameter, flowering stem length, and fresh weight of the flowers increased. In addition, the quality and vase life of the flowers were improved (Llewellyn et al., 2019). In the same study, the growth of gerbera flowers was accelerated in greenhouse conditions under LED lighting (Llewellyn et al., 2020). Bud break, flower bud abortion, re-production of flowering shoots, harvest intervals, and petal coloration in roses highly depend on light (Zieslin and Tsujita, 1990). In potted Phalaenopsis, the number of inflorescences, florets, and flower size increased in response to supplemental LED light (Magar et al., 2019).

The tight binding of calcium ions to pectin molecules maintains cell wall rigidity, thereby providing mechanical support to the plants (Hawkesford et al., 2012; Li et al., 2012). Moreover, polygalacturonase enzyme activity and degradation of middle-lamella are reduced in the presence of calcium (Wehr et al., 2004). Stem lignification by calcium application plays vital roles in water uptake by cut stems (Vanholme et al., 2010). Foliar application with calcium chloride in a bending-sensitive gerbera (*Gerbera jamesonii* 'Rosaline') increased water absorption, followed by delayed stem bending (Aghdam et al., 2019). The authors indicated that lignin, cellulose, and hemicellulose contents of the flower stems increased significantly. Combrink (2017) reported that application with CaCl₂ hardened cell walls of gerbera scape. Applying 50 mM calcium chloride with other compounds that increased vase life delayed the occurrence of bent necks in gerberas (Perik et al., 2014).

In the present work, we studied the physiological responses and flower stem quality of cut gerbera (*Gerbera jamesonii* 'Bayadere') plants grown under different light quality and calcium

supplementation. The role of assimilates in improving physiological processes and stem strength was examined by LED artificial lighting. The role of calcium in enhancing stem stiffness was tested by pre-harvest foliar application with calcium chloride.

Material and Methods

Plant material and experimental conditions

Tissue cultured plugs of *Gerbera jamesonii* 'Bayadere' with 3-4 leaves were purchased from a local supplier. The plugs were transplanted into round 20 cm diameter × 20 cm height plastic pots filled with cocopeat: perlite (70:30 v/v) as the substrate. The pots were placed in a glasshouse and the plants were fertigated by Hoagland's half strength solution (1950) for 10 days. After acclimatization to glasshouse conditions, the plants were fertigated by recommended nutritional formula provided by the supplier. The current experiment was conducted in Rafsanjan city (latitude 30°24'24" N, longitude 55°59'38" E, 1514 m above mean sea level). Climate control equipment in the glasshouse were adjusted to provide day and night temperatures of 25 ± 2 °C and 20 ± 2 °C, respectively. Relative humidity was maintained at approximately 60%. The average photosynthetic photon flux density (PPFD) of natural light in the glasshouse was $115 \mu\text{mol m}^{-2}$

s^{-1} . All light treatments were applied with a photoperiod of 16 h light and 8 h dark.

Light and calcium treatments

The artificial light sources were LEDs of red light (100%), blue light (100%) and combined light sources of red (70%) + blue (30%) with an intensity of $200 \mu\text{mol m}^{-2} \text{s}^{-1}$. Four LED fixtures (Iran Grow Light, Iran) were installed along the center of each bench and were fixed 25 cm above the pot level. Gerbera pots were located centrally on the benches. Each fixture consisted of 24 LEDs and a coverage area of $100 \times 40 \text{ cm}^2$ per meter. The current flow, power, and voltage of the lamps were 350 mA, 24 W, and 265-85 V, respectively. Artificial lighting on the plants was performed from 6 A.M. to 10 P.M. To prevent light contamination, each bench was separated with a cardboard. Plants grown under natural light were regarded as the control. Artificial lighting started after the acclimatization of plants to glasshouse conditions and continued until the end of the experiment. Fig. 1 shows plants under light treatments and spectral graphs of light sources. After the appearance of the first flower bud, calcium chloride was sprayed at 0, 0.5, 1, and 1.5 g L⁻¹. Spraying intervals were 10 days and the plants were sprayed 10 times in total.

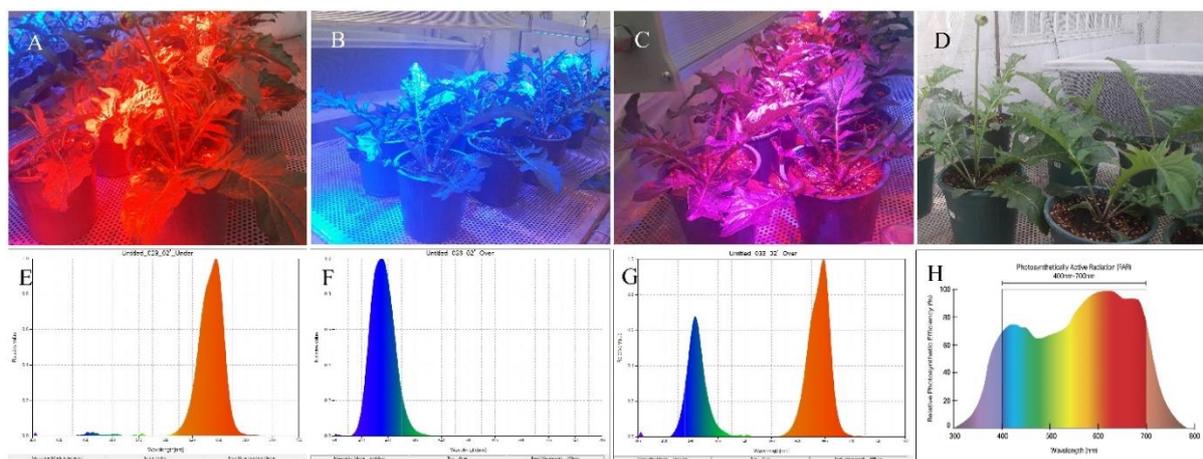


Fig. 1. Photos of the plants grown under 100% red (A), 100% blue (B), and 70% red + 30% blue (C) LED lights and control plants under natural sunlight (D). The light spectra produced by LED lights (E-G) and sun (H) are shown in graphs.

Determination of chlorophyll content and fluorescence

For the destructive measurement of chlorophyll concentration, 0.25 g of fresh leaf was ground in 10 mL of 80% (v/v) acetone. Ground tissue was centrifuged at 3500 xg for 10 min. The pigment

content of the supernatant was determined using a spectrophotometer, described by Lichtenthaler (1987). Moreover, chlorophyll content was measured non-destructively as the chlorophyll content index (CCI) of young fully expanded leaves using a chlorophyll meter (SPAD, Konica,

P502, Japan). The average of three measurements per leaf was recorded as CCI.

Chlorophyll fluorescence was measured with a portable Handy Plant Efficiency Analyzer (PEA, Hansatech, UK) as described by Bulgari et al. (2015). Before measurements, leaf clips (4 mm diameter) were attached to leaves, and after 15 min, the maximal quantum yield of PSII efficiency (F_v/F_m) and Performance Index (PI) were recorded on dark-adapted leaf tissues. Here, PI shows the performance index for energy conservation from photons absorbed by the PSII antenna to the reduction of QB during photosynthesis.

Gas exchange measurements

Gas exchange was measured on attached and fully expanded healthy leaves at the beginning of flowering between 9:30 and 11:30 am. using a portable LCI photosynthesis system (ADC BioScientific Ltd., UK). Net photosynthetic rate (P_n , $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$), stomatal conductance of H_2O (g_s , $\text{mol H}_2\text{O m}^{-2} \text{ s}^{-1}$), mesophyll conductance of CO_2 (g_m , $\text{mmol m}^{-2} \text{ s}^{-1}$), internal CO_2 (c_i , vpm), and transpiration rate (E , $\text{mol H}_2\text{O m}^{-2} \text{ s}^{-1}$) were determined on each leaf. The ratio of P_n to E was calculated and reported as instantaneous water use efficiency (WUE_{ins} , $\mu\text{mol CO}_2 \text{ mol}^{-1} \text{ H}_2\text{O}$).

Analysis of leaf calcium and nitrate concentration

The calcium concentration of the leaves grown under different light conditions and chloride calcium sprays was determined. Four fully expanded leaves were sampled per treatment. Sampling occurred at harvest time. The samples were dried in a hot air oven for 48 h at 60 °C and then ground to get a fine powder. The powdered samples (0.5 g) were digested in 2 N HCl to make an extract. Calcium concentration in this extract was measured by titration with EDTA (Ryan et al., 2001). The nitrate concentration was determined using a LAQUA Twin Nitrate (NO_3^-) rapid response digital meter (LAQUA Twin Nitrate Meter, Spectrum Technologies, Inc., USA). This instrument provides reliable data on nitrate content in plant tissues (Chang and Chang, 2014).

Evaluation of scape quality

Flowers were harvested after one complete ring of matured anthers had formed (the opening of

two rows of flower flags). Cut flowers were individually placed in 250 mL glass bottles containing distilled water. To evaluate flower quality, stem neck diameter (the portion just below the capitulum) and stem bending were measured on each harvested flower. Stem neck diameter was measured using a digital caliper. To estimate stem bending, angles of the capitulum surface concerning the vertical side were measured. Therefore, the scape curvature was measured based on the method described by Çelikel and Reid (2002). In this method, the angle between the main stem (scape) and neck was determined.

Statistical analysis

The experiment was arranged as factorial based on a completely randomized design (CRD) with two factors, including light quality (four levels) and spraying of calcium chloride (four levels) with four replications. Data were subjected to a two-way analysis of variance (ANOVA) and means were separated using Duncan's new multiple range test (DNMRT) at 5% level of significance ($p \leq 0.05$) using SAS 9.1.

Results

Photosynthetic pigments

Results of the photosynthetic pigments measurements of gerbera leaves influenced by different light quality and calcium chloride concentrations are shown in Table 1. Based on the ANOVA, the interaction effect of light and calcium chloride on chlorophyll concentration was significant. However, no considerable variations in chlorophyll a concentration occurred among the different light and calcium treatment groups. Plants grown under R light and sprayed with 1 g L^{-1} calcium chloride exhibited the highest chlorophyll a content (1.26 mg g^{-1} FW). However, this treatment was not significantly different from many other treatments. The lowest concentrations of chlorophyll a under B light (0.73 mg g^{-1} FW) and RB light (1.08 mg g^{-1} FW) occurred using 1 g L^{-1} calcium chloride that showed significant differences compared to the control. Exposure to RB light approximately doubled the amount of chlorophyll b compared to monochromatic R or B light.

Table 1. Chlorophyll concentration (mg g⁻¹ FW) and chlorophyll content index (CCI) of gerbera (*Gerbera jamesonii* 'Bayadere') leaves under different light spectra and spraying with CaCl₂.

Light spectrum	CaCl ₂ (g L ⁻¹)	Chlorophyll a	Chlorophyll b	CCI
NL	0	1.18 ± 0.089 ^{ab}	0.703 ± 0.111 ^{abc}	57 ± 0.753 ^{ab}
	0.5	1.23 ± 0.043 ^{ab}	0.900 ± 0.171 ^{ab}	54 ± 1.256 ^{bcd}
	1	1.16 ± 0.107 ^{ab}	0.588 ± 0.120 ^{abc}	52 ± 0.478 ^{cd}
	1.5	1.25 ± 0.014 ^{ab}	0.730 ± 0.071 ^{abc}	55 ± 0.485 ^{abc}
R	0	1.25 ± 0.010 ^a	0.680 ± 0.049 ^{abc}	45 ± 1.285 ^e
	0.5	1.26 ± 0.015 ^a	0.750 ± 0.087 ^{abc}	50 ± 1.452 ^d
	1	1.23 ± 0.020 ^{ab}	0.668 ± 0.110 ^{abc}	58 ± 0.267 ^a
	1.5	1.18 ± 0.022 ^{ab}	0.445 ± 0.043 ^c	54 ± 1.695 ^{bcd}
B	0	1.19 ± 0.030 ^{ab}	0.520 ± 0.039 ^c	47 ± 0.526 ^e
	0.5	1.18 ± 0.058 ^{ab}	0.588 ± 0.032 ^c	54 ± 0.928 ^{bcd}
	1	0.73 ± 0.050 ^c	0.458 ± 0.201 ^c	57 ± 0.711 ^{ab}
	1.5	1.20 ± 0.004 ^{ab}	0.415 ± 0.054 ^c	53 ± 0.749 ^{cd}
R + B	0	1.25 ± 0.017 ^a	0.730 ± 0.087 ^{abc}	57 ± 0.383 ^{ab}
	0.5	1.23 ± 0.037 ^{ab}	0.738 ± 0.061 ^{abc}	57 ± 0.383 ^{ab}
	1	1.08 ± 0.102 ^b	0.558 ± 0.103 ^c	53 ± 2.881 ^{bcd}
	1.5	1.20 ± 0.014 ^a	0.938 ± 0.088 ^a	55 ± 0.621 ^{abc}

Mean values marked with the same letter within columns do not differ significantly based on Duncan's Multiple Range Test at $p \leq 0.05$.

Chlorophyll content index (CCI) and chlorophyll a fluorescence

The chlorophyll content index (SPAD value) was significantly affected by the interaction effect of CaCl₂ and light quality. The highest value (58) was associated with the treatment of 1 g L⁻¹ CaCl₂ + R light which showed no significant difference compared to 1 g L⁻¹ CaCl₂ + B light. The lowest values were obtained under R (45) or B (47) lighting in the absence of calcium chloride treatment (Table 1).

Chlorophyll a fluorescence shows the maximal photochemical efficiency of photosystem II. PI represents the performance of photosynthetic systems and was only affected by artificial lighting. Plants grown under B light increased in F_v/F_m compared to plants that received natural or R light. However, adding R light to blue LED lamps did not significantly affect this parameter. The PI values of gerbera plants were not significantly affected by exposure to different light qualities. However, the PI values of plants grown under NL conditions were much lower than LED-grown plants (Fig. 2).

Leaf gas exchange parameters

The average net photosynthetic rate and stomatal conductance significantly increased in LED light-treated gerbera plants compared to the effect of the NL treatment. Plants grown under RB light

showed the highest rate of photosynthesis. Using RB light, Pn was approximately 1.5 times higher than leaves under NL. There was no significant difference among NL, R, and B light treatments. Internal CO₂ concentration (ci) and mesophyll conductance of CO₂ (gm) were not significantly affected by light or calcium treatments. Maximal stomatal conductance occurred under combined red and blue light (Fig. 3A and B). The lowest gs occurred under R light, about half of its value in plants exposed to RB LEDs.

The transpiration rate and instantaneous water-use efficiency significantly changed under different light qualities. Calcium treatments did not highly alter these traits. Like the trends obtained for Pn and gs, the E of plants grown under RB light was higher than those grown under the other light sources. The transpiration rate approximately doubled after adding the B light source to the R light. Blue and NL light conditions exhibited no significant impact on E values. In contrast to E and gs, the highest amount of WUEins occurred in plants treated with R light which was approximately two times higher than WUEins in plants exposed to B light (Fig. 3C and D).

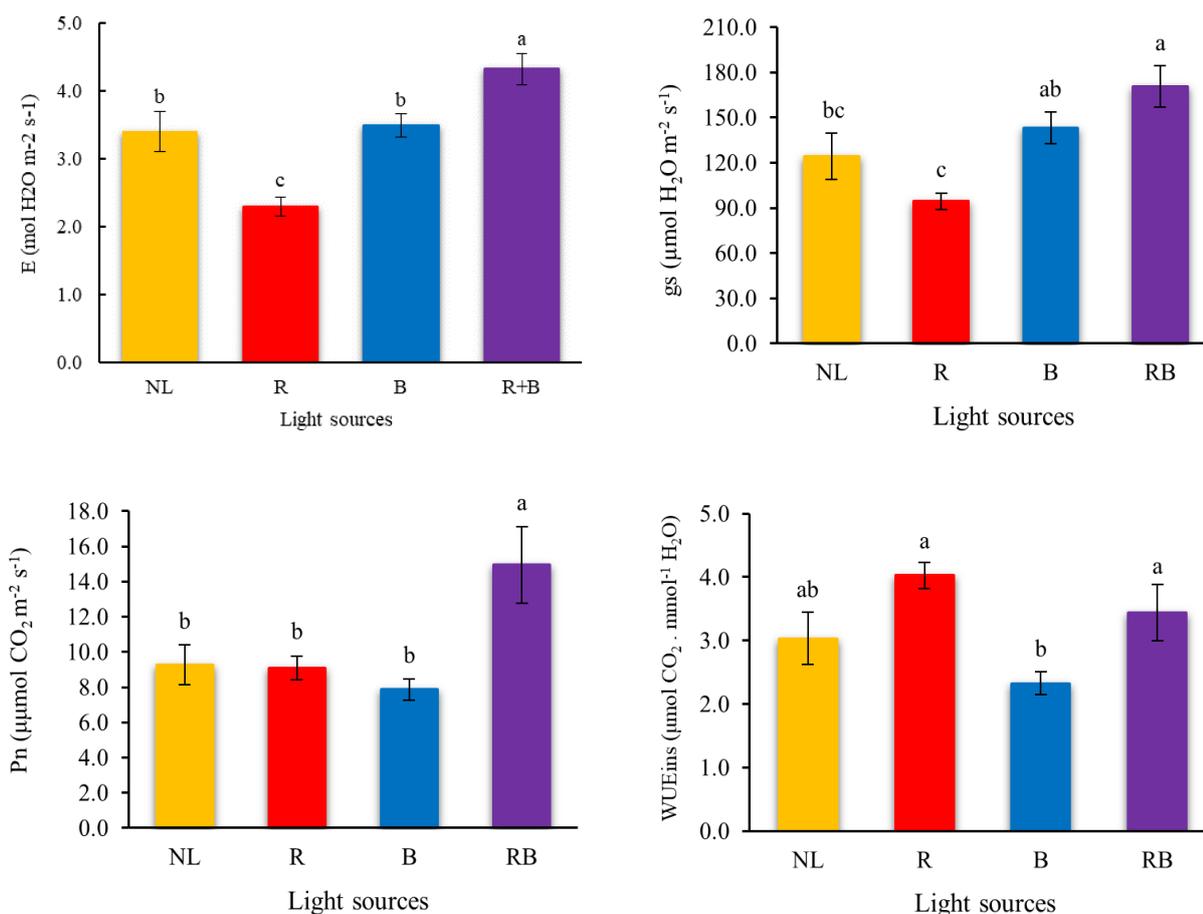


Fig. 3. The photosynthesis indices of gerbera plants grown under four different light sources including natural light (control, NL), red light LEDs (R), blue light LEDs (B) and combined 70% red + 30% blue light LEDs (RB). (A) Net photosynthetic rate (Pn); (B) stomatal conductance (gs); (C) transpiration rate (E); and (D) instantaneous water use efficiency (WUEins). Bars represent mean \pm SD, n = 4. Bars with the same letter are not significantly different ($p \leq 0.05$) according to Duncan's Multiple Range Test.

Leaf calcium and nitrate content

Both calcium and nitrate concentrations in gerbera leaves were significantly affected by the interaction between light sources and calcium chloride. Calcium concentrations in the leaves of plants grown under RB light, treated with $1.5 \text{ g L}^{-1} \text{ CaCl}_2$, showed an increase of more than 3.5-fold, compared to the lowest value (0.05) under NL conditions (Fig. 4). In comparing R and B light LEDs, red light showed a higher increase in leaf calcium content.

The presence of RB light in the spectrum interestingly reduced the amount of leaf nitrate to 60% of its highest value observed in plants sprayed with $1 \text{ g L}^{-1} \text{ CaCl}_2$ under the NL (Fig. 5). In comparing NL conditions and LED lighting, the gerbera plants had more leaf nitrate under the NL.

Scape quality

Statistical analysis showed that light and calcium chloride significantly improved floral scape

quality. Application with RB light or 1.5 g L^{-1} calcium chloride significantly increased neck diameter compared to controls (NL conditions or no application with calcium chloride). However, no significant differences occurred among different light qualities or other concentrations of calcium chloride (Fig. 6). Moreover, measuring stem bending indicated that flower stem strength differed by exposure of gerbera plants to different light qualities or applying calcium chloride. Higher calcium chloride concentrations dramatically reduced stem bending by more than 10-fold. Moreover, stem bending of plants exposed to LED light was at least half of those grown under NL. The flower stem of plants grown under NL conditions, with no calcium spray, showed the highest curvature (65.8 degrees). In contrast, foliar application with $1.5 \text{ g L}^{-1} \text{ CaCl}_2$ on plants under RB light resulted in the lowest curvature (2.4 degrees) (Fig. 7).

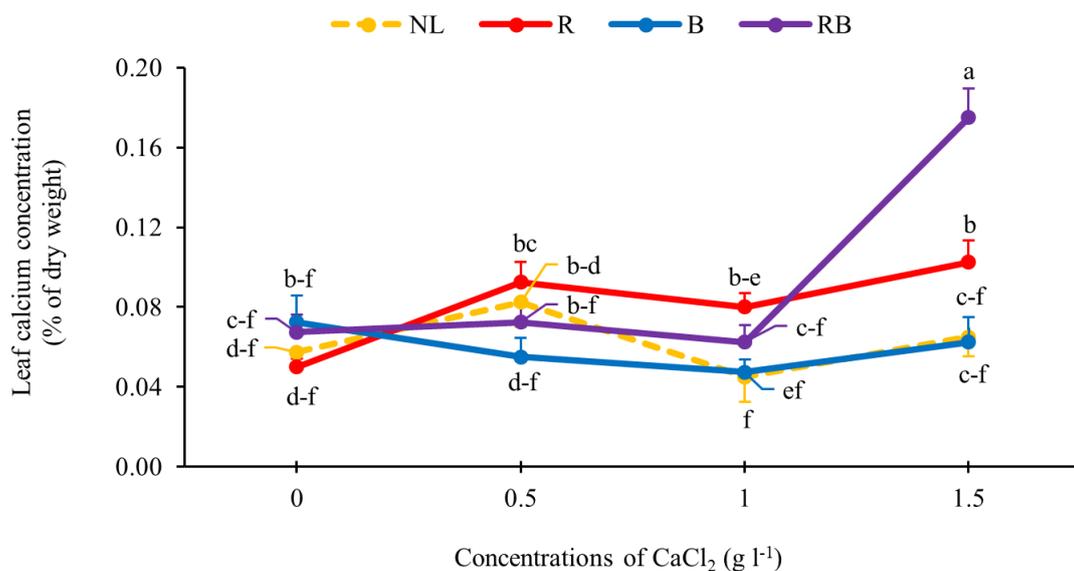


Fig. 4. The interaction effect between calcium and light on leaf calcium concentration of gerbera plants grown under four different light sources, i.e. natural light (control, NL), red light LEDs (R), blue light LEDs (B) and 70% red + 30% blue light LEDs (RB). Bars represent standard errors of four replicates. Values with different letters are significantly different according to Duncan’s Multiple Range Test at $p \leq 0.05$.

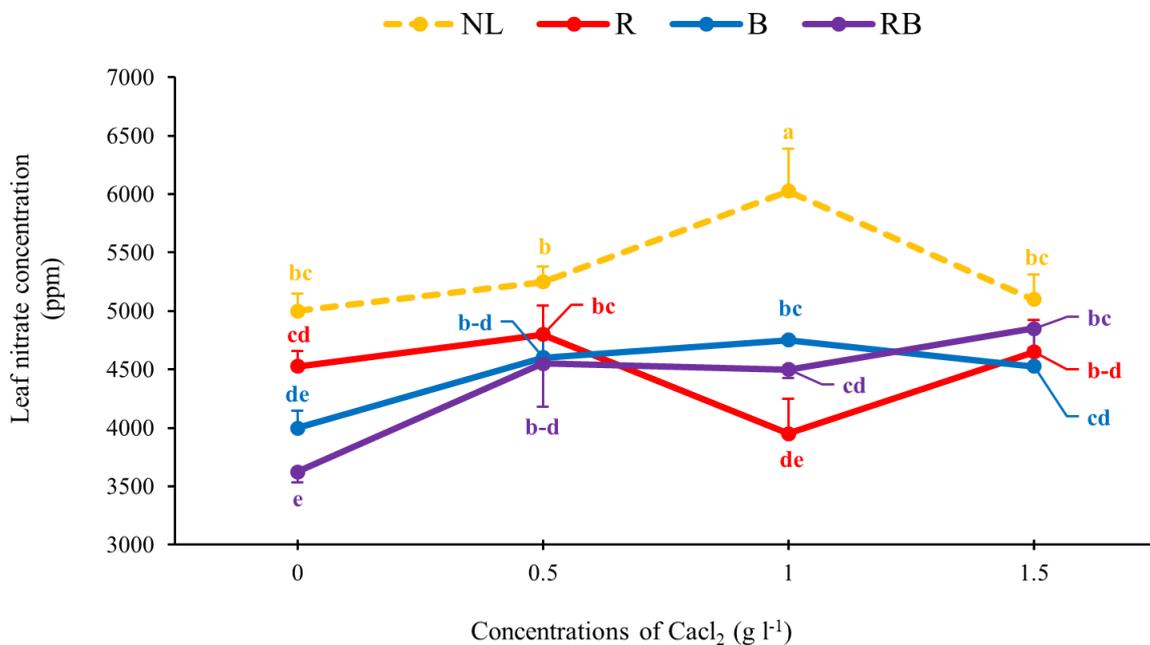


Fig. 5. The interaction effect between calcium and light on leaf nitrate concentration of gerbera plants grown under four different light sources, i.e. natural light (control, NL), red light LEDs (R), blue light LEDs (B) and 70% red + 30% blue light LEDs (RB). Bars represent standard errors of four replicates. Values with different letters are significantly different according to Duncan’s Multiple Range Test at $p \leq 0.05$.

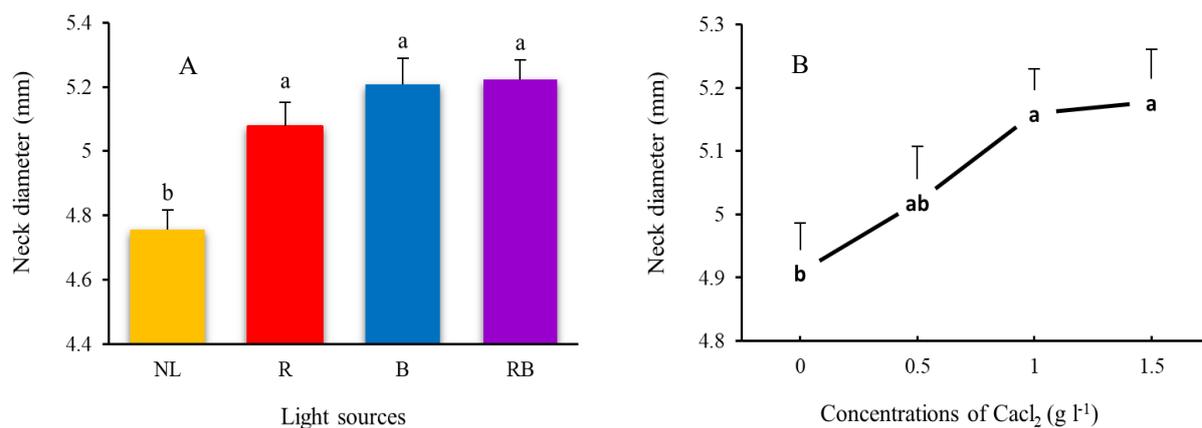


Fig. 6. Effects of different light spectra (A) and concentrations of calcium chloride (B) on neck diameter of gerbera plants grown under four different light sources, i.e. natural light (control, NL), red light LEDs (R), blue light LEDs (B) and 70% red + 30% blue light LEDs (RB). Bars represent standard errors of four replicates. Values with different letters are significantly different according to Duncan's Multiple Range Test at $p \leq 0.05$.

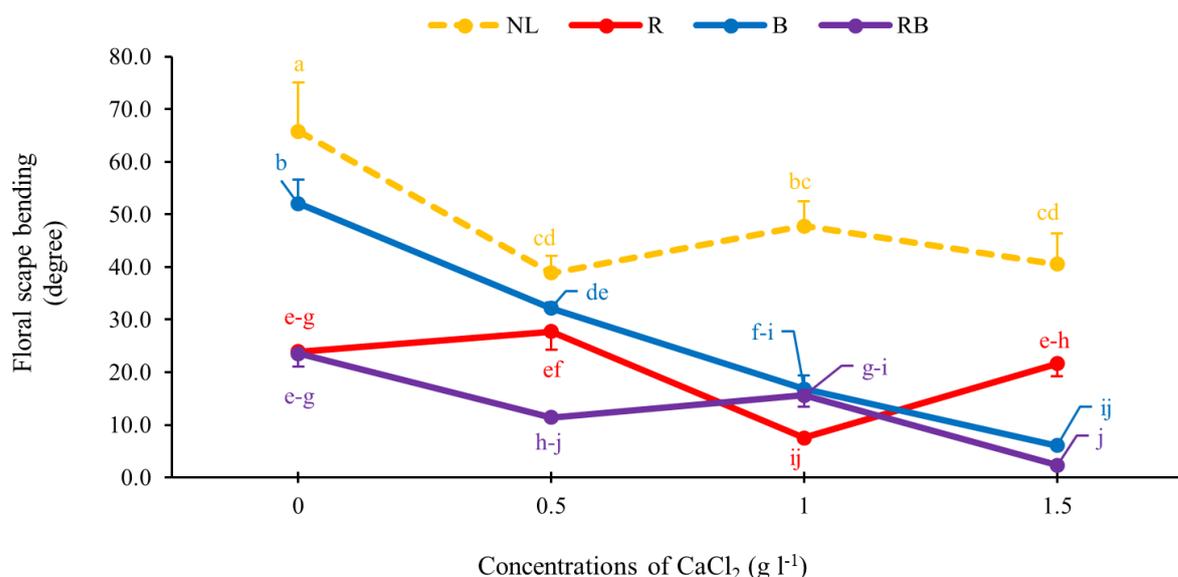


Fig. 7. Scape bending of gerbera flowers under four different light sources, i.e. natural light (control, NL), red light LEDs (R), blue light LEDs (B) and 70% red + 30% blue light LEDs (RB). Vertical bars show standard errors of four replicates. Values with different letters are significantly different according to Duncan's Multiple Range Test at $p \leq 0.05$.

Discussion

Scape bending is a major cause that limits the vase life of cut gerbera. However, manipulating environmental factors and plant nutrition can mitigate stem bending and therefore prolong the postharvest life of this attractive cut flower. In the present study, we report that cut gerbera (*Gerbera jamesonii* 'Bayadere') plants enhanced

their tolerance to bending of floral scape by improving their photosynthesis and stem mechanical strength.

Based on the results of destructive and non-destructive (SPAD values) evaluation of chlorophyll concentration, RB LED light was favorable for biosynthesizing chlorophyll a and b. Mixed LED lights reportedly promoted

chlorophyll production in gerbera (Cioć and Pawłowska, 2020). In chrysanthemums, the highest content of chlorophyll a + b was achieved under R (80) + B (20) LED light (Thanh-Tung et al., 2018). They indicated that both types of LEDs that emit red and blue light are required for the growth of chrysanthemum plants. Fan et al. (2013) proved that chlorophyll biosynthesis in *Brassica campestris* L. increased by simultaneous application with red and blue LED lights. Chlorophylls are involved in light absorption and directly affect photosynthesis in plants. Light quality has a direct effect on photosynthetic pigments in plants. Cryptochromes and phototropin receive blue light and regulate the formation of chlorophyll in plants (Fankhauser and Chory, 1997). We conclude that RB LED light improved the biosynthesis of photosynthetic pigments.

Besides artificial lighting, foliar application with calcium spray showed positive effects on the biosynthesis of chlorophylls. Calcium promotes plant growth by enhancing the uptake of essential nutrients (Ahmad et al., 2015). Some minerals, such as nitrogen and magnesium, are involved in chlorophyll structure. Moreover, when Ca^{2+} binds to calmodulin (CaM), a calcium-binding messenger protein, it triggers biological processes. In chloroplasts, there is an interaction between CaM and NAD kinase, regulated by the Ca^{2+} /CaM complex, and is essential for the biosynthesis of chlorophyll. Chlorophyll content in *Arabidopsis* NADK2 mutants (*nadk2*) reportedly decreased (Hochmal et al., 2015). Moreover, calcium plays an indirect role in chlorophyll biosynthesis by controlling the hydration state of the membranes and the cytoplasm (Pal and Laloraya, 1972).

The current study showed that the performance index (PI) pertaining to the photosynthetic apparatus was much higher under all light qualities compared to NL. Moreover, the highest values of chlorophyll fluorescence (F_v/F_m) occurred under B light. Based on the PI and F_v/F_m data, Cioć et al. (2021) indicated that the performance of the photosynthetic apparatus of in vitro gerbera leaves was reduced under R light. In contrast, involving B LED light could significantly increase the photosynthetic performance. Aliniaieifard et al. (2018) reported the detrimental effects of monochromatic red light on the morphology and photosynthesis of English marigolds. They indicated that red light resulted in leaf deformation (epinasty) and lower F_v/F_m and PI, compared to the full light spectrum. Moreover, most of the energy absorbed by the photosystems dissipated as heat under the monochromatic red-light treatment. In pepper,

seedlings grown under LEDs exhibited significantly higher PI values, compared to plants grown under HPS lamps (Sobczak et al., 2021). Seedapalee et al. (2021) showed that B lighting resulted in the highest F_v/F_m value of *Helianthus tuberosus* L. seedlings compared to the other light sources. Generally, PI is a very sensitive indicator that shows the performance of photosynthetic apparatus. LED-grown leaves reportedly contained more chloroplasts with thicker grana resulting in higher PI value (Sobczak et al., 2021). The F_v/F_m ratio often indicates the maximum quantum efficiency of photosystem II. It appears as the maximum photochemical yield of PSII in the dark-adapted state. Thus, the higher F_v/F_m values under B light, compared to NL conditions, indicated that gerbera plants had a better growth status under artificial lighting.

In this study, the net photosynthetic rate and stomatal conductance increased using different light qualities. The lowest photosynthetic rate was observed under monochromatic blue light. The photosynthesis rate under RB LED light was higher than other lights. No significant differences were observed in internal CO_2 concentration and mesophyll conductance, suggesting that differences in the photosynthetic rate were not due to a limitation of internal CO_2 concentration or gas exchange through mesophyll. The literature review indicates that the effect of light sources on photosynthetic parameters is completely species-dependent. In two *Rosa hybrida* cultivars, plants grown under monochromatic blue light exhibited declined photosynthesis rates despite increasing levels of stomatal conductance and internal CO_2 concentrations (Abidi et al., 2013). The highest photosynthesis rate of in vitro chrysanthemum plantlets occurred under RB LED light (Kim et al., 2004). Lian et al. (2002) reported that the greatest extents of bulblet growth in *Lilium* under RB light may be due to greater photosynthesis. Net photosynthesis in petunia leaves declined during the transition to the flowering phase. However, this reduction was slower in plants grown under RB LEDs than those grown under white light (Phansurin et al., 2017). Photosynthesis in higher plants is a wavelength-dependent process. The spectral energy emitted by RB LEDs is consistent with chlorophyll biosynthesis and absorption spectra (Kim et al., 2004). Thus, LED lights, specifically RB, may enhance photosynthesis. RB light increases photosynthetic pigments and their respective excitation levels (Joshi et al., 2019). Moreover, blue light acts vitally in chlorophyll formation and chloroplast development. Also, blue light

regulates stomata opening for CO₂ uptake during photosynthesis (Mao et al., 2005). Besides blue photons, red light is also involved in signaling to control stomata opening (Tennessen et al., 1994). Thus, combined application with red and blue light could enhance stomatal conductance and improve photosynthesis.

We found that the highest E value occurred in plants treated with RB light. In contrast, water use efficiency was lower under B or RB light. Stomata regulate water evaporation in plants. Light quality clearly impacted the stomatal function and the leaf E value (Matsui et al., 1981). Stomata differentiation is encouraged under blue light (Kang et al., 2009). Moreover, phototropins are responsible for regulating the stomatal function under blue light. Thus, higher E under RB light in our experiment shows that blue light promoted stomata opening and resulted in higher transpiration. However, water use efficiency reflects the ratio of photosynthesis to transpiration. Stomatal conductance has an inverse relation with water use efficiency. Therefore, the higher the stomatal conductance, the lower the water use efficiency. In our research, higher stomatal conductance under B and RB light increased the E value and thus led to lower water use efficiency.

Our measurements showed that RB light and calcium spray resulted in the lowest leaf nitrate. In contrast, plants grown under NL conditions contained the highest nitrate. In the case of leaf calcium content, it was the opposite. Due to health concerns, most research cases on light and leaf nitrate were aimed at vegetable crops. The literature review shows that light quality alters leaf nutrient content. In a research, Nicole et al. (2018) reported that RB light reduced the nitrate level of various lettuce cultivars. Moreover, the leaf nitrate content of baby-leaf spinach declined under different light qualities. In some vegetable species, nitrate content declined under LED lighting compared to those grown under white light (Nájera and Urrestarazu, 2019). Besides food crops, controlling nitrate levels in ornamental plants is particularly important. Nitrogen fertilizers significantly affect the stem properties of trees. In poplar, the structure and composition of the cell wall and lignification pattern was changed in response to the high nitrogen supply (Pitre et al., 2007; Li et al., 2012; Plavcová et al., 2012). In a hybrid of poplar, the lignin content of stems declined using N fertilization (Novaes et al., 2009). A reduction in nitrogen supply improved stem lignification in eucalyptus (Camargo et al., 2014). Nitrate reductase activity is regulated by light. The molecular basis of this regulation was reviewed

by Lillo and Appenroth (2001). They reported that a photosynthesis-dependent post-translational modification is involved in this process. Photosynthesis may influence nitrate reductase activity via photo-assimilates, substrates (nitrate, NADH), and/or by influencing Ca²⁺ flux. We conclude that a small nitrate concentration and a higher calcium concentration in leaves under RB light improved floral scape rigidity. Thus, besides an improved photosynthesis rate, the better balance between NO₃⁻ and Ca²⁺ ions reduced floral scape bending. Artificial LED lighting and calcium chloride spray drastically mitigated stem bending, the major post-harvest disorder of gerbera, compared to plants grown under NL with no calcium chloride treatment. Low mechanical strength and high water loss, especially in the neck, are the main reasons for stem bending in gerbera. Thus, treatments that promote stem lignification and neck diameter result in simultaneous better mechanical strength and improved water uptake, followed by a reduction in stem bending (Camargo et al., 2014). In the case of light, more research on the impact of light intensity and stem mechanical strength provided precise information. Few reports are available on the effect of light spectrum on stem diameter. Yousef et al. (2021) studied the influence of light quality on the morpho-physiological traits of grafted tomato seedlings. They reported that stem diameter was not significantly affected by light quality. However, the higher value of this trait occurred under B light, and plants developed well under RB light. Other research indicated that a mixture of the red and blue LED provides the best light quality to encourage the formation of vascular bundles in grafted tomato seedlings (Wei et al., 2018; Lee et al., 2016).

The main proportion of the total calcium in plant tissues is in cell walls as calcium pectate, which helps the cell wall be rigid and turgid. Thus, calcium plays a direct role in stem strength. Aghdam et al. (2019/G2-6) used calcium chloride spray on a bending-sensitive gerbera (*Gerbera jamesonii* 'Rosaline'). They realized that the lignin, cellulose, and hemicellulose content in floral scapes increased. It resulted in higher water uptake and delayed stem bending. Van Ieperen and van Gelder (2006) showed that calcium contributes to the cell wall formation of the chrysanthemum stem and thus facilitates water uptake. However, there is a positive correlation between light intensity and calcium flow in xylem sap. Light encourages the formation of vascular bundles and thus improves calcium flow and accumulation. Higher calcium transportation, accumulating in sclerenchyma tissues, resulted in

higher mechanical stiffness and strength. Consistent with other studies, we conclude that calcium treatment could enhance the mechanical strength of floral scape by hardening cell walls. Moreover, a higher photosynthesis rate under RB LED improved neck diameter and better formation of vascular bundles, which led to a significant reduction in stem bending.

Conclusion

In the current study, RB LED lighting and spraying 1-1.5 g L⁻¹ calcium chloride improved the main parameters involved in the photosynthesis of cut gerbera (*Gerbera jamesonii* 'Bayadere'). The treatments improved the neck diameter and mechanical strength of the floral scape. Thus, scape bending in these plants decreased, compared to those under NL without the calcium spray. Thus, our findings indicated that the manipulation of the light spectrum and foliar application with calcium sources can assist in preventing the bent neck disorder in cut gerbera.

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Conflict of Interest

The authors indicate no conflict of interest in this work.

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