

## Effects of red and blue light ratio on the morphological traits and flower sex expression in *Cucurbita moschata* Duch.

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### Abstract

Squash (*Cucurbita moschata* Duch.) is an important fruit vegetable that can be long-term transport and storage. Light-emitting diodes (LEDs) are commercially used light sources applied to improve the producing of leaf vegetables in plant factory. However, the influences of LEDs on the plant growth and flower development of fruit vegetables remain unknown. In this study, five effective light quality treatments, including white light, a 10:8 ratio of blue (B) to red (R) light, a 10:4 mixture of blue/red light, red light, and blue light, were used for growing squash and inducing female flowers to maximize production. Our results show that varying light quality influence morphological traits and flower appearance. Both blue and red light improved the development of first and second internodes and induced larger leaves and petiole lengths, whereas 10:4 mixture caused shorter plant heights and decreased internode and petiole lengths. Although 10:8 mixture treatment reduced chlorophyll content, this spectral regime increased leaf number and influenced flower sex development, inducing more female flowers and more fruits. Light quality manipulation thus beneficially influences the growth and flower sex proportion in squash plants. Squash plants under 10:8 mixture treatment exhibited increase in yield, and can be used as a supplementary light treatment in plant factory.

**Keywords:** *Cucurbita moschata* Duch.; light-emitting diodes; floral sex; light quality; morphological traits; squash

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### Introduction

Native to Central and South America, squash plants include several species of creeping and climbing annual herbaceous vines (Ara *et al.*, 2013) in the genus *Cucurbita*, family Cucurbitaceae. The most economically important members of the genus are cultivars of *Cucurbita maxima*, *C. moschata*, Duch. and *C. pepo*. Their global acreage and production are 2.1 million hectares and 22.9 million tons, respectively. Asian countries are the major producers, where China and India together contribute more than 60% of the world's total squash production (FAOSTAT Statistical Database). The *C. moschata* squash cultivar is mostly grown in

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tropical and subtropical regions for its adaption to warmer climates, and is more tolerant of hot and humid weather compared to *C. maxima* (Aydinli *et al.*, 2019; Lu *et al.*, 2019). The *C. moschata* Duch. contains high nutrition, being rich in starch, minerals, and vitamin A, vitamin B1, niacin, and vitamin C, and is widely used in medicine (Lu *et al.*, 2019). In Taiwan, squash is an important vegetable that not only withstands long-term transport and storage, but also increases the supply of summer vegetables and diversifies the vegetable market.

The development of the female flower ovary is the basis of fruit production, and the yields and economic benefits of the squash plant are related to the number and proportion of female flowers. Having more female flowers per plant is essential for higher yields. The cucurbit family is remarkable for its diversity of sex types. They are mainly monoecious, but some cultivars are andromonoecious, trimonoecious gynoecious, or androecious (Cebrián *et al.*, 2022). The expression of flower sex is controlled by numerous factors, including phytohormones, sex determining genes, and environmental conditions. Multiple phytohormones are related to the regulation of sex differentiation in *C. moschata* Duch., among which, ethylene is the main regulator of sex determination, and increases the ratio of female flowers in *Cucurbitaceae* (Li *et al.*, 2021; Manzano *et al.*, 2013). Ethylene promotes carpel development and arrests stamen development in female flowers (Bai *et al.*, 2004). Phytohormones can affect sex expression in *Cucurbitaceae* crops, but the relationships between environmental light factors and sex expression in the family is worth investigating.

Light-emitting diodes (LED) technology can make plant factory production more sustainable because it reduces the carbon footprint of the individual light photon with better manipulation in terms of light quality and intensity (Yang *et al.*, 2017; Lee *et al.*, 2020; Choi *et al.*, 2022; Vitale *et al.*, 2022). As important environmental signals, lights, especially blue light (BL) and red light (RL) regulate plant developmental and physiologic processes through photoreceptors (Casson and Hetherington, 2014). Photoreceptors monitor the light environment and hence help plants determine the timing of crucial developmental events, including flowering and seed germination (Kami *et al.*, 2010). As a plant adaptation mechanism, photo-morphogenesis enables plants to adapt to their surroundings, and is influenced by light receptors such as phytochromes and cryptochromes (Galvão and Fankhauser, 2015). Phytochrome generally assimilates RL, while cryptochrome essentially absorbs BL, and both photoreceptors assist in controlling flowering. The flowering process is widely known to involve photoreceptors associated with BL, promoting flowering and increasing the number of flower buds in chrysanthemums (Park and Jeong, 2020). Moreover, BL enhances plant performance, generates higher levels of Chl content, increases flower bud formation, and promotes flowering in *Kalanchoe blossfeldiana* (Yang *et al.*, 2022). Jeong *et al.* (2014) showed that BL stimulated the elongation of stems and internodes without affecting chrysanthemum flower bud development. Ye *et al.* (2021) demonstrated that BL promoted the flowering of cultivated strawberry seedlings, analysed the global transcriptome of their leaves, and described how BL regulated the flowering at the gene expression level. Besides, photosynthetic performance of grafted watermelon seedlings was improved by BL during healing and acclimatization (Moosavi-Nezhad *et al.*, 2021). Johnson *et al.* (2020) found that compared to continuous RL, continuous BL promotes hypocotyl and petiole growth in arugula (*Brassica eruca*) and mustard (*Brassica juncea*) plants. In *Arabidopsis* and tomato, RL increased shoot growth and development, whereas BL reduced shoot growth and development (Spaninks *et al.*, 2020). Verma *et al.* (2018) reported that mixing RL and BL at a ratio of 4:1 largely increased the leaf number/width/area, root length, stomata width, as well as fresh and dry weights of leaves and roots than RL or BL alone in foxglove (*Digitalis purpurea*). Naznin *et al.* (2016) discussed that the yield of coriander plant had the highest biomass when the exposure of RL and BL was 10:1 rather than 5:1. An application of 7:3 RL:BL to grafted tomato seedlings significantly increased total leaf area, dry weight, total Chl/carotenoid ratio, and soluble protein and sugar contents, and maintained better photosynthetic performance (Yousef *et al.*, 2021). Li *et al.* (2018) implied that pod numbers and yields were greatest in peanut plants under R/B = 3:7 LED lights. A 1:1 mixture of BL:RL allows optimum leaf development, maximum photosynthesis, and Chl content in the cucumber (Hogewoning *et al.*, 2010). However, 1:1 BL:RL did not lead to optimal growth in basil comparing

to any combination of BL, RL, and green light (GL) wavelengths (Carvalho *et al.*, 2016). In addition, Son and Oh (2013) observed that lettuce grew better in the ratio of 47% BL + 53% RL under  $171 \pm 7 \mu\text{mol m}^{-2} \text{s}^{-1}$ . Growing *Rosa* plants under RL and RBW light + FR (far-red) at high a R:FR ratio stimulated flower bud formation (Matysiak *et al.*, 2021). When light spectra change, morphogenetic and photosynthetic responses in different plant species are not the same (Cope and Bugbee, 2013). Adjusting light spectra as required improves plant cultivation in controlling plant growth, development, and flower sex.

Although light spectra have been studied in the development and productivity of many plant species, up-to-date, in squash plants the influence of LEDs on growth and flowering has not been revealed yet, and the effects of different mixtures of RL and BL for inducing female flowers remain unknown. Being monoecious, squash plants have been utilized as a model for studying the genetic control of floral sex differentiation and determination (Martínez and Jamilena, 2021). Although light adjustments help optimize the yield and quality of squash plants, currently, no information is available on the effect of light adjustments for flowering of *C. moschata*. Therefore, we chose different proportions of RL and BL irradiation to identify optimal and suitable light quality conditions for regulating female flowers and growing squash to maximize production. In this study, it is assumed that the morphological response and flower development of squash plants would vary under different RL and BL regimes. An optimal strategy for regulating LED spectra will help induce more female flowers to obtain higher yields in *Cucurbitaceae* crops and maximize economic benefits for *Cucurbita* growers.

## Materials and Methods

### *Plant material and cultural practice*

Seeds of the 'Strong Man' cultivar were purchased from Known-you Seed Company (Taipei, Taiwan). 'Strong Man' is a grafting rootstock variety with cold-tolerant ability for watermelon and bitter melon in Taiwan. Culture practices, including watering and fertilization, were described in our previous paper (Lin *et al.*, 2020). In brief, seeds were sterilized with 2% sodium hypochlorite containing a few drops of Tween-20 (Sigma-Aldrich, MO, USA) for 5 min, washed with sterilized distilled deionized H<sub>2</sub>O five times, and seeds then germinated at 30 °C for three days. Germinated seeds were sown in a commercial potting mix of sand, peat moss, and Perlite (1:1:1, v/v/v; Known-you Seed Co., Taipei, Taiwan) in 3-inch plastic pots (9 cm diameter), and grown in a growth chamber with a 12 h photoperiod (25/20 °C day/night temperature and 70% relative humidity) under  $150 \mu\text{mol m}^{-2} \text{s}^{-1}$  photosynthetic photon flux density (PPFD) light. Plants were watered with a half-strength Hoagland solution (Hoagland and Arnon, 1950), with 200 mL<sup>-1</sup> being applied to each pot every other day for 43 days after sowing (DAS). Squash plants with uniformly growth at the cotyledon-open stage (7 DAS) were raised and maintained in the above-mentioned growth room under different light treatments at a constant 25 °C with a 12/12 h light/dark photoperiod for another two weeks (21 DAS). Plant growth was monitored starting from the two-leaf developmental stage (21 DAS).

### *Light treatments*

Light spectra treatments for squash seedling growth began at 7 DAS, and consisted of the five following commercially available LED arrays designed by Hi-Point Technology (Kaohsiung, Taiwan):

- (1) white light (cool white diodes) control treatment (WL),
- (2) blue light and red light (10:8) as 1BL/0.8RL, short as R/B (0.8),
- (3) blue light and red light (10:4) as 1BL/0.4RL, short as R/B (0.4),
- (4) monochromatic red light (short as RL), and
- (5) monochromatic blue light (short as BL).

The BL (with a peak at 453 nm), RL (with a peak at 660 nm), and WL (at 410~755 nm) spectral distributions from LED devices were measured with a spectral radiometer (Hi-Point Technology, Kaohsiung,

Taiwan) in the 380~780 nm range with 1 nm steps (Figure S1). A calibrated spectroradiometer (LI-COR1800, Lincoln, NE, USA) was placed horizontally in the experiment cabinets and the sensor covered by the vessel's glass lid. The lamp's 1008-LED luminous area was 60× 80 cm, and the spectrum was recorded at the youngest leaf of the squash plants. Each lighting experiment was run in a growth room maintained a 12/12 h light/dark photoperiod under a light intensity of 150  $\mu\text{mol m}^{-2} \text{s}^{-1}$ , which was measured daily above the squash plant canopy and maintained by adjusting the distance of the LEDs above it. For each light treatment, the spectral distribution was measured at three different locations within the plant growing areas.

Ten squash plants for each light treatment were employed with a completely randomized design, and used for the below-listed growth measurements for a month. The position effects were minimized by randomly located replications among light treatments.

#### *Plant growth measurements and determination of total chlorophyll content*

Plant growth measurements included plant height, leaf number, stem perimeter, first and second internode lengths, leaf (fully expanded third leaf) length and width, and petiole length of the fully expanded third leaf. During lighting treatments, data were collected at 21 DAS (14 d exposure to light treatments), 27 DAS (20 d exposure to light treatments), 30 DAS (23 d of exposure to light treatments), and 43 DAS (36 d exposure to light treatments) from March to April 2022. Plant height was measured between the cotyledon and youngest leaf node. The first internode length was measured between the cotyledon and first leaf node, and the second internode length between first and second leaf nodes. Stem perimeter was measured by wrapping the stem with a cotton thread and then measuring the thread with a vernier ruler. Leaf length and width, and petiole length from the third leaf, were measured by vernier ruler. All leaves longer than 1 cm were counted to determine the number of leaves per plant.

The non-destructive measurement of leaf total chlorophyll content (TCC) can be rapidly and accurately achieved by SPAD (soil and plant analysis development) analyzer (SPAD-502 Chlorophyll Meter, Konica Minolta, Tokyo, Japan), and can help advance the interpretation of the photochemical process in plants. The positive linear relationship between SPAD values and TCC has been reported (Ling *et al.*, 2011). Squash leaf TCC was measured with a SPAD-502, and was represented by the SPAD readings. The fully expanded first, second, and third leaves were separately inserted into the measuring head, and three readings were collected from different location of each leaf and averaged. Data from the first, second, and third leaves per plant and ten plants per treatment for each treatment were taken for further analysis.

Squash cv. "Strong Man" is monoecious, with separate male and female flowers. At the flowering stage, the number of flowers was monitored at 43 DAS, and the sum of male and female flowers in all flower-stalk branches per plant were counted at 43 DAS. The number of fruits per plant was recorded at the end of the experimental period (43 DAS).

#### *Statistical analysis*

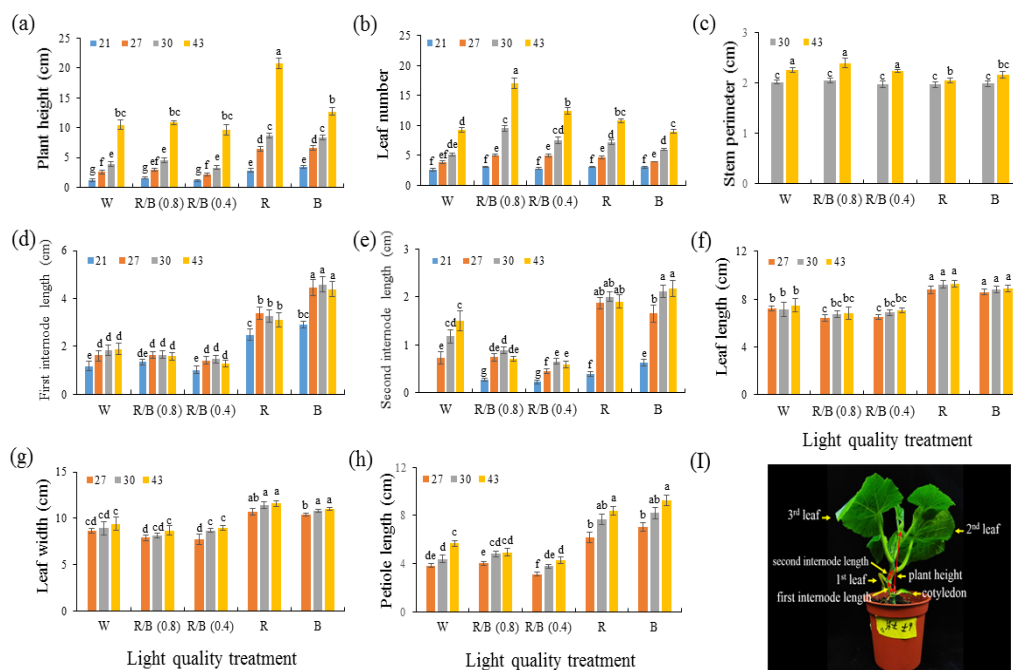
Ten squash plants were randomly planted under each light treatment, and all analyses were performed until the end of the experimental period (43 DAS). One-way ANOVA was used here to determine the significance differences of leaves, total CC, flowers, and fruits measurements among light regimes to different time points. The least significant difference (LSD) test at  $p \leq 0.05$  was applied to separate the means of significant values by using SAS 9 (SAS Institute, Cary, NC, USA).

## **Results**

#### *Plant growth and morphological performance*

Figure 1 shows distinct responses to different light quality treatments. All plant heights and leaf numbers of squash grown for 43 DAS had significantly taller and greater values than at other measurement dates (Figures

1a, b). Notably, the tallest plant height (21 cm) and highest leaf number (17) grown for 43 DAS were detected under RL and R/B (0.8) treatments, respectively. Additionally, stem perimeters of squash grown for 43 DAS under WL, R/B (0.8), and R/B (0.4) treatments had significantly larger values than at 30 d (Figure 1c), and the largest stem perimeter (2.4 cm) at 43 DAS was measured under R/B (0.8) treatment. Stem perimeters were only measured at 30 and 43 DAS due to their being too small prior to then. Significantly larger lengths of first (3.1~4.4 cm) and second (1.9~2.18 cm) internodes occurred when grown under RL and BL treatments compared to other lighting treatments at 27, 30, and 43 DAS (Figures 1d, e). In all lightings, the first and second internodes of all plants at 27, 30, and 43 DAS were larger than those at 21 DAS. Plant leaf length (8.8~9.3 and 8.6~8.9 cm), leaf width (10.7~11.6 and 10.4~11.0 cm), and petiole length (6.2~8.4 and 7.1~9.3 cm) of the third leaf at 27, 30, and 43 DAS of exposure to RL and BL treatments were significantly larger than those in other lighting treatments (Figures 1f, g, h). Moreover, no significant differences in leaf length were observed in all plants grown in same lighting treatment at all measurement date (Figure 1f). As the number of days of growth increased, slowly increasing petiole length was observed under all light applications (Figure 1h). Leaf length and width and petiole length in all tested plants were only measured at 27, 30, and 43 DAS due to the third leaf not being fully open until 27 DAS.



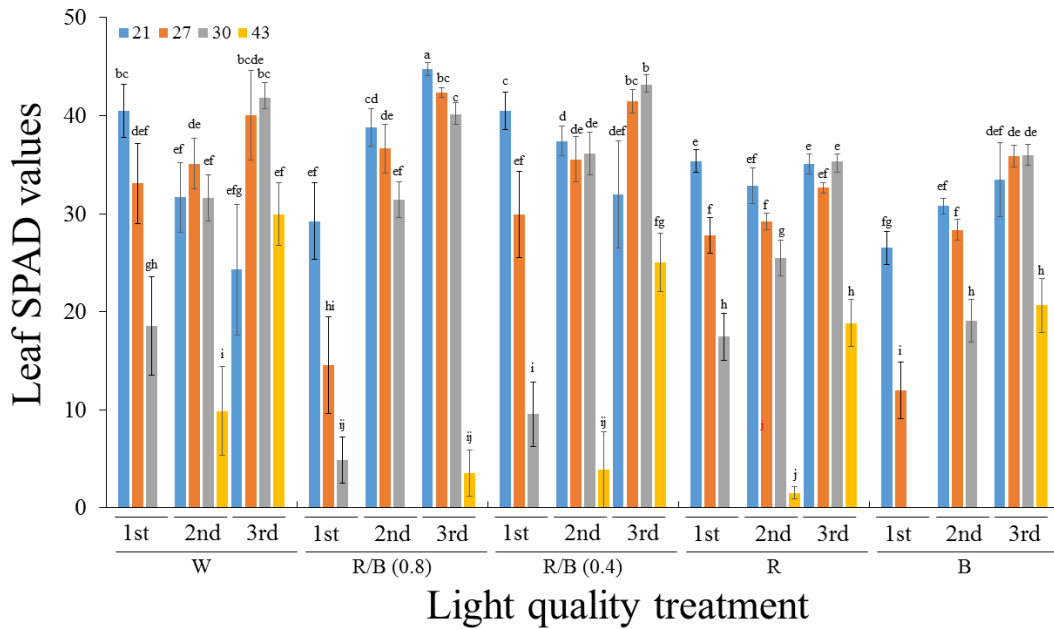
**Figure 1.** Morphological traits of squash plants under different light quality treatments. (a) plant height, (b) leaf number, (c) stem perimeter, (d) first internode length, (e) second internode length, (f) third leaf length, (g) third leaf width, and (h) third petiole length of the fully expanded third leaf at 21, 27, 30, and 43 days after sowing (DAS) of exposure to light treatments with white light (W), mixture of blue and red light as R/B (0.8), mixture of blue and red light as R/B (0.4), red light (R), and blue light (B). (I) This diagram shows the data collection site

Vertical bars indicate the means  $\pm$  standard error ( $n = 10$ ). Different letters indicate statistically significant differences among light treatments over the time period ( $p < 0.05$ ) according to one-way ANOVA.

### Total chlorophyll content

Leaf CC values of squash plants cultured under various light regimes are shown in Figure 2. Generally, as the number of days increased, first and second leaf CC decreased under all lighting treatments except for the second leaf CC under WL treatment. The highest first leaf CC (40.5 SPAD value) was found in WL and R/B

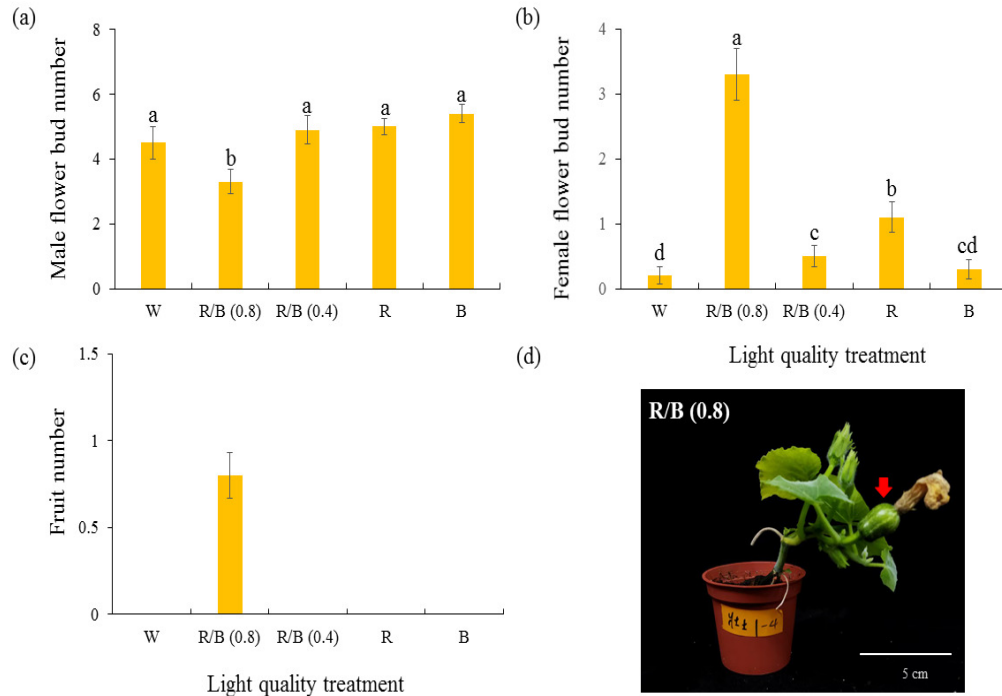
(0.4) treatments at 21 DAS, while the highest second and third leaf CCs (38.8 and 44.7 SPAD value) were found in R/B (0.8) treatment at 21 DAS. No significant differences were observed in all second leaf CCs among all light spectra at 21 and 27 DAS. Significant higher third leaf CCs were detected in plants grown under R/B (0.8), RL, and BL treatments at 21, 27, and 30 DAS, in comparison with 43 DAS. Furthermore, the highest (44.7 SPAD value) and lowest (3.5 SPAD value) third leaf CCs were found in R/B (0.8) treatment at 21 and 43 DAS, respectively. No SPAD values of the first leaves at 43 DAS were recorded due to their being already abscised from stems. Second leaves at 43 DAS under R/B (0.8) and BL treatments were also abscised, leading to no SPAD data on them.



**Figure 2.** Total chlorophyll content (SPAD value) of first, second, and third leaves of squash plants at 21, 27, 30, and 43 days after sowing (DAS) of exposure to light treatments with white light (W), mixture of blue and red light as R/B (0.8), mixture of blue and red light as R/B (0.4), red light (R), and blue light (B). Vertical bars indicate the means  $\pm$  standard error ( $n = 10$ ). Different letters indicate statistically significant differences among light treatments over the time period ( $p < 0.05$ ) according to one-way ANOVA.

#### *Flower and fruit numbers*

The sexes of squash plant flowers displayed differences in response to different light quality regimes. After applying R/B (0.8) treatment, male and female flower numbers at 43 DAS were significantly fewer, although still greater than under other lighting treatments (Figures 3a, b), resulting in their fruit appearing in the R/B (0.8) treatment only at the end of the experiment (Figures 3c, d). The fruit was considered parthenocarpic, since it grew in the absence of pollination due to the corolla being closed. Moreover, there were no significant differences in the effects of WL, R/B (0.4), RL, and BL on male flowers, although RL stimulated more female flowers than WL, RL, and BL irradiances.

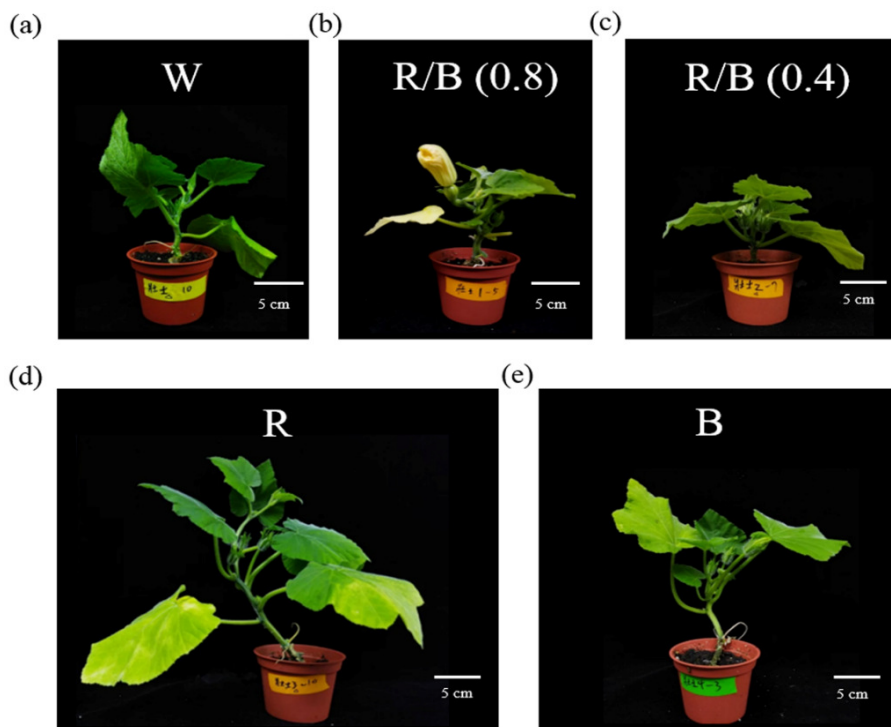


**Figure 3.** Flower traits of squash plants under different light quality treatments. Numbers of male flower buds (a), female flower buds (b), fruits (c), and ovaries of pistillate flowers continued until producing a parthenocarpic fruit labeled with an arrow (d) at 43 days after sowing (DAS) of exposure to light treatments with white light (W), mixture of blue and red light as R/B (0.8), mixture of blue and red light as R/B (0.4), red light (R), and blue light (B) treatments

Vertical bars indicate the means  $\pm$  standard error ( $n = 10$ ). Different letters indicate significant differences in statistics among light adjustments ( $p < 0.05$ ) according to one-way ANOVA. Scale bar = 5 cm.

## Discussion

Different light spectral distributions applied in this study from 7~43 DAS influenced squash plant growth and flower appearance, but these effects were not lethal and all plants did not suffer damage to their leaves. The lower leaves of most plants looked epinastic and senescent (Figures 4b, d), but most leaves appeared healthy and tended to be unaffected, and exhibited adaptive morphologic plasticity. Squash leaves appeared to be sensitive to R/B (0.4), which caused shorter plant heights and decreased internode and petiole lengths at 43 d, but still produced compact plants with greener leaves (Figure 4c). Although there was no difference in stem perimeter at 30 DAS in all light qualities, stem perimeters under RL and BL were visually thinner than the other lightings at 43 DAS, resulting in RL- and BL-treated plants being spindlier, leggier, and taller than other treatments (Figures 4d, e). BL or RL alone applied to squash plants obviously improved the development of first and second internodes and induced larger leaves in terms of leaf length, leaf width, and petiole length. This indicated that blue and red wavelengths stimulated cell division and expansion to act directly on plant morphogenesis, especially in the early stage of development. Moreover, the RL regime was more beneficial for natural plant growth, with increased plant height exhibiting a tall appearance. RL might better penetrate the plant canopy than the other light treatments. Perhaps RL treatment achieved a balanced spectral environment by supplementing a favorable amount of red irradiance to plants at  $150 \mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD, followed by inducing leaf structure and increasing internode length and plant height.



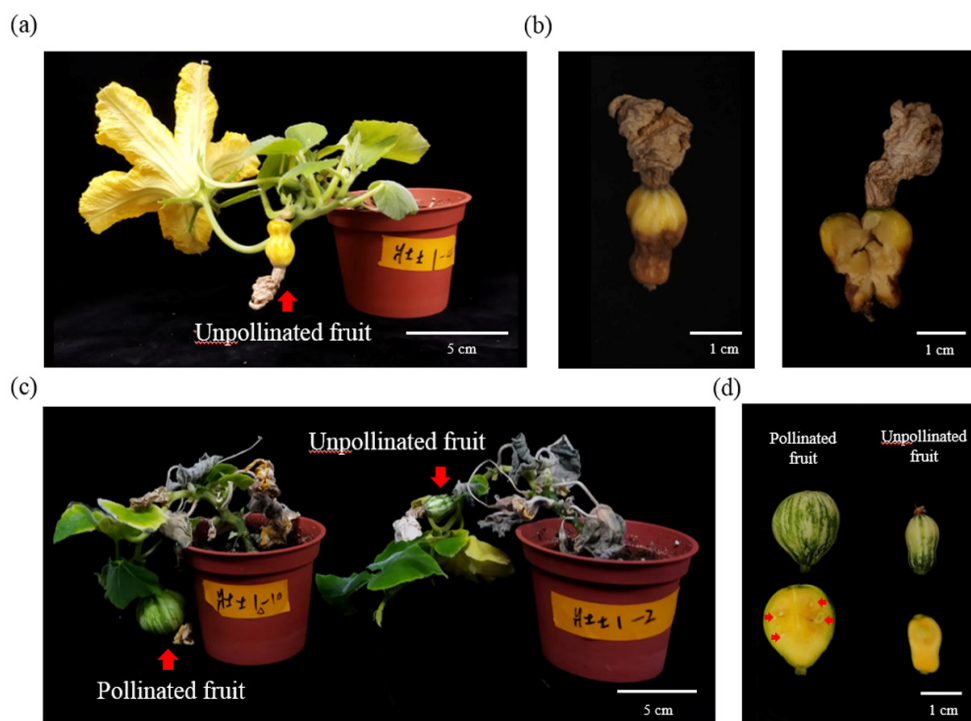
**Figure 4.** Representative views of squash plants grown under five different light quality regimes: (a) white light as W, (b) mixture of blue and red light as R/B (0.8), (c) mixture of blue and red light as R/B (0.4), (d) red light as R, and (e) blue light as B at 43 days after sowing (DAS). Scale bars = 5 cm.

In this study, the peak emissions of WL, BL, R/B (0.8), and R/B (0.4) treatments coincide with the absorption peaks of Chl *a* and *b*, and reported wavelengths are at their respective maximum photosynthetic efficiencies. Figure 2 shows that the TCCs of third leaves under RL and BL treatments were relatively low compared to R/B (0.4) at 43 DAS. In addition, obviously faster aging and senescence rates of the third leaf were observed in the R/B (0.8) treatment at 43 DAS compared to other lighting treatments. Thus, TCCs of third leaves at 43 DAS were significantly reduced as R/B (0.8) lighting was applied, so the leaves of the R/B (0.8) treatment were a visually and slightly lighter green compared to other lightings (Figure 4b). It is possible that R/B (0.8) treatment suppressed Chl synthesis, resulting in leaves becoming chlorotic, epinastic, and senescent at 43 DAS. Nevertheless, R/B (0.8)-treated plants had the greatest leaf numbers and stem perimeters, showing that R/B (0.8) further increased plant growth and development. Most interestingly, R/B (0.8) treatment markedly induced flowering, resulting in increases in flower and fruit numbers. Thus, R/B (0.8) plays an important role in flower development by obviously promoting female flowers and inhibiting male flowers. A high proportion of R/B (0.8) within the light spectrum, being more energetic, may cause light avoidance phenomena in chloroplasts, reducing TCC and increasing female flowering, and confirming that modulating the light spectrum is a valuable tool for controlling and selecting specific characters in this cultivar, especially in indoor environments. Therefore, R/B (0.8) light supplementation may be the most effective technique for inducing flowering, and it can contribute to the commercial production of squash plants.

R/B (0.8) treatment at 43 DAS induced more female flowers than other light treatments (Figure 3b), developed more pistillate flowers and ovaries/fruits, and fruit grew normally in the absence of pollination (parthenocarpic fruit) (Figures 5c and d, right panels). The petal growth rate was reduced in female flowers, and exhibited visual deterioration, abscission, and disintegration, followed by ovary abortion and subsequently shrunken and dwindled parthenocarpic fruit set when not pollinated (Figures 5a and b). Conversely, when



these female flowers were artificially pollinated during anthesis, their ovaries continued to grow and fruit set was concomitant with seed production in the 7 DAS after anthesis (Figures 5c and d, left panels). Pollination was attempted in female flowers and fruits were able to set seeds, since pollen was fertile in other male flowers. *C. moschata* Duch. cultivar ‘Strong Man’ like many cucurbits, is a monoecious plant with unisexual flowers. If the flower was pollinated, then development of the ovary continued and the fruit set, whereas growth of the ovary was aborted in the absence of pollination and fertilization. Generally, the number of male flowers was much larger than female flowers in squash plants. However, increasing female flowers per plant is necessary for higher yields. Meanwhile, more and more attention was drawn on the close relationship between sex type and yield in the breeding of cucurbits (Li *et al.*, 2019). Because of their suitable plant height (averaging 10.8 cm in the R/B (0.8) treatment at 43 DAS), squash plants can be produced using three-dimensional (3D) industrial production systems of plant factory, and the cultivation systems can facilitate both production and crossbreeding operations in squash without land.



**Figure 5.** Fruit development under mixture of blue and red-light R/B (0.8) treatment. (a) Phenotype of anthesis on the development of the ovary and corolla of pistillate flowers cultivated under R/B (0.8) treatment at 43 days after sowing (DAS). (b) Petal growth rate was reduced in the female flower and it was visually deteriorated and abscised (right panels), followed by ovary abortion and subsequent shrinkage and dwindled parthenocarpic fruit set when not pollinated (left panel). (c) Comparisons of the growth of corolla and ovary without artificial pollination (left panels) and unpollinated parthenocarpic fruit with deteriorated female flower (right panel). (d) Longitudinal sections comparing the growth of fruit set with seed production (labeled with red arrows) following anthesis (left panel), and shrunken and dwindled parthenocarpic fruit set without pollination (right panel). Scale bars for A and C = 5 cm; B and D = 1 cm.

Flower sexual differentiation was influenced by multiple cues, including phytohormones, sex determining genes, and environmental factors. Auxin, ethylene, gibberellin, cytokinin, abscisic acid, brassinosteroid, and salicylic acid can direct the sex expression of *Cucurbitaceae* crops (Zhang *et al.*, 2017). So far, the role of light quality in the sex determination of squash plants is unknown. Our study helps elucidate

different light regulation in squash flowering. The R/B (0.8) light illumination positively regulated the formation of female flowers during floral sex differentiation in the squash plant. Further research is required to understand the role that R/B (0.8) plays in the process of ethylene induction of female flowers and whether the ethylene induced by R/B (0.8) treatment also regulates hormonal balance in female flowers. It is also worthwhile to conduct research that explores the exact genomic mechanism behind the development of hormones under the influence of R/B (0.8).

### **Conclusions**

RL, BL, R/B (0.8), R/B (0.4), and WL were used for growing squash plants to study the effects of different light qualities on the development its leaves and flowers. We found that different LED spectra were capable of triggering various morphological traits and flower sexual appearances. RL increased first and second internode lengths, leaf length and width, petiole length, and plant height at 43 DAS, whereas R/B (0.4) decreased plant height, both internodes, and petiole lengths. BL was conducive to squash growth by increasing internode length, leaf length and width, and petiole length. Moreover, R/B (0.8) promoted leaf number and stem perimeter, accelerated aging and senescence, and increased female flowering but decreased male flowering. Since the regulation of sex determination can directly influence plant yield and quality, our findings can upgrade the LED culture systems with comply with commercial requirements for the rapid, large-scale, and precision management of squash production.

### **Authors' Contributions**

Conceptualization, HHL and KHL; methodology, YCC; validation, HHL; investigation, YCC and QEW; resources, HHL and KHL; data curation, HHL; writing—original draft preparation, HHL and KHL; writing-review and editing, HHL and KHL; supervision, HHL; funding acquisition, HHL and KHL.

All authors read and approved the final manuscript.

### **Ethical approval** (for researches involving animals or humans)

Not applicable.

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## Conflict of Interests

The authors declare that there are no conflicts of interest related to this article.

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