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Research Article

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The effects of preharvest LED light, melatonin and AVG treatments on the quality of postharvest snapdragon and vase life

Diying Xiang[1,3†] Chi D. Nguyen[1,2†], Liz Felter[1], David Clark[2], Heqiang Huo[1,2*]

[1]Mid-Florida Research and Education Center, University of Florida, Apopka, FL, USA 32703, [2]Department of Environmental Horticulture, University of Florida, Gainesville, FL, USA 32611, [3]College of Horticulture, Hebei Agricultural University, Baoding, Hebei, China, 07100
[†]Equal contribution to this study

ABSTRACT

Snapdragon (*Antirrhinum majus*) is one of the top ten fresh-cut flowers in the United States; however, its vase life is shorter than other flowers, limiting its marketability. The purposes of this study were to test the effects of LED light, exogenous melatonin and ethylene production inhibitor, AVG, on the quality of snapdragon and the prolongation of vase life. Our results showed that snapdragon treated with 10 h white light followed by 6 h blue light (WB) inhibited stem elongation and lengths of the inflorescences, reduced the number of florets and vase life. On the contrary, snapdragon treated with 10 h white light, 3 h red light, 3 h blue light (WRB) significantly promoted stem elongation, lengths of the inflorescences, and increased the size and number of florets. The lengths of stems and inflorescences increased significantly in all melatonin treatments while quantity and size of florets only increased with 200 $\mu mol \cdot L^{-1}$ melatonin application. Noticeably, vase life was significantly extended with 200 $\mu mol \cdot L^{-1}$ melatonin application and shortened with WB treatment. In contrast to melatonin, all AVG treatments resulted in decreases of the floret size; and changes in stem elongation and inflorescence length were only observed in the treatment with 100 $\mu mol \cdot L^{-1}$AVG. These results showed that pre-harvest treatment with WBR and melatonin can effectively improve the postharvest quality of snapdragon's flowers and 200 $\mu mol \cdot L^{-1}$ of melatonin extended the vase life.

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***Corresponding Author:**
Heqiang Huo,
Email: hhuo@ufl.edu

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INTRODUCTION

Snapdragon (*Antirrhinum majus* L.) is a popular ornamental plant known for its colorful and fragrant flowers. Snapdragons have decorated many gardens as bedding plants and their slender spikes establish line and structure in flower arrangements. Fresh-cut snapdragon sales value reached $12.18 million in the United States, making it a top ten fresh-cut flower in 2015 [1]. Due to snapdragon's sensitivity to ethylene, which induces flower abscission and plant wilting; postharvest problems such as poor vase life and stem bending hinder their production and marketability [2]. Treatments with silver thiosulfate (STS) or 1-methylcyclopropene (1-MCP) to inhibit ethylene action after harvesting to increase shelf life of fresh-cut flowers are commercially practiced. However, STS may raise environmental and health concerns, and it is still difficult to massively and uniformly apply gaseous form of 1-MCP to ornamental plants [3,4,5,6]. Extensive studies have focused on extending vase life through postharvest treatments, but few studies have examined how treatments during the production stage of snapdragons would affect plant quality and subsequent post harvest longevity [7,8]. Therefore, the objective of the present study was to examine how snapdragons treated at the pre-harvest stage would affect the flowers quality and longevity.

Light is crucial for plant growth and development because it controls not only photosynthesis but also flowering time and morphogenesis. Specifically, different wavelengths of light affect the synthesis of phytochemicals that regulate the cellular signaling processes involved with growth and development. Light-emitting diodes (LEDs) have been extensively utilized to investigate many aspects of horticulture research, including bioactive compounds production, disease resistance, and postharvest quality [9,10]. The LEDs are gaining popularity and attracting a broad application to agricultural industry due to their energy efficiency, durability, and the ability to control different spectral wavelengths in comparison to the traditional fluorescent light. The cost-effectiveness and benefits of LEDs also make it an attractive tool for nursery production. Postharvest studies utilizing LEDs primarily focused on fruits and vegetables while information of LEDs effects on ornamental plants is scarce. It has been demonstrated that White, Red, and Green light extend the shelf life and improve nutritional qualities of leafy vegetables, tomato, strawberry, and banana [11,12,13]. Recently, the use of LEDs was shown to delay postharvest senescence of garland chrysanthemum [14]. Considering the multifunctional application of LEDs, it is appropriate to test the effects of specific wavelength of LEDs on snapdragons flowers quality and vase life.

In addition to preharvest treatment of snapdragons using LEDs, exogenous application of N-acetyl-5-methoxytryptamine (melatonin) and aminoethoxyvinylglycine (AVG) were also carried out to examine their effects on plant quality of snapdragon. Melatonin was first discovered in plants in 1995 and present in different parts of plants including leaves, stems, roots, fruits, and seeds depending on the plant species [15,16]. Since then, various studies on the protective effects of melatonin by acting as an antioxidant to remove free radicals have been reported extensively for combating abiotic stresses [17,18,19]. Recent studies expanded melatonin's effects and widen the research to include seed germination, plant tissue culture, and post harvest [20,21]. Notably, exogenous melatonin was shown to improve tomato fruit storage quality, reduce cold injury in peach fruit, alleviate browning in fresh-cut pear fruit and degradation of cassava roots [22]. Although evidences of melatonin's beneficial effects on plants are widespread, little information is found on fresh-cut flowers. AVG is a well-known inhibitor of ethylene biosynthesis and routinely applied to preserve fruits freshness [23,24,25]. Compared with 1-MCP, water-soluble AVG is easily measured and can generate more consistent and reliable effects than gaseous applications of 1-MCP.

Post-harvest treatments are usually carried out to improve the quality and vase life of fresh-cut flowers. However, plant vigor, growth and pre-harvest quality can significantly impact post-harvest quality. Thus, pre-harvest treatments can provide a combinational effect with post-harvest treatments, which may increase fresh-cut snapdragons quality, longevity and marketability.

MATERIALS AND METHODS

Plant Materials and Treatments

Due to the difficulty of obtaining fresh cut long stem snapdragon in Florida, we used "Snapshot White" instead for the experiment. The results from this study could be translated to different varieties within the same species. Snapdragon (*Antirrhinum majus* L.) seedlings in 6" pot of the cultivar "Snapshot White" were kindly provided by KNOX Nursery Inc. (Winter Garden, FL). After budding, plants with consistent growth were selected as experimental materials. All potted plants were arranged in trays randomly. Six pots were used for each treatment, and three replicates were performed. The experiment was divided into two groups. One group was subjected to foliar application of either melatonin or AVG at different concentration solutions (100, 150 and 200 $\mu mol \cdot L^{-1}$) on both abaxial and adaxial sides of the leaves while water was sprayed as the control. After the plants were sprayed, they were grown under fluorescent (white) light for 16 h light and 8 h dark photoperiod at constant room temperature. The other group was exposed to different LEDs (Heliospectra, Sweden) regimes (10h White with 6h Red at 660nm spectral output (WR), 10h White with 6h Blue at 460nm spectral output (WB), and 10h White with 3h Red and 3h Blue (WRB)) at constant room temperature. The potting mix was maintained at a moist condition during the whole experiment. For both groups of treatments, flower inflorescences or spikes were harvested in the morning and immediately placed in distilled water when the first 1-2 florets were fully opened. To avoid blocking of water transport by air at the cut section, flower spikes were re-cut under the water at a 5cm angle and then individual spike was transferred to a vase containing 250mL of distilled water. All the vases were sealed with aluminum foil to prevent water evaporation, and kept in a conditioned room at 23°C, 60% relative humidity, and 12 h light per day. Three replications were performed with four spikes for each treatment. The complete list of treatments can be seen in Table 1.

Morphological Measurements and Determination of Vase Life: Plant Growth, Inflorescence (Spike) Length, Number of Florets Per Spike, and Transverse/Vertical Diameter

Plant growth was calculated by measuring plant height from the base to the tip of the plant using a measuring tape before treatment and before harvesting. Inflorescence (spike) length was measured from the inflorescence base (at the first floret node) to the top of the plant. The number of florets was counted after harvesting and the flower diameter was defined as the maximum width of each flower and was measured every day with a Vernier caliper while the flowers were in vases. The vase life was determined as the number of days from harvest to the day when flowers in vases lost their decorative value which defined as pedicel bending or petal browning and fading [26].

Statistical Analysis

All data were statistically analyzed by one-way analysis of variance (ANOVA) using SPSS software version 17.0. Graphs

Table 1: Treaments category at the beginning of flower emergency

Treatment	Light Quality	Chemical
Control (W)	16 h White	Water
WR	10 h White + 6 h Red	--
WB	10 h White + 6 h Blue	--
WRB	10 h White + 3 h Red + 3 Blue	--
M100	16 h White	100 $\mu mol \cdot L^{-1}$ Melatonin
M150	16 h White	150 $\mu mol \cdot L^{-1}$ Melatonin
M200	16 h White	200 $\mu mol \cdot L^{-1}$ Melatonin
A100	16 h White	100 $\mu mol \cdot L^{-1}$ AVG
A150	16 h White	150 $\mu mol \cdot L^{-1}$ AVG
A200	16 h White	200 $\mu mol \cdot L^{-1}$ AVG

were generated using Microsoft Excel using means and standard errors for each category. The means were separated by Duncan test at 0.05 probability level.

RESULTS

Stem Growth and Spike Length

Plant height is an important indicator of cut flower quality, and a minimum length of stems is required for each type of flower [27]. Both LEDs and melatonin treatments significantly affected the growth of snapdragon (Figure 1A). While there was no significant difference between WRB or WR treatments and the control, WB strongly inhibited stem elongation with a 27.0% decrease in growth. Application of exogenous melatonin resulted in significant stem growth, with an increase of 17.68%, 21.19%, and 22.03% under the treatment with 100,150, and 200 μmol·L^{-1} melatonin, respectively. In contrast, plant growth only exhibited a significant increase with 100 μmol·L^{-1} of AVG application while 150 and 200 μmol·L^{-1} AVG treatments showed no differences from the control. Comparable to the effect on stem growth, spike length increased with all melatonin treatments while it only increased with 100 μmol·L^{-1} of AVG treatment (Figure 1B). Interestingly, WR treatment significantly increased spike length but not plant growth. The longest spike was observed under WRB treatment where the mean value reached 11.3cm and the shortest mean inflorescence was 7.5cm under WB treatment.

Vase Life

Similar to the morphological characteristics, snapdragon vase life was extended with an increase in melatonin concentration. Melatonin at 200 μmol·L^{-1} delayed spike wilting and increased vase life with a mean value of 12.8 days, which is significantly different from the white light control with an average vase life of 10.5 days (Figure 1D). Although WRB and WR treatments showed an obvious positive change in vase life, WB significantly reduced snapdragons vase life compared to both the control and other treatments, resulting in the vase life with an average of only 9 days. To our surprise, the preharvest treatment with the ethylene production inhibitor AVG did not improve vase life of fresh-cut flowers and the vase life of cut flowers pre-treated with all concentrations of AVG decreased (Figure 1D).

The Number of Florets Per Spike and the Size of Florets

The ornamental value of cut flower depends on the number of florets per spike [28]. Additionally, more florets may indicate a longer vase life. The highest numbers of florets were observed after 200 μmol·L^{-1} of melatonin treatment, reaching a mean value of 14.8 florets per spike, an increase of 16.08% from the white light control (0) (Figure 1C). The WRB treatment also significantly increased floret number per spike, with a mean value of 14.75 or a 15.69% increase from the white light control. The fewest florets were obtained from WB treatment where the mean value was only 10.50 or 17.65% reduction compared to the white light control. No significant difference was observed between the rest of the treatments and the control (Figure 1C). Likewise, both transverse and vertical diameter floret sizes were increased under WRB treatment and the transverse diameter was wider only with 200 μmol·L^{-1} application of melatonin. The WB treatment did not show an obvious effect on the size of florets although this treatment exhibited negative effects on all other characteristics (Figure 1, 2, and 3). On the other hand, all AVG treatments inhibited the development of floret sizes both in the transverse and vertical diameter (Figure 2). Noticeably, the size of florets decreased significantly with the increase of AVG concentration where the smallest transverse diameter (2.41cm) and vertical diameter (3.51cm) were observed after treatment with 200 and 150 μmol·L^{-1} AVG, resulting in a reduction of 6.30% and 9.90% from the control, respectively (Figure 2A, 2B, and 3).

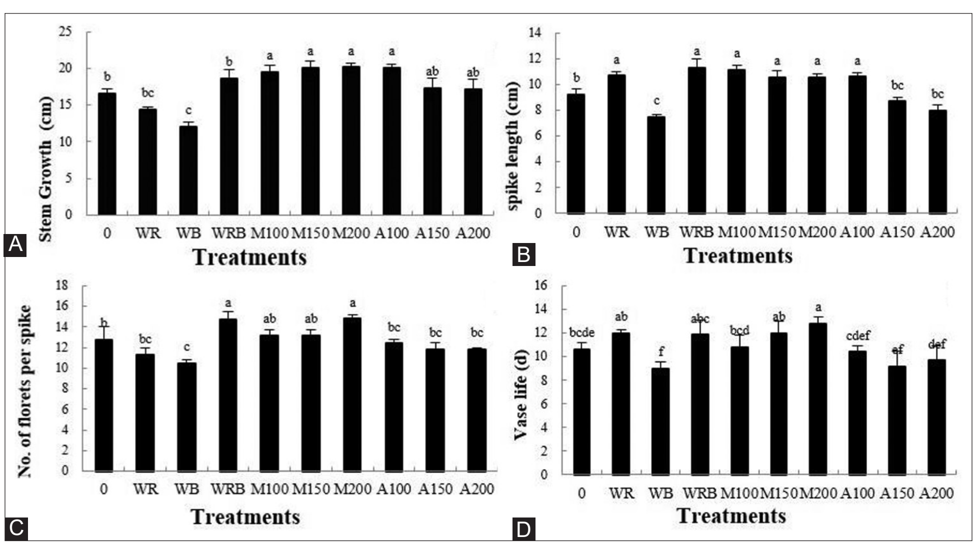

Figure 1: Morphological measurement and vase life of under different treaments. A) Stem growth, B) Spike length, C) Number of florets per spike, D) Vase life. Different letters on bars indicate significant difference at $p < 0.05$

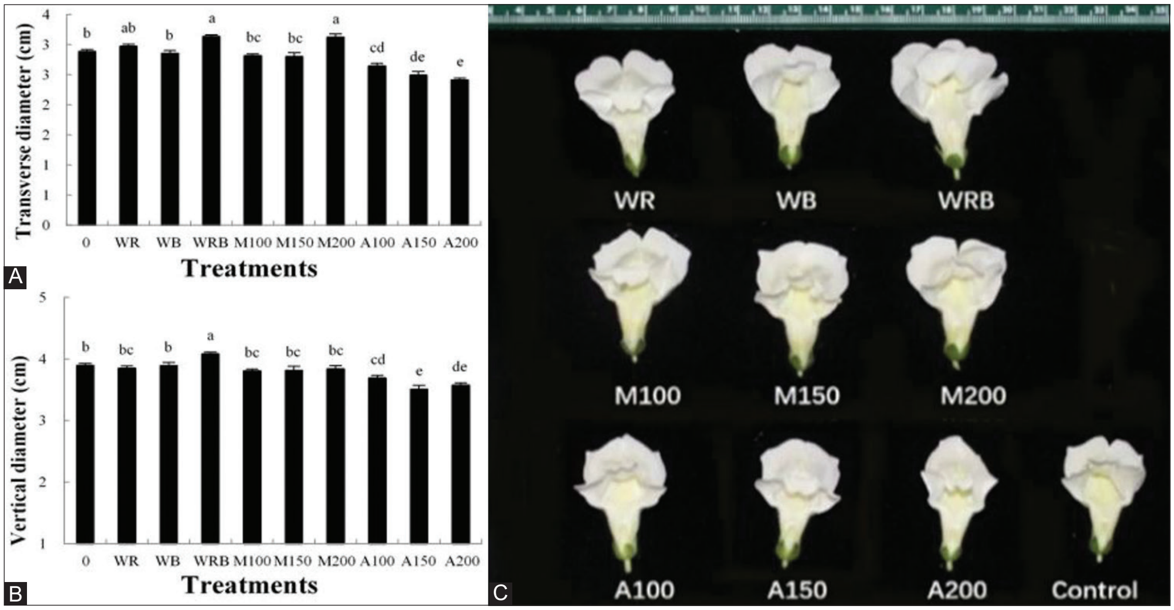

Figure 2: The size of florets under different treaments. A) transverse diameter; B) vertical diameter; C) Flower size representation images. Different letters on bars indicate significant difference at $p < 0.05$

Figure 3: Representive Image of spikes for each treament. A) Control (White light); B) From left to right; WRB, WR and WB C) Melatioin at 100, 150 and 200 μmol L^{-1}; D) AVG at 100, 150 and 200 μmol L^{-1}

DISCUSSIONS

LEDs Effects on Snapdragons

Previous studies examining the effects of LEDs on ornamental plants generally started treatments at the seedlings stage or after harvesting cut flowers. While treating the plants with different wavelengths of LEDs during the seedling stage can significantly impact their morphological characteristics such as stem length, the quality of the seedlings of many ornamental plants was similar under LEDs and high-pressure sodium lamps [29]. Additionally, continuous LEDs treatments could negatively affect flowering of treated seedlings [30]. In contrast, the effects of postharvest LEDs treatments were limited to the deterioration of the fresh-cut flowers [31]. The present study examined how preharvest treatments of snapdragons with different wavelengths of LEDs affect quality of postharvest plants. Our results showed that pre-harvest treatments with specific wavelength(s) of LEDs have significant but distinct impact on postharvest quality of snapdragon. For example, WRB and WR increased spike length, while it was shortened under WB treatment (Figure 1B and Figure 3). Similarly, the number of florets and the size of the flowers were negatively impacted by WB treatment and increased most notably under WRB treatment. A slight increase in vase life was also observed for WR and WRB treatments. Although blue light irradiation can increase shelf life of many leafy vegetables after post-harvesting, white light supplemented with blue light negatively affects snapdragon flower development, resulting in fewer florets per spike (Figure 2) [32]. Additionally, white light supplemented with both blue and red light was able to improve flower quality of snapdragons more than white light supplemented with red light alone. These results indicate that different light wavelengths combinations may generate distinct effects on plant quality. Therefore, LEDs effect on plant quality may be species-dependent and considerations should be taken when utilizing LEDs treatments.

In addition to growth characteristics and flower development, LEDs have been shown to affect anthocyanin content in a variety of plant species. Wojciechowska et al. indicated that blue light increased the anthocyanin content in *Lachenalia's* "Rupert" corolla by 35% when compared with red light treatment as well as the control [33]. An et al. also showed that irradiation with both blue and red light increased anthocyanin production more than monochrome blue and monochrome red light in *Prunus x yedoensis's* "Somei-yoshino" cherry blossom cultivar [34]. In this study, we have not observed any alteration in pigment of snapdragon flower and vegetative organs, suggesting that anthocyanin production in snapdragon may not be as responsive to LED blue or red light. However, we cannot rule out the possibility that genotypic difference, treatment duration with LEDs, and the intensity of LEDs may account for this absence of physiological change.

Melatonin and AVG Effects on Snapdragons

Melatonin can alleviate adverse effects of abiotic stresses by acting as an antioxidant and radical scavenger. The beneficial effects of melatonin recently expanded to seed germination and post harvesting [35]. Melatonin treatment ameliorated postharvest decay in strawberry, promoted ripening and improved tomato quality, and alleviated browning of fresh-cut pear [36,37,38]. However, little is known of melatonin's effects on flower development and postharvesting quality of fresh-cut-flowers. Here, we reported the first use of melatonin to improve flower quality of snapdragon. All three concentrations at 100, 150, and 200 $\mu mol \cdot L^{-1}$ positively increased snapdragon growth and spike length. While 100 and 150 $\mu mol \cdot L^{-1}$ of melatonin slightly increased the number and size of florets, 200 $\mu mol \cdot L^{-1}$ of melatonin significantly increased the quality of the flowers when compared to the control. Most notably, only the 200 $\mu mol \cdot L^{-1}$ melatonin treatment has a significant vase life extension out of all the treatments in comparison to the control. Although the molecular mechanisms are unclear, studies have suggested that melatonin ameliorates post-harvesting decay through the reduction of ethylene production by inhibiting the expression of genes related to ethylene biosynthesis [39]. In addition, exogenous melatonin can inhibit bacterial, viral, and parasitic infections by increasing the expression of pathogenesis-related genes such as nitric oxide and salicylic acid to improve post-harvest quality in fruits and vegetables [40]. In 2016, Ichimura et al. introduced mannitol as a source of carbohydrate to stimulate bud development and extend vase life of snapdragon flowers [41]; future studies may be performed to examine how the combined application of melatonin and mannitol affects snapdragon flower quality and vase life.

To our surprise, preharvest application of AVG only increased plant growth and spike length at the lowest concentration at 100 $\mu mol \cdot L^{-1}$, and high concentration of AVG negatively affect flower size and vase life. These results indicated that the application of high concentrations of AVG may significantly inhibit endogenous ethylene production. Ethylene is a type of stress hormone that plays an essential role in plant development in many plant species, which might explain the deleterious effect of high concentrations of AVG on plant development such as flower size. Another possibility is that snapdragon is highly responsive to endogenous ethylene changes; inhibition of endogenous ethylene production may cause developmental defects even at very late developmental stages. Previous reports have shown that the application timing of AVG is critical for its effect. Hayama et al. found that application of 1-MCP at early developmental stages generated a better effect on delaying peach fruit softening, while AVG should be applied at the latter developmental stages to have a better effect [42]. Therefore, application timing of the two chemicals is critical for reducing peach fruit softening. Although the application of AVG alone did not generate significant beneficial effects on snapdragon vase life, combinations of AVG and other plant hormones may have different effects. Shimizu-Yumoto and Ichimura observed that fresh-cut *Eustoma* flowers pulse-treated with AVG in combination with 1-naphthaleneacetic acid (NAA) significantly extend vase life up to three times more than the control [43]. Therefore, AVG might have a significant impact on snapdragons vase life if used properly. A complementary study using either LEDs or melatonin as treatment during snapdragons flower development stage; and AVG as a pulse treatment after flowers are cut may provide a practical solution to increasing postharvesting quality of snapdragons fresh-cut flowers.

CONCLUSIONS

In summary, both WRB or melatonin treatments positively affect flower quality during pre-harvest and post-harvest stages; and 200 $\mu mol \cdot L^{-1}$ of melatonin alone extended the vase life of cut snapdragons. While AVG treatments negatively affect snapdragon flower development in this study, the effects of AVG at different concentrations and developmental time may have a significant impact on post-harvesting quality indicated by other studies. In conclusion, post-harvest information obtained from pre-harvest treatment with LEDs and melatonin have provided new insights to enhancing snapdragon flower quality and may provide practical solutions to increase their marketability.

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