

Floriculture

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Section Editor

Screening Photosynthetic Performance of Bedding Plants Using Chlorophyll Fluorescence

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Index Words Supplemental lighting, photosystem II, electron transport rate

Significance to Industry Supplemental lighting can be used to improve plant quality and speed production cycles for floricultural crops. Although supplemental lighting generally increases production costs due to its high energy requirements, newly-developed lighting strategies have the potential to greatly reduce this expense. However, understanding crop-specific light use dynamics is necessary in order to maximize the efficiency of supplemental lighting systems. In this study, we used diurnal chlorophyll fluorescence monitoring to evaluate the light use efficiency of three bedding plant species. Geraniums, petunias, and impatiens were continuously monitored over 24-hour periods using a chlorophyll fluorometer and quantum sensor to determine the efficiency of photosynthetic light use (Φ_{PSII}), the intensity of photosynthetically-active light (photosynthetic photon flux; *PPF*), and the electron transport rate (*ETR*), a proxy for photosynthesis. For all species, *ETR* increased as *PPF* increased up to a maximum *ETR*, with little or no increase at higher light intensities. This was due to a decrease in Φ_{PSII} at higher *PPFs*. Impatiens had lower *ETRs* and Φ_{PSII} than geraniums and petunias at *PPFs* greater than $200 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$. Similarly, geraniums and petunias reached *ETRs* of $100 \pm 10 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ at much lower *PPFs* than impatiens. Diurnal chlorophyll fluorescence monitoring is an effective means of determining crop-specific light requirements.

Nature of Work The use of supplemental lighting for floricultural crops can speed production cycles and improve plant quality. Gianino and van Iersel (1) recently developed a supplemental lighting control system that provides additional light only up to a pre-defined *PPF* threshold, automatically dimming when ambient light intensity increases, and brightening when it falls below the desired level. Preliminary results from greenhouse-grown begonias have shown that this approach to supplemental lighting control can improve plant growth and quality with minimal energy expense (our unpublished results). Plant responses to light intensity vary among species, and are affected by growing conditions (2, 3). Therefore, to optimize supplemental lighting control systems it is necessary to understand the photosynthetic performance of individual species within a specific production environment (4). Chlorophyll fluorescence measurements are a rapid and non-destructive means of determining photosynthetic light use efficiency (5). The aim of this study was to evaluate diurnal chlorophyll fluorescence monitoring as a method for screening the light use efficiency of different floricultural crop species under realistic growing conditions.

Photosynthetic performance of three commercially-important bedding plant species was evaluated in a glass-covered greenhouse in Athens, GA, during January and February, 2016.

Geranium (*Pelargonium x hortorum* 'Maverick Violet'), petunia (*Petunia x hybrida* 'Daddy Blue') and impatiens (*Impatiens walleriana* 'Super Elfin XP Violet') seeds were germinated in plug trays and transplanted into 6 inch (15 cm) round pots filled with a peat:perlite substrate (Fafard 1P; Sun Gro Horticulture, Agawam, MA) two weeks after germination. Plants were watered daily and fertilized weekly with a 200 ppm N liquid fertilizer (15-5-15 Cal-Mag; Everris, Marysville, OH). Continuous chlorophyll fluorescence measurements were taken 6 to 8 weeks after transplanting using a chlorophyll fluorometer and attached leaf clip with quantum sensor (JUNIOR-PAM, Heinz Walz; Effeltrich, Germany). Leaves were measured once every ten minutes during the day and once an hour at night over 24-hour periods; this was repeated five times for each species. Chlorophyll fluorescence measurements were used to determine the quantum yield of photosystem II (Φ_{PSII}), a measure of the efficiency with which absorbed light is used by plants to drive photosynthesis (6). Electron transport rate (*ETR*) was calculated from Φ_{PSII} and *PPF* using the following equation: $ETR = \Phi_{PSII} \times PPF \times 0.84 \times 0.5$.

Results and Discussion Generally, Φ_{PSII} decreased and *ETR* increased up to a maximum *ETR* as *PPF* increased, as demonstrated over the course of a single day of measurements with geranium and impatiens (Fig. 1). This asymptotic response of *ETR* to increasing light intensity occurs because plants use light less efficiently (Φ_{PSII} decreases) as they receive more light. Since excess light energy can cause damage to the photosynthetic apparatus, some of the light absorbed by a leaf is dissipated as heat and not utilized for the light reactions of photosynthesis. As light intensity increases, a larger proportion of the absorbed energy is lost as heat, and photosynthesis becomes less efficient (lower Φ_{PSII}) (7, 8). Thus, providing supplemental light at supra-optimal intensities will have little or no positive effect on photosynthesis and growth, but could instead damage the photosynthetic apparatus and decrease photosynthetic efficiency.

Compared to geraniums, impatiens consistently had a lower *ETR* and Φ_{PSII} at light intensities above $200 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$, and similar *ETR* and Φ_{PSII} at light intensities below $200 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ (Figure 1). This indicates that, although impatiens are generally considered to be shade plants, they do not use low-intensity light more efficiently than sun-loving geraniums. Rather, these results suggest that impatiens have an inherent inability to utilize higher light intensities. This is likely related to limitations in the photosynthetic carbon reactions because the signal which initiates heat-dissipative processes is generated when the light reactions proceed at a faster rate than carbon fixation (9).

During the course of this study, maximum daily *PPFs* were 130 to $1,360 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ (mean $802 \pm 439 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$). This is generally lower than would be observed under full sunlight conditions during the growing season but well within the range of light intensities that could be provided using supplemental lights. Table 1 illustrates the mean *PPF* and Φ_{PSII} at which each species reached an *ETR* of 50, 100, and 150 (± 10) $\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$. All three species reached *ETRs* within the range of $50 \pm 10 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ at a similar *PPF* (200

to $220 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$) and Φ_{PSII} (0.5 to 0.59). Impatiens had *ETRs* of $\sim 100 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ at a *PPF* of $1039\pm 153 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ and Φ_{PSII} of 0.23 ± 0.03 , while geraniums and petunias reached such *ETRs* at much lower light intensities with higher values of Φ_{PSII} (Table 1). Since *ETR* correlates to the overall rate of photosynthetic carbon fixation and is likely correlated to growth rate (5), supplemental light intensities should be controlled within ranges that allow plants to use light most efficiently (high Φ_{PSII}). Thus, optimal supplemental light levels for impatiens are lower than those for petunia and geranium. The results of this study demonstrate that diurnal chlorophyll fluorescence monitoring can be used to understand light use efficiency of different crop species under actual growing conditions, and could be a valuable tool for developing supplemental lighting control regimes.

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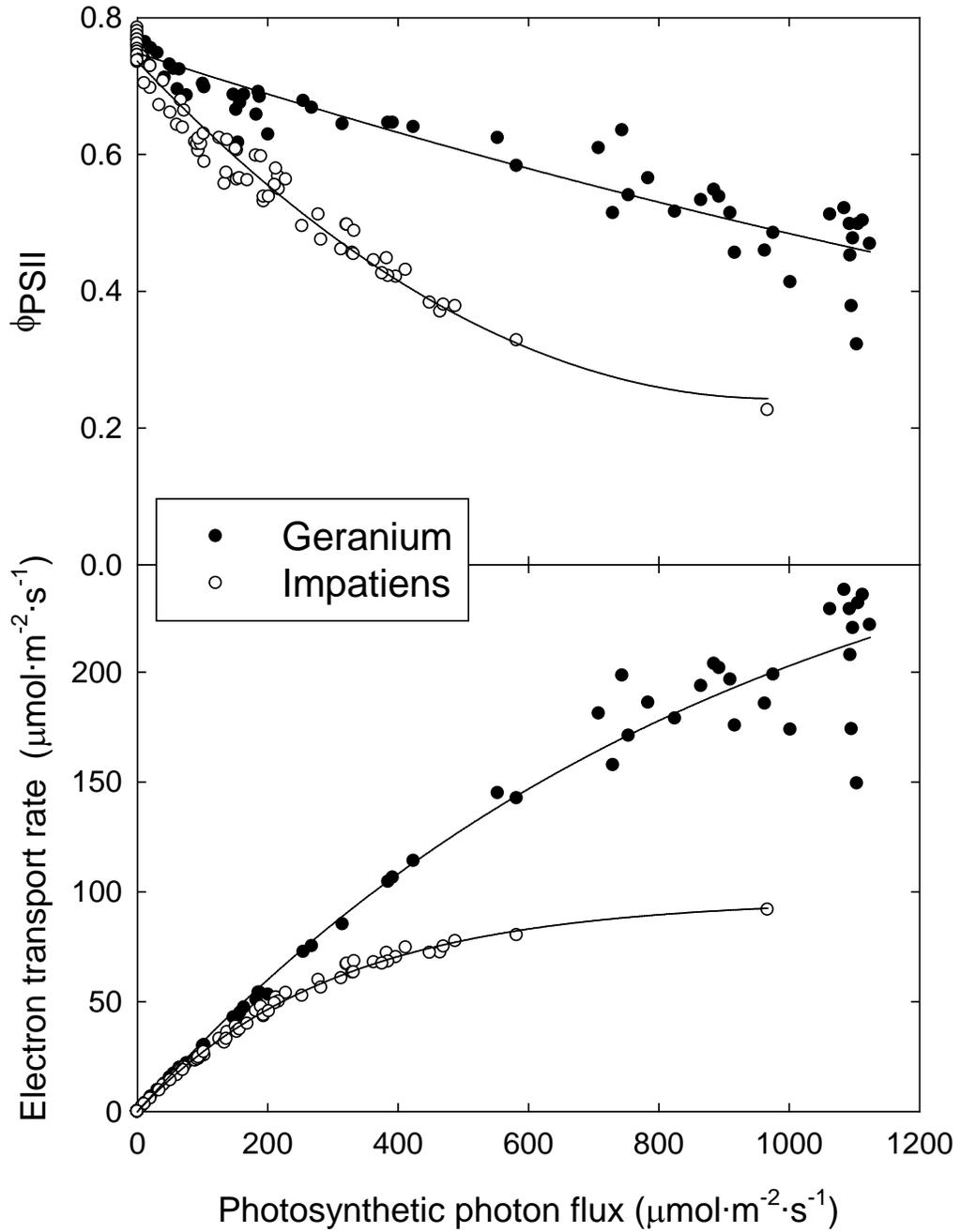


Figure 1. Photosynthetic light use efficiency (Φ_{PSII}) and electron transport rates of geranium and impatiens at a range of light intensities as measured over a single day.

Table 1. Photosynthetic photon flux (a measure of light intensity) needed to achieve three specific electron transport rates, and Φ_{PSII} values at which these electron transport rates were achieved for petunias, geraniums, and impatiens. Values are mean \pm standard deviation.

Species	Electron transport rate ($\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$)	Photosynthetic photon flux ($\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$)	Φ_{PSII}
Petunia	49.7 \pm 5.1	201.1 \pm 29.6	0.59 \pm 0.04
	97.1 \pm 5.5	462.3 \pm 91.6	0.51 \pm 0.08
	151.9 \pm 6.5	877.8 \pm 56.6	0.41 \pm 0.03
Geranium	47.7 \pm 5.8	201.1 \pm 39.0	0.58 \pm 0.08
	96.0 \pm 6.9	482.4 \pm 112.4	0.50 \pm 0.11
	146.8 \pm 4.7	964.0 \pm 217.1	0.38 \pm 0.10
Impatiens	48.7 \pm 5.8	217.6 \pm 47.8	0.54 \pm 0.06
	99.8 \pm 5.8	1038.9 \pm 153.2	0.23 \pm 0.03
	159.2 \pm 3.7	1161 \pm 4.9	0.33 \pm 0.01

Adaptive LED Lighting Can Benefit Greenhouse Production

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Index Words Light emitting diode, duty cycle, adaptive lighting, energy efficiency, photosynthetic photon flux

Significance to Industry Light emitting diodes (LEDs) are increasing in popularity, thanks to continuous research on improving their efficiency and performance. LEDs have been reported to reduce electricity use for supplemental lighting by 35-70% compared to high pressure sodium lamps [1, 2, 3]. Although LEDs are not yet widely used by greenhouses, they can provide important benefits over more commonly used high-pressure sodium (HPS) lights. When HPS lights are used, they typically are used at full power. In contrast, LEDs can be dimmed which can lead to potential energy savings. However, the dimming capability of LEDs rarely is taken advantage of in supplemental lighting applications. We have developed an adaptive LED lighting system that prevents the light level at the crops from dropping below grower-determined thresholds. The LEDs dim or brighten automatically in response to changing levels of sun light in a matter of seconds and provide only enough light to reach the threshold. Greenhouses can benefit from this system by assuring that the crop always receives appropriate levels of light, while using the least amount of energy possible. This adaptive LED lighting system has the potential to reduce electricity costs, offsetting the high capital costs of LED lighting arrays.

Nature of Work Light-emitting diodes (LEDs) are gradually being adopted by large greenhouses and are popular in many research institutions. LED grow lights for supplemental lighting use wavelength-specific diodes that emit light in the photosynthetically-active range of the light spectrum [1, 5, 6]. LED grow lights have been reported to improve growth and reduce energy consumption [1, 4, 5]. Seedlings of Douglas fir (*Pseudotsuga menziesii*) and Engelmann spruce (*Picea engelmannii*) have greater shoot height, root length, stem diameter, and more chlorophyll when grown under LEDs compared to HPS lamps [1]. LED supplemental lighting may reduce energy use by 35% to 70%, when compared to HPS lighting [1, 5]. For example, tomato fruit yield was similar when grown with supplemental, intracanopy LED lighting compared to overhead HPS supplemental lighting, but the LEDs used only 24% of the energy that HPS lamps used for supplemental lighting [5]. The main reason that growers are reluctant to purchase LED lights is the high initial cost [6]. LEDs are still more expensive than HPS and metal halide lamps, so most growers opt for the less expensive lighting system. To make LED lights more economical, we want to take advantage of some unique capabilities of LEDs. For example, the light output from LED lights can be precisely, easily, and cheaply controlled by adjusting their duty cycle, which controls the amount of power used by the lights. By decreasing the duty cycle, the LEDs are dimmed, reducing light output and energy consumption.

Our goal was to design a light sensor-based LED control system that can adjust the duty cycle of LED lights in response to changing amounts of sunlight. We wanted to assure that the adaptive LED light can prevent light levels, or photosynthetic photon flux density (PPFD), from dropping below grower-specified thresholds by providing just enough supplemental light to reach that threshold. By not providing more energy than needed, such a light can realize substantial energy savings.

One LED light bar (54 W; PopularGrow, Shenzhen, China) with a 5:1 ratio of red to blue LEDs was connected to a duty cycle controller, which was connected to a datalogger (CR1000, Campbell Scientific, Logan, UT) (Fig. 1). One quantum sensor (Li-190, LI-COR, Lincoln, NE) was placed underneath the center of the LED light bar to measure the PPFD from the LEDs and sun light combined. A second quantum sensor was placed above the LED bar to measure the PPFD from sunlight. The datalogger took readings from the quantum sensor under the LED bar and if the PPFD reading was below the threshold level, then the duty cycle was increased to prevent the light level from dropping below the threshold and vice versa. PPFD thresholds were set daily to 50, 100, 150, 200, or 250 $\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ to test the ability of the adaptive LED light to prevent PPFD levels from dropping below those thresholds under a range of different sun light conditions. Each threshold was maintained for 14 hours on four separate days.

Results and Discussion For each threshold, and on each day, the adaptive LED system was able to prevent light levels from dropping below the assigned threshold. For example, the adaptive LED system was able to prevent the light levels under the LED bar from dropping below a PPFD level of 200 $\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ in a 14-hr period (Fig. 2). As the duty cycle was adjusted, the amount of electricity provided to the LED light bar was adjusted, in order to prevent the PPFD from dropping below the threshold. When the measured sunlight increased, the duty cycle automatically decreased (Fig. 3). Only when sunlight exceeded the threshold did the adaptive system turn off the LED light bar.

The system operated on cloudy and sunny days with daily light integrals (DLI) ranging from 2.8 to 9.5 $\text{mol}\cdot\text{m}^{-2}\cdot\text{d}^{-1}$. PPFD levels under the LED bar were maintained at or above the thresholds, regardless of the threshold or weather conditions. This shows that the system can change the duty cycle to adapt and adjust within seconds to accurately achieve desired PPFD levels. With a threshold of 200 $\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$, the adaptive LED light used 40% (at a DLI of 2.8 $\text{mol}\cdot\text{m}^{-2}\cdot\text{d}^{-1}$) to 60% less energy (DLI of 9.3 $\text{mol}\cdot\text{m}^{-2}\cdot\text{d}^{-1}$), compared to using the LED light at full power. Overall, energy savings from the adaptive light ranged between 20 and 92%, with greater savings on days with higher DLI and lower PPFD thresholds. The energy savings that can be achieved with this adaptive LED system can offset the initial investment of LED supplemental light systems. Since this is an automated system, it does not require constant monitoring, thus requiring little grower time.

A preliminary study showed growing *Begonia semperflorens* with this system reduced energy consumption by 60% with only a 10% reduction in biomass, compared to LEDs that were on at full power. This adaptive LED light will be commercialized by PhytoSynthetix, an LED lighting company in Athens, GA.

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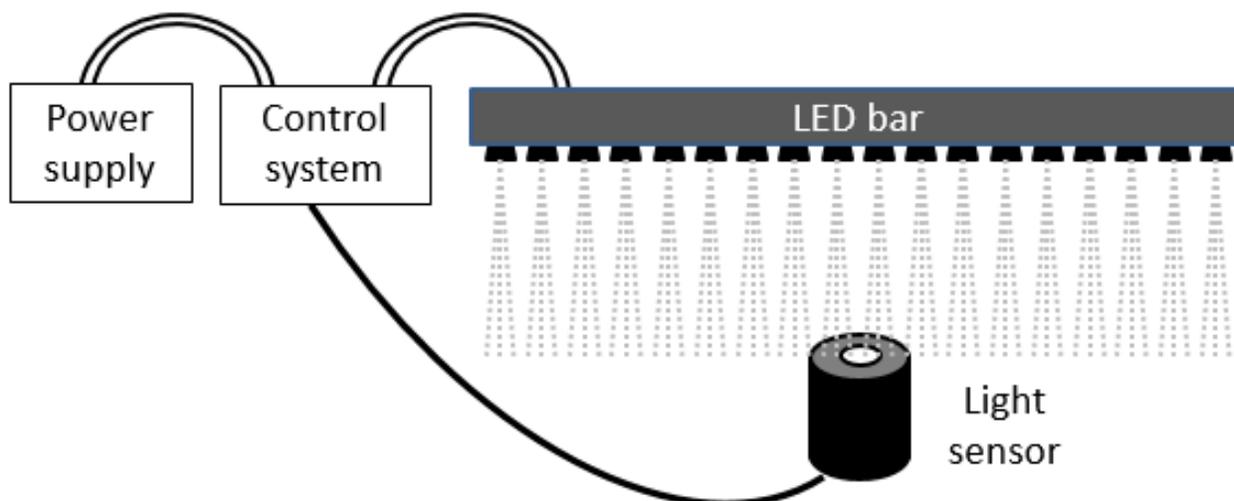


Figure 1. A diagram of the adaptive LED light. The controller consists of a datalogger and duty cycle control board. It uses the light sensor to determine how much light is present underneath the LED bar. If there is less light than needed, the controller automatically increases the duty cycle of the light bar, and vice versa.

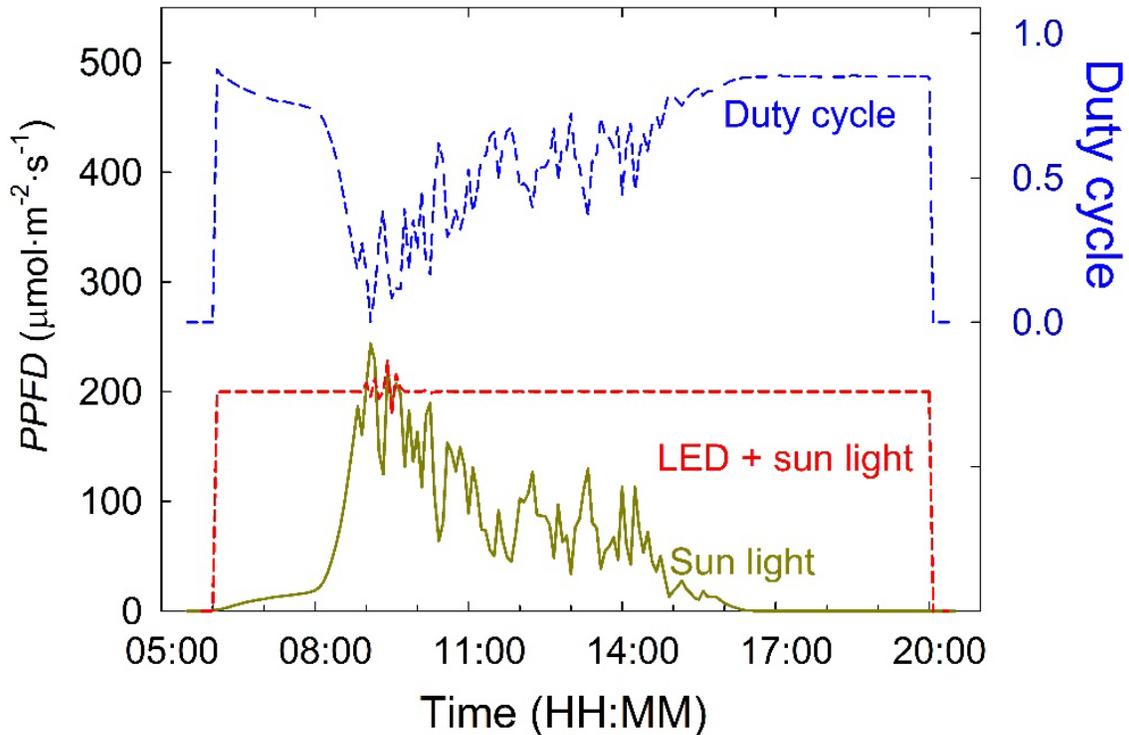


Figure 2. The amount of sunlight, the amount of sunlight plus the light from the LED underneath the LED light bar, and duty cycle of the LED light during a cloudy, winter day. The adaptive LED system was set to maintain a target photosynthetic photon flux density (PPFD) of $200 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$, over the course of a 14-hr period. Note that the duty cycle of the LED light decreases as sunlight increases. The adaptive light maintained the PPFD very close to the set point of $200 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ throughout the 14-hr period.

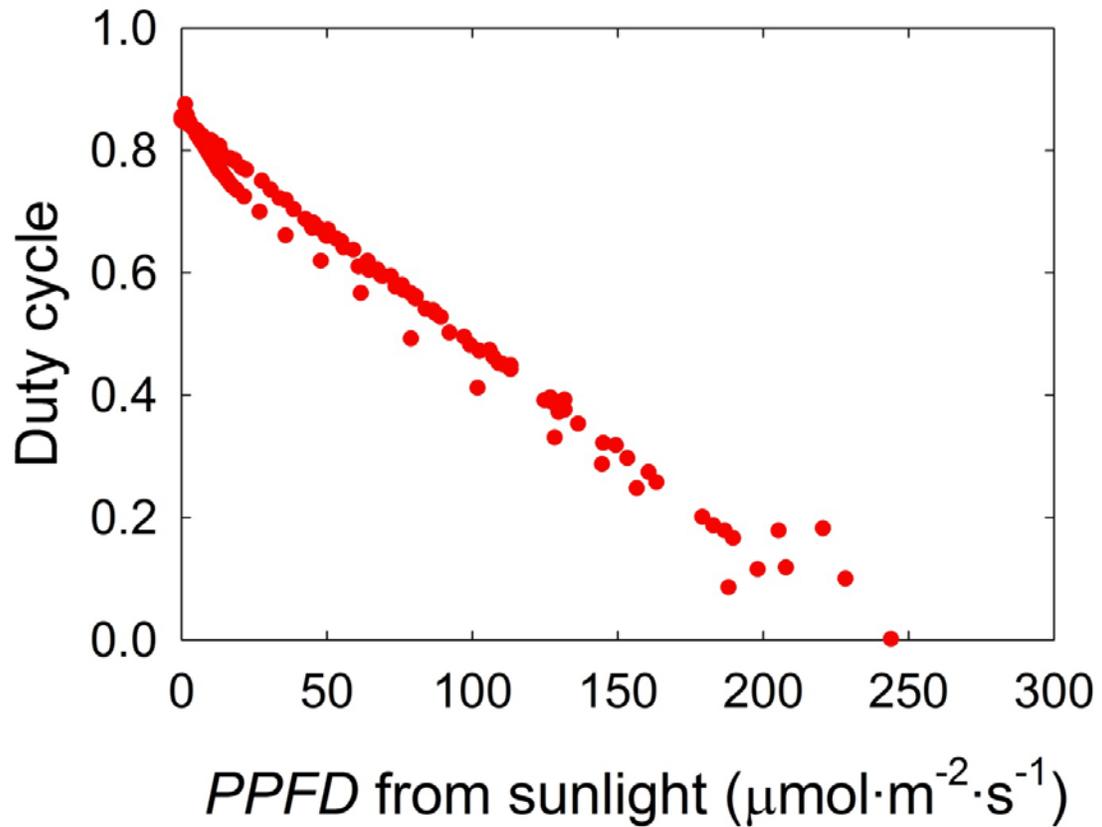


Figure 3. The relationship between the amount of sunlight (photosynthetic photon flux density, PPFD) and the required duty cycle of the LED light bar to maintain a PPFD of at least $200 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$.

Inoculating Coated Marigold Seed with Fungal Entomopathogens: Effects on Plant Growth

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Index Words Endophytes, seed coatings, *Beauveria bassiana*, *Isaria fumosorosea*, marigold

Significance to Industry Inoculation of field crops with endophytic fungal entomopathogens can improve crop growth, resistance to pests and pathogens, and tolerance of environmental stress (1, 2). It is unknown whether these growth effects apply to the production of marigolds (*Tagetes erecta*), one of the top 15 flowering annuals sold in the U.S. Seed coating technology could provide an economical system for delivering endophytes to *T. erecta* (3). While we found mixed results from the impacts of seed coating and entomopathogenic endophytes on a number of *T. erecta* horticultural characteristics, these results should also be evaluated within the context of possible pest control effects, which are currently under study.

Nature of Work Fungal endophytes are symbiotic fungi that inhabit plant tissues without causing harm to their plant hosts. *Beauveria bassiana* and *Isaria fumosorosea* are commercially available fungal insect pathogens and seed inoculation with these fungi may reduce feeding damage and reproduction of greenhouse pests infesting a number of flower crops. We investigated the effect of these endophytes on Discovery Yellow marigold (Ernst Benary of America, Inc.) growth and form in the presence of a commercial antimicrobial seed coating (DISCO VEG Yellow L-072). We used a full factorial design to test two seed treatments (non-coated and coated) with 3 endophyte treatments (*B. bassiana*, *I. fumosorosea*, and sterile control). Fungi were cultured on sabouraud dextrose agar and incubated for 5 weeks at 25°C. Fungi were suspended in 0.1% TritonX-100 and the suspensions were diluted in 2% methyl cellulose to a concentration of 1×10^8 conidia/mL according to (4). *Tagetes erecta* seeds were submerged in treatment suspensions containing *B. bassiana*, *I. fumosorosea*, or a 2% methyl cellulose sterile control for 1 h and then air-dried. Each *T. erecta* seed was planted in a 3.5-inch pot with a peat-perlite substrate (Sunshine Mix #1) on March 10, 2016. Pots were maintained in a fan-pad cooled glasshouse for 3 weeks, and irrigated every 2-3 days with reverse-osmosis treated water without fertilizer. Seeded pots were grouped in seed lots containing 10 pots for each of the six treatments, with 12 seed lots in total per treatment. Seed lots were distributed in a complete randomized block design. The germination frequency of each seed lot was recorded every 12 hours during the first week after sowing. After 3 weeks, a random sample of 30 seedlings from each treatment group was collected. The height to width ratio (height:width) of each plant was recorded, then the plants were rinsed in water

to remove soil from the roots and the fresh mass was measured. A probit regression model was used to predict the median germination time (MGT), or time at which 50% germination was reached, for each seed lot. A two-way ANOVA was performed separately for each dependent variable (MGT, final germination proportion one week after sowing, plant fresh weight, and plant height:width) with the seed type and endophyte type as the categories with 2 and 3 levels in each category, and the interaction between seed type and endophyte. The germination proportion and height:width were arcsine-transformed before ANOVA was applied. If the F-statistic from the ANOVA was significant, Tukey HSD tests were used to identify significance between treatment responses.

Results and Discussion Seed coating had a significant impact on MGT ($F_{2,72}, p < 0.0001$) (Table 1), with a higher MGT in non-coated seeds. Endophytes also affected MGT ($F_{2,72}, p = 0.0323$), which was higher in *I. fumosorosea*-treated seeds than *B. bassiana*-treated seeds. Neither *I. fumosorosea* ($F_{2,72}, p = 0.8426$) nor *B. bassiana* ($F_{2,72}, p = 0.1202$) differed from the sterile control. There was no interaction between seed and endophyte treatments ($F_{2,72}, p = 0.4076$). Seed coating increased germination proportion ($F_{2,72}, p < 0.0001$) (Table 1). Endophytes had an impact on the proportion ($F_{2,72}, p = 0.0245$); *I. fumosorosea* treatments were significantly lower than the controls ($F_{2,72}, p = 0.0245$), and *B. bassiana* did not differ from the control ($F_{2,72}, p = 0.9739$) or *I. fumosorosea* ($F_{2,72}, p = 0.0615$). Seed coating and endophyte effects on germination proportion did not interact ($F_{2,72}, p = 0.4292$). Seed coating increased seedling fresh mass ($F_{2,180}, p < 0.0001$), and endophytes were significant ($F_{2,180}, p < 0.0001$) (Fig. 1). *Beauveria bassiana* and *I. fumosorosea* treatments decreased fresh weight compared to control groups ($F_{2,180}, p < 0.0001$; $F_{2,180}, p = 0.0366$), with *B. bassiana* treatments significantly lower than *I. fumosorosea* treatments ($F_{2,180}, p = 0.0052$). Coated and non-coated treatments elicited different endophyte effects on plant fresh weight ($F_{2,72}, p = 0.0003$). In non-coated seeds, *Isaria fumosorosea* treatments had significantly lower fresh weight than the control ($F_{2,180}, p < 0.0160$); and there were no differences between *B. bassiana* and *I. fumosorosea* ($F_{2,180}, p < 0.0001$) or the control ($F_{2,180}, p < 0.0001$). Within the coated seed treatments, *B. bassiana* treatments weighed significantly less than *I. fumosorosea* ($F_{2,180}, p < 0.0001$) and control treatments ($F_{2,180}, p < 0.0001$) while there was no difference between *I. fumosorosea* and control treatments ($F_{2,180}, p = 0.9999$). The height:width was affected by seed treatment ($F_{2,180}, p = 0.0180$), and was greater in non-coated seeds (Fig. 2). It was also affected by endophytes ($F_{2,180}, p < 0.0104$). *B. bassiana* showed a lower ratio than *I. fumosorosea* ($F_{2,180}, p = 0.0090$), but *B. bassiana* and *I. fumosorosea* did not affect plant height:width when compared to the control treatment ($F_{2,180}, p = 0.6507$; $F_{2,180}, p = 0.0835$). Coated seed treatments influenced endophyte effects on height:width ($F_{2,180}, p < 0.0001$). Within the non-coated treatments, *I. fumosorosea* and *B. bassiana* had increased height:width compared to the control ($F_{2,180}, p < 0.0001$; $F_{2,180}, p = 0.0183$) but there was no difference between *B. bassiana* and *I. fumosorosea* ($F_{2,180}, p = 0.6726$). Within the coated treatments, *I. fumosorosea* showed no difference to the control or *B. bassiana* treatments ($F_{2,180}, p = 0.4945$; $F_{2,180}, p =$

0.0640), while the ratio was significantly lower in *B. bassiana* treatments compared to the controls ($F_{2,180}, p < 0.0001$).

In summary, endophyte seed treatments had important effects on *T. erecta* development. *Isaria fumosorosea* negatively affected germination and seedling fresh weight, and produced seedlings with a less compact form. *Beauveria bassiana* reduced seedling fresh weight. Also, the speed and frequency of *T. erecta* germination, as well as plant mass and shape responded differently to each of the endophytes. Seeds inoculated with *B. bassiana* performed better than *I. fumosorosea*-inoculated seeds, which required more time to germinate and had lower germination rates. *Beauveria bassiana* treatment also led to smaller plants with a shorter, wider shape than those treated with *I. fumosorosea*. Seed coating did not affect germination responses to endophyte inoculation, but was involved in the endophyte effects on plant growth. *Isaria fumosorosea* effects on plant mass and form were negated when the seed coating was present. Alternatively, *B. bassiana* showed more effects on plant growth in the presence of the seed coating and may be more compatible than *I. fumosorosea* with other industry seed coating chemicals.

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Table 1. Mean (\pm standard error) percentage germination and median germination time (MGT) (hours) of non-coated and methylisothiazolinone-coated seeds inoculated with *B. bassiana*, sterile control, and *I. fumosorosea*.

	Non-coated			Coated		
	Control	<i>B. bassiana</i>	<i>I. fumosorosea</i>	Control	<i>B. bassiana</i>	<i>I. fumosorosea</i>
Germination (%)	81.7 \pm 8.85	80.8 \pm 4.50	63.0 \pm 9.69	93.3 \pm 8.61	95.0 \pm 7.42	91.7 \pm 7.38
MGT (h)	77.5 \pm 3.12	70.0 \pm 3.23	79.0 \pm 3.10	52.3 \pm 1.40	50.2 \pm 1.21	53.5 \pm 1.08

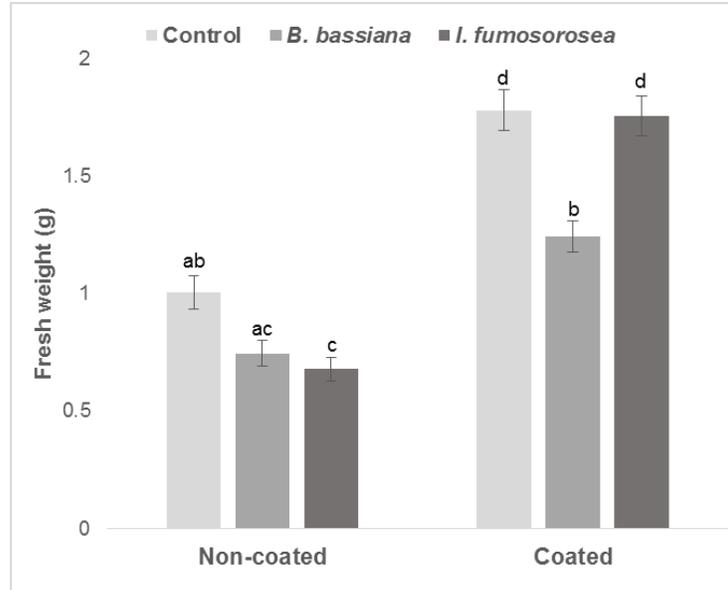


Figure 1. Mean fresh weight (g) of 3-week old *T. erecta* plants grown from non-coated and methylisothiazolinone-coated seeds inoculated with *B. bassiana*, sterile control, and *I. fumosorosea*. Shared letters indicate no significant difference between the treatments (Tukey HSD, $p > 0.05$). Error bars represent standard error.

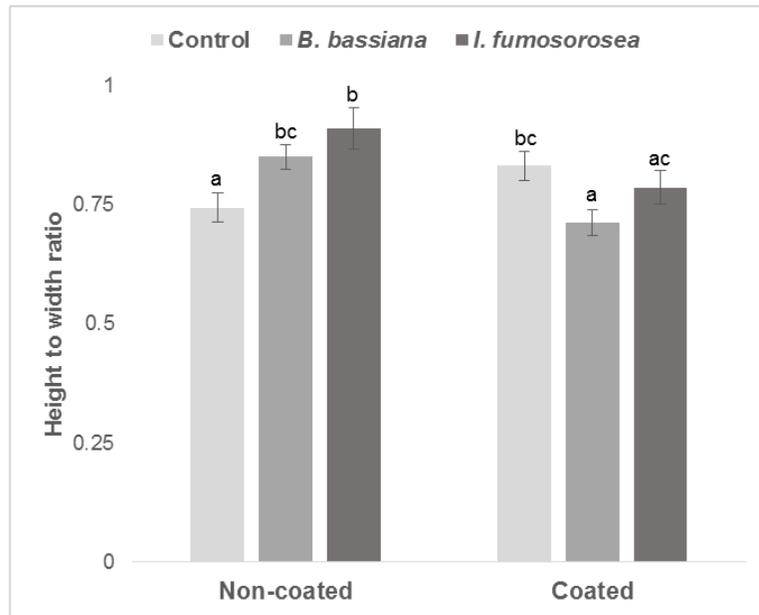


Figure 2. Mean height to width ratio of 3-week old *T. erecta* plants grown from non-coated and methylisothiazolinone-coated seeds inoculated with *B. bassiana*, sterile control, and *I. fumosorosea*. Shared letters indicate no significant difference between the treatments (Tukey HSD, $p > 0.05$). Error bars represent standard error.

Relative Salt Tolerance of Six Texas Superstar® Annuals

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Index Words Angelonia, brackish groundwater, pentas, petunia, salinity, reclaimed water

Significance to Industry High-quality water is becoming a limited resource for nursery production and public use. It may be inevitable to use alternative water sources such as municipal reclaimed water to irrigate landscapes and brackish groundwater to irrigate nursery crops and landscapes (1). Reclaimed water contains relatively high levels of soluble salts, which adversely affect plant growth and development (2). Using reclaimed water to irrigate nursery crops and landscapes is not a widely acceptable practice for nursery growers and homeowners due to limited information on the salt tolerance of nursery and landscape plants. To expand the usage of reclaimed water, salt tolerant plants should be identified in areas where reclaimed waters may be used for irrigation.

The Texas Superstar® program is established to identify landscape plants that show superb performance in Texas based on replicated field trials across the state (3,4). Those plants demonstrating outstanding landscape performance and superior pest tolerance are awarded the Texas Superstar® designation. However, salt tolerance is not considered during the evaluation process. *Angelonia angustifolia* 'Serena Lavender' ('Serena Lavender' angelonia) and 'Serenita Raspberry' ('Serenita Raspberry' angelonia), *Pentas lanceolata* 'Butterfly Deep Pink' (Butterfly Deep Pink pentas), *Petunia x hybrida* 'Tidal Wave Silver' ('Tidal Wave Silver' petunia), *Petunia x violacea* (laura bush petunia), and *Portulaca oleracea* 'Toucan Hot Mix' ('Toucan Hot Mix' purslane) are in the list of Texas Superstar® annuals. However, their responses to irrigation water with elevated salinity remain unknown. A greenhouse study was conducted by irrigating six Texas Superstar® annuals with a nutrient solution at an electrical conductivity (EC) of 1.2 dS·m⁻¹ (control) or salt solution at EC of 5.0 or 10.0 dS·m⁻¹ (EC 5 or EC 10) for seven weeks. Results showed that 'Toucan Hot Mix' purslane was the highest salt tolerant species without any foliar damage and growth reductions. Other annuals experienced minor foliar damage with varied growth reductions. 'Tidal Wave Silver' petunia, laura bush petunia, and 'Serenita Raspberry' angelonia plants had less growth reductions and were relatively more salt tolerant than 'Serena Lavender' angelonia and 'Butterfly Deep Pink' pentas.

Nature of Work 'Toucan Hot Mix' purslane, butterfly deep pink pentas, 'Tidal Wave Silver' petunia, and 'Serenita Raspberry' angelonia seeds were received from Ball Horticulture (West Chicago, IL), whereas laura bush petunia and 'Serena Lavender' angelonia seeds were purchased from Wildseed Farms (Fredericksburg, TX). On 31 January 2013, seeds

were sown into 72-cell trays (4 cm × 4 cm × 5.5 cm) containing a Red-earth® Plug and Seedlings germination mix (SunGro® Horticulture, Agawam, MA). All trays were placed under mist system with bottom heat (~25 °C). On 15 March, seedlings were transplanted to 2.3-L poly-tainer container (No.1P, 16.5 × 16.5 cm) filled with LP5 (SunGro Hort., Bellevue, WA). All plants were watered with a nutrient solution until treatments were initiated. The nutrient solution at EC of 1.2 dS·m⁻¹ was prepared by adding 1 g·L⁻¹ of 15 N-2.2 P-12.5 K (Peters 15-5-15; Scotts) to reverse osmosis (RO) water.

Treatments were applied on a weekly basis by irrigating plants with 1 L nutrient solution (control) or saline solutions at EC of 5.0 or 10.0 dS·m⁻¹ with 10% to 30% leaching fraction from 19 March to 2 May. In addition, plants were watered with 500 mL nutrient solution whenever the substrate surface was dry between treatment solutions. The saline solution was prepared by adding sodium chloride (NaCl) and calcium chloride (CaCl₂) at 2:1 (molar ratio) to the nutrient solution described above. All solutions were prepared in 100-L tanks with confirmed EC of 1.2 ± 0.1 (control, nutrient solution), 4.8 ± 0.2 (EC 5), and 9.0 ± 0.5 dS·m⁻¹ (EC 10) (mean and standard deviation) for the nutrient and saline solutions, respectively. The pH of the solutions was adjusted to 6.0 ± 0.4.

The substrate EC was determined using the pour-through method (Cavins et al., 2000) using an EC meter (Model B-173, Horiba, Ltd., Japan). The temperature in the greenhouse was maintained at 26.8 ± 4.3 °C (mean ± standard deviation) during the day and 22.0 ± 2.2 °C at night. The daily light integral (photosynthetically active radiation) was 16.1 ± 6.2 mol·m⁻²·d⁻¹.

Plant growth parameters were taken at the initiation of treatments (i.e. 19 March) and end of experiment (i.e. 7 May). Height (cm) from the pot rim to the tallest growth point of the plant was measured for 'Serena Lavender' angelonia, 'Butterfly Deep Pink' pentas, and 'Serenita Raspberry' angelonia. Due to their prostrate growth habits, the *length of the longest shoot (cm) was recorded for 'Tidal Wave Silver' petunia, Laura Bush petunia, and 'Toucan Hot Mix' purslane plants. Two perpendicular widths (cm) were recorded for all plants. Growth index was calculated as (height + width + perpendicular width)/3. Number of flowers (or inflorescences) was counted.*

Foliar salt damage (leaf edge burn, necrosis, and discoloration) was rated at the end of experiment on a visual score of 0 to 5, where 0 = dead; 1 = severe (over 90%); 2 = moderate (50% to 90%); 3 = slight (less than 50%); 4 = good quality with minor foliar damage; and 5 = excellent with no visible foliar damage. Leaf greenness (or relative chlorophyll content) of all plants was measured using a hand-held Soil-Plant Analysis Development (SPAD) chlorophyll meter (Minolta Camera Co., Osaka, Japan). For each plant, four leaves were randomly chosen for the measurements. Gas exchange was measured as described in Sun et al. (5). Upon termination of the experiment, shoots were severed at the substrate surface, and dry weight (DW) was determined after plants were oven-dried at 65 °C to constant weight.

The experiment followed a split-plot design with salinity (three levels) as the main plot and six species as subplot with eight replications. One-way analysis of variance (ANOVA) was

performed separately for each species for all data because the species had different growth habits. When salinity effect was significant, means separation among treatments was conducted using Tukey's honest significant difference (HSD) multiple comparison. All statistical analyses were performed using JMP 11 (SAS Institute Inc., Cary, NC).

Results and Discussion Substrate EC determined using the pour-through method provides clues about plant performance before deficiency or toxicity symptoms appear. During the period of the experiment, the leachate EC stayed around $3.0 \text{ dS}\cdot\text{m}^{-1}$ for plants irrigated with a nutrient solution (control) (Fig. 1). When plants were watered with a saline solution at EC of $5.0 \text{ dS}\cdot\text{m}^{-1}$ or $10.0 \text{ dS}\cdot\text{m}^{-1}$, the leachate EC increased from 5.1 to 9.1 $\text{dS}\cdot\text{m}^{-1}$ and from 7.8 to 11.9 $\text{dS}\cdot\text{m}^{-1}$, respectively. These results indicate that salt often accumulates in peat-based substrate. To reduce salt accumulation, saline solution was applied on a weekly basis instead of every irrigation. If saline solution is applied with every irrigation, salt accumulates more rapidly as reported in Niu et al. (6).

'Butterfly Deep Pink' pentas, 'Tidal Wave Silver' petunia, Laura Bush petunia, and 'Toucan Hot Mix' purslane plants had similar visual scores across treatments (Table 1). All plants had good quality with minor foliar damage or no visible foliar damage. Although salinity at EC 10 reduced the visual scores of 'Serena Lavender' angelonia and 'Serenita Raspberry' angelonia, plants still had good quality with minor foliar damage.

Growth index of Laura Bush petunia and 'Toucan Hot Mix' purslane plants were not different among treatments (Table 1). Salinity at EC 10 reduced the growth index of 'Serenita Raspberry' angelonia and 'Tidal Wave Silver' petunia by 13% and 11%, respectively. The growth index of 'Serena Lavender' angelonia and 'Butterfly Deep Pink' pentas decreased by 11% and 21% at EC 5 and 20% and 42% at EC 10, respectively.

Dry weights of 'Toucan Hot Mix' purslane plants were not different among treatments (Table 1). Salinity at EC 10 reduced the dry weight of 'Serenita Raspberry' angelonia, 'Tidal Wave Silver' petunia, and Laura Bush petunia plants by 17%, 17%, and 20%, respectively. The dry weights of 'Serena Lavender' angelonia and 'Butterfly Deep Pink' pentas decreased by 17% and 27% at EC 5 and 36% and 53% at EC 10, respectively.

The number of flowers or inflorescences of 'Serenita Raspberry' angelonia, 'Tidal Wave Silver' petunia, Laura Bush petunia, and 'Toucan Hot Mix' purslane plants was not different among treatments (Table 2). Both EC 5 and EC 10 significantly reduced the number of flowers of 'Serena Lavender' angelonia by 19%, whereas EC 10 decreased the number of flowers of 'Butterfly Deep Pink' pentas by 30%.

All plants had similar SPAD readings among treatments with the exception of 'Butterfly Deep Pink' pentas and Laura Bush petunia. The SPAD readings of 'Butterfly Deep Pink' pentas were reduced by 11% at EC 5, whereas those of Laura Bush petunia decreased by 17% at EC 5 and 19% at EC 10, respectively. Salinity at both EC 5 and EC 10 did not affect the leaf E , g_s , and P_n , indicating that salt treatment did not influence their photosynthesis (data not shown).

In conclusion, Texas Superstar® annuals have different growth and physiological responses to salinity. 'Toucan Hot Mix' purslane, a succulent plant, exhibited relatively the highest salt tolerance without any foliar damage and growth reductions. 'Tidal Wave Silver' petunia, Laura Bush petunia, and 'Serenita Raspberry' angelonia plants also had high salt tolerance with minor foliar damage, 17% to 20% reductions in shoot DW at EC 10, and without any changes in the number of flower. 'Serena Lavender' angelonia and 'Butterfly Deep Pink' pentas experienced minor foliar damage with varied reductions in shoot DW (17% and 36% at EC 5 and 27% and 53% at EC 10, respectively) and in the number of inflorescences (19% and 30% at EC 10 respectively). These indicated that they are relatively less tolerant to salinity compared to others. It is important to note that the results from this study may serve as a reference for selecting salt-tolerant Texas Superstar® annuals for landscapes where low quality water may be used. In general, all these annual species may be irrigated with alternative water sources with an EC of 5 dS·m⁻¹ or less with little or no growth reduction and foliar damage, as long as site is well drained.

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Table 1. Visual score, growth index, and shoot dry weight of Texas Superstar® annuals irrigated with nutrient solution [Control, electrical conductivity (EC) =1.2 dS·m⁻¹] or saline solutions (EC = 5.0 or 10.0 dS·m⁻¹, EC 5 or EC 10) in the greenhouse (n=8). Abbreviation in name: SL, 'Serena Lavender'; SR, 'Serenita Raspberry'; TWS, 'Tidal Wave Silver'; LB, Laura Bush.

Common name	Visual score ^z			Growth index (cm) ^y			Dry weight (g)		
	Control	EC 5	EC 10	Control	EC 5	EC 10	Control	EC 5	EC 10
SL Angelonia	5.5 a ^x	5.0 a	4.8 b	35.7 a	31.9 b	28.7 b	43.7 a	36.1 b	27.9 c
SR Angelonia	5.5 a	5.0 a	4.8 b	26.5 ab	27.5 a	23.0 b	25.6 ab	28.9 a	21.2 b
Pentas	4.9 a	4.8 a	4.4 a	21.2 a	16.7 b	12.4 c	32.5 a	23.8 b	15.2 c
TWS Petunia	5.0 a	5.0 a	5.0 a	110.3 a	112.1 a	97.8 b	105.6 a	108.3 a	87.6 b
LB Petunia	5.0 a	4.6 a	4.9 a	69.4 a	66.9 a	66.3 a	88.5 a	77.5 ab	70.4 b
Purslane	5.0 a	5.0 a	5.0 a	37.7 a	35.4 a	39.2 a	72.7 a	73.5 a	72.7 a

^z Visual score: 0 = dead; 1 = over 90% foliar salt damage (burning, necrosis, and discoloration); 2 = moderate (50% to 90%) foliar salt damage; 3 = slight (less than 50%) foliar salt damage; 4 = good quality with minor foliar salt damage; and 5 = excellent with no visible foliar salt damage.

^y Growth index = (height+width+perpendicular width)/3.

^x For each species, same letters among treatments are not significantly different by Tukey's honest significant difference (HSD) multiple comparison at $P < 0.05$.

Table 2. Number of flowers/inflorescences and relative chlorophyll content (SPAD) of Texas Superstar® annuals irrigated with nutrient solution [Control, Electrical conductivity (EC) =1.2 dS·m⁻¹] or saline solutions (EC = 5.0 or 10.0 dS·m⁻¹, EC 5 or EC 10) in the greenhouse (n=8). Abbreviation in name: SL, 'Serena Lavender'; SR, 'Serenita Raspberry'; TWS, 'Tidal Wave Silver'; LB, Laura Bush.

Common name	No. flowers/inflorescences			SPAD		
	Control	EC 5	EC 10	Control	EC 5	EC 10
SL Angelonia	57 a ^z	46 b	46 b	52.7 a	54.1 a	54.0 a
SR Angelonia	50 a	47 a	45 a	47.3 a	46.8 a	47.6 a
Pentas	11 a	9.4 ab	7.8 b	63.6 a	57.8 ab	57.0 b
TWS Petunia	255 a	232 a	212 a	50.3 a	49.9 a	48.3 a
LB Petunia	124 a	104 a	92 a	38.5 a	31.9 b	31.4 b
Purslane	30 a	32 a	27 a	33.5 a	36.2 a	34.9 a

^z For each species, same letters among treatments are not significantly different by Tukey's honest significant difference (HSD) multiple comparison at $P < 0.05$.

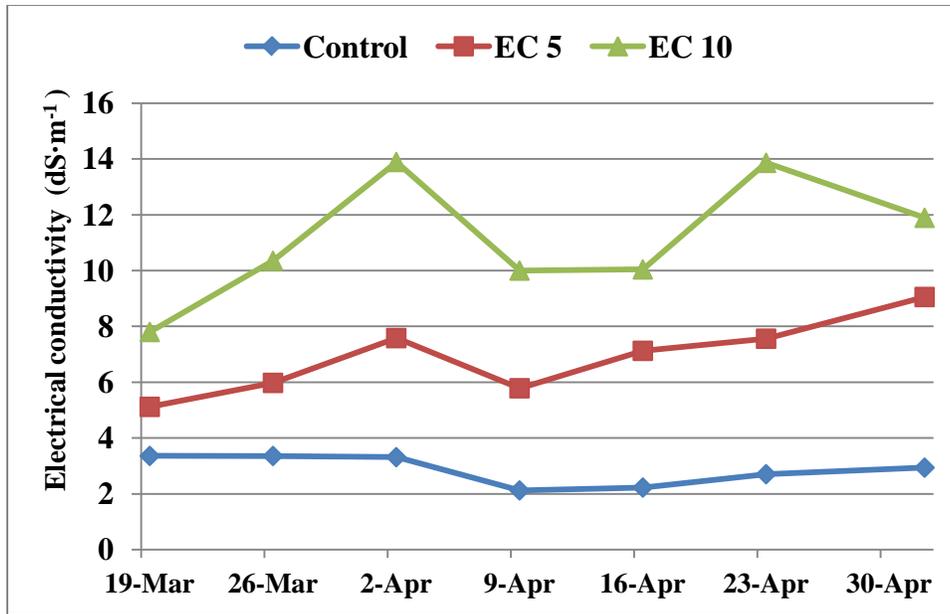


Figure 1. Leachate electrical conductivity (EC) measured after treatment solutions applied. Control, EC 5, and EC 10 represent nutrient solution at EC of 1.2 dS·m⁻¹, salt solutions at EC of 5.0 and 10.0 dS·m⁻¹, respectively. During the experimental period, nutrient and salt solutions applied weekly from 19 March to 2 May.