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Prohexadione-calcium affects growth and flowering of petunia and impatiens grown under photoselective films

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Abstract

The response of petunia (*Petunia x hybrida* Vilm.-Andr. 'Countdown Burgundy') and impatiens (*Impatiens wallerana* Hook 'Accent Orange Tempo') to Prohexadione-calcium was evaluated under a clear and a far-red light absorbing greenhouse (A_{FR}) film to investigate the dosage effect of Prohexadione-Ca and to determine if it can overcome the flowering delay under FR deficient greenhouse environments. Prohexadione-Ca reduced stem elongation of petunia and impatiens under A_{FR} and clear films when applied 3 weeks after germination. Late applications were less effective. In both crops, main stem length decreased in a quadratic pattern as the concentration of Prohexadione-Ca increased. Under both films, 50–100 mg l^{-1} Prohexadione-Ca resulted in ≈30% shorter petunia plants. Greater concentrations (500 and 1000 mg l^{-1}) resulted in excessively short plants (over 70%). Prohexadione-Ca at 100 mg l^{-1} delayed anthesis of petunia by 8 and 3 days under the clear film and the A_{FR} film, respectively during less inductive photoperiods but had no effect during inductive photoperiods. In impatiens, Prohexadione-Ca at 100 mg l^{-1} delayed anthesis over 10 days under clear or A_{FR} film. Greater concentrations (200 and 300 mg l^{-1}) inhibited flowering of impatiens. Prohexadione-Ca treatments significantly affected flower color development. Untreated petunia plants had dark burgundy flowers. Prohexadione-Ca treatment increased L*, a*, and C* values and decreased hue angle indicating that the flowers were faded. Flowers of untreated impatiens plants were bright orange color. Prohexadione-Ca at 100 mg l^{-1} increased L* value and decreased a*, b*, and C* values indicating that significant petal fading had occurred. Flowers of treated plants were

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nearly white under both films. Although effective in height control, loss of color would be a major limitation to the use of Prohexadione-Ca on flowering crops.

Keywords: Plant growth regulation; Spectral filters; Bedding plants

1. Introduction

Bedding plant industry typically uses chemical growth retardants to control height. A-rest (ancymidol), B-nine (daminozide), Bonzi (paclobutrazol), Cycocel (chlormequat chloride), and Sumagic (uniconazole) are among the most commonly used growth retardants to control bedding plant height. Bonzi and Sumagic are relatively new compounds compared to the rest. Although Bonzi is highly effective on most crops, it metabolizes slowly and therefore, is highly persistent. This is especially important in bedding plant production because residual effects can delay field growth and flowering. Growers often have problems with Bonzi and Sumagic causing excessive height reduction and slow growth after transplanting. Restrictions on the use of growth regulating chemicals have been set on food crops because of potential health risks to workers and consumers. The ornamentals industry is also faced with strict regulatory measures regarding chemical usage. Therefore, the horticultural industries are looking for alternative methods for height control.

Various non-chemical alternatives for plant growth regulation are being investigated. In collaboration with Mitsui Chemicals Inc. (Tokyo, Japan), we have been working to develop photoselective greenhouse covers that can reduce the transmission of far-red light as a non-chemical alternative to chemical height control. These covers were effective in controlling height of a wide range of herbaceous plants without using chemical growth retardants (Rajapakse and Young, 1999). However, flowering of certain short night requiring (long day, LD) plants [snapdragon (*Antirrhinum majus* L.) and petunia (*Petunia x hybrida* Vilm.-Andr.)] was delayed under FR light deficient environments (Cerny et al., 2003).

Prohexadione-Ca is a new plant growth retardant patented by Kumiai Chemical Industry Co. and has been registered for growth control of rice (*Oryza sativa* L.) in Japan (Evans et al., 1999). Recently, Prohexadione-Ca has been registered in the United States (under the trade name Apogee), as a replacement for daminozide, for use on apples (*Malus x domestica* Borkh.). Currently, the United States Environmental Protection Agency has classified Prohexadione-Ca as a reduced risk compound, because of the negligible toxicological effects on mammals. Prohexadione-Ca has been reported to degrade in higher plants with a half-life of few weeks and in soil with a half-life of less than a week. Therefore, Prohexadione-Ca is not persistent in the environment and has a low potential for bioaccumulation in the environment.

Most of the work with Prohexadione-Ca has been conducted on fruit and grain crops. Foliar application of Prohexadione-Ca at the rates of 125–250 mg l^{-1} has been reported to provide effective vegetative growth control in apples (Evans et al., 1999). Costa et al. (2001) reported that 100 mg l^{-1} of Prohexadione-Ca reduced shoot growth and the occurrence of fire blight in pear (*Pyrus communis* L.). Although Prohexadione-Ca may be used by the ornamental industry, there are very few published reports available. Foliar application of Prohexadione-Ca at the rate of 200 mg l^{-1} reduced stem elongation of ‘Bright Golden Anne’ chrysanthemums compared to untreated plants (Tatineni et al., 2000). Hisamatsu et al. (1998, 2000) reported that Prohexadione-Ca promoted flowering of stock (*Matthiola incana* L.) while Uniconazole prevented flowering. They also reported that Prohexadione-Ca enhanced the effect of applied gibberellins due to reduced inactivation. Maki et al. (2002) reported that the gibberellin metabolism (GA_{12} and GA_{19}) in chrysanthemum was slower under FR light deficient environments and that FR light deficient environments may also enhance the inactivation of biologically active gibberellins (GA_1). These observations suggest that Prohexadione-Ca may be used overcome the flowering delay under FR light deficient environments. This study was conducted to evaluate the use of Prohexadione-Ca on ornamental crops. Specific objectives were to investigate: (1) the dosage effects of Prohexadione-Ca on petunia and impatiens to select optimum concentrations, (2) the time of application to achieve optimum height control, and (3) if Prohexadione-Ca can overcome the flowering delay under FR deficient environments.

2. Material and methods

2.1. Plant material and culture

Petunia x hybrida Vilm.-Andr. ‘Countdown Burgundy’ and *Impatiens wallerana* Hook ‘Accent Orange Tempo’ were used in all experiments. Seeds were sown into 288-cell (6 mL per cell) plug trays filled with a commercial germination mixture (Fafard Germination Mix, Fafard Inc., Anderson, SC) and germinated on a greenhouse mist bench (20 s of mist every 30 min) set at 22 ± 2 °C. Two weeks after complete germination, uniform seedlings were transplanted individually into 165 mL pots containing a commercial potting mix (Fafard 3-B Mix). Plants were acclimatized for 1 week in the greenhouse before treatments. Plants were watered as needed and fertilized at each irrigation with 1 g l^{-1} of 20N–4.4P–16.7K water-soluble fertilizer (Peters 20-10-20 Peat-Lite Special, Scotts-Sierra Horticultural Products Co., Marysville, Ohio) during the experiments. Greenhouse cooling/heating set points were 27 °C/18 °C.

2.2. Photoselective growth chambers

A clear film and an experimental photoselective greenhouse film with a far-red light absorbing dye (A_{FR} film) were used in all experiments (Mitsui Chemicals Inc., Tokyo, Japan). Four polyvinyl chloride framed growth chambers (1.2 m × 1.2 m × 1.2 m; two for each film) were covered with these films. In each chamber, two fans (one attached to the lower left corner and the other attached to the upper right corner of the opposite side) were oriented in the same flow direction to prevent heat buildup. All growth chambers were randomly placed inside a glass greenhouse. Photosynthetic photon flux (PPF) transmission of the clear and A_{FR} film was 90 and 75%, respectively. Photosynthetic photon flux was adjusted for uniformity in all chambers with neutral density filters. Daily light integral

(DLI) inside the photoselective chambers were recorded with a LI-1000 data logger (LI-COR, Lincoln, Neb) fitted with LI-190SB quantum sensors (programmed to collect readings every 5 min) during each set of experiments. The photoperiod was determined based on the U.S. Naval Observatory Sunrise and Sunset Table for Greenville, SC. Hourly air temperature inside chambers was monitored using type T thermocouples placed above the plant canopy and connected to an Omnidata EasyLogger (EL 824, Omnidata International Inc., Logan, Utah).

Spectral quality inside the chambers was measured on clear days between 12:00 and 14:00 h at the beginning and end of each experiment using a LI-1800 spectroradiometer fitted with a LI-1800-10 remote cosine sensor. The dye in the A_{FR} film intercepted FR and *R* wavelengths with maximum absorption at 730 nm. The phytochrome photoequilibrium (P_{fr}/P_{total}) estimates of transmitted light for the clear and A_{FR} films were 0.71 and 0.77, respectively (Sager et al., 1988). The broad band *R*/FR ratios (*R* = 600–700 nm; FR = 700–800 nm) for the clear and A_{FR} films were 1.05 and 1.51, respectively.

2.3. Dose response to Prohexadione-Ca

In the initial experiment with petunia, Prohexadione-Ca (BAS 125 10W, BASF Corp., Research Triangle Park, NC) at 0, 100, 500, or 1000 mg l^{-1} was evaluated. Each solution contained 0.1% Tween 20 as a surfactant (Fisher Scientific, NJ). Forty-eight plants were transferred to each chamber after 1-week acclimatization period. A set of 12 plants in each chamber was sprayed to run off three times at 2-week intervals with each of the above Prohexadione-Ca solutions. First application of Prohexadione-Ca was made 3 weeks after germination (plants had five to six leaves). Plants received average daily photoperiod of 10.5 h day^{-1} and an average daily photosynthetic light integral of 6.6 ± 3.1 mol m^{-2} day^{-1}. Average day and night temperatures were not different among film treatments and were 21.6 ± 0.2 °C and 19.2 ± 0.1 °C, respectively.

Concentrations over 100 mg l^{-1} caused excessive height reduction and delay in flowering of petunia. Therefore, the experiment was repeated with lower concentrations to select the optimum concentration of Prohexadione-Ca. This experiment was conducted only under the clear film because the same trend was observed under the A_{FR} film. Forty-eight plants were transferred to clear chamber. A set of 12 plants was sprayed to run off three times at 2-week intervals with 0, 50, 100, or 150 mg l^{-1} Prohexadione-Ca solution. Plants received an average daily photoperiod of 12 h day^{-1} and an average daily photosynthetic light integral of 10.3 ± 4.8 mol m^{-2} day^{-1}. Average day and night temperatures were not different among film treatments and were 21.1 ± 0.2 °C and 19.2 ± 0.1 °C, respectively.

In a separate experiment, effect of Prohexadione-Ca on growth and flowering of impatiens plants was evaluated. Plants were grown as described previously. Twenty-four plants were transferred to each chamber. A set of six plants in each chamber was sprayed to run off three times at 2-week intervals with 0, 100, 200, or 300 mg l^{-1} Prohexadione-Ca solution. Plants received an average daily photoperiod of 14 h day^{-1} and an average daily photosynthetic light integral of 14.8 ± 6.0 mol m^{-2} day^{-1}. Average day and night temperatures were not different among film treatments and were 25.3 ± 0.1 °C and 21.0 ± 0.1 °C, respectively.

2.4. Timing of applications

Plants were grown as described previously inside a glass greenhouse. A set of 24 petunia and 12 impatiens plants were treated with 100 mg l^{-1} Prohexadione-Ca solution containing 0.1% surfactant at 4, 5, or 6 weeks after germination. Control plants were treated with water and surfactant. One week after application, half of the plants (12 petunia and six for impatiens) received a second application of Prohexadione-Ca solution. The experiments were terminated when all plants had flowered. Plants received an average daily photoperiod of 14.5 h day^{-1} and an average daily photosynthetic light integral of 28.5 ± 6.7 mol m^{-2} day^{-1}. Average day and night temperatures were 26 ± 0.1 °C and 23 ± 0.1 °C, respectively.

2.5. Experimental design, data collection, and analysis

The experimental chambers were randomly placed inside the greenhouse. Two replicate chambers were used for each film treatment. In the dose response experiment, Prohexadione-Ca concentrations were randomly arranged within a chamber. The data were analyzed as a split plot with photoselective films (clear and A_{FR}) as the whole plot factor and concentration as the split plot factor. Twelve single plants of petunia and six plants of impatiens were used in each treatment combination/replicate. Main stem length (from growing medium surface to apex), days to anthesis, total number of flowers (open flowers and unopened buds), leaf greenness, shoot dry weight, and flower color were recorded. Leaf greenness was measured with a Spad-502 meter (Spectrum Technologies Inc., Plainfield, IL) on four leaves below the apex of each plant. For dry weight measurements, shoots were oven-dried for 2 days at 90 °C. Flower color was measured using a Minolta Chroma meter with a CIE Illuminant C light source and calibrated using white CR-A43 calibration plate (DP-301 with CR-300 measuring head; Minolta Corp., Ramsey, NJ). Color was recorded using the CIE 1976 L*a*b* uniform color space (CIE-Lab). Chroma (C*) and hue angles (h°) were recorded. The L* value is a useful indicator of darkness or lightness of petals. The hue is the angle in a color wheel of 360°, with 0°, 90°, 180°, and 270° representing the hues red/purple, yellow, blue/green, and blue, respectively (McGuire, 1992; Nunes et al., 2002). Chroma is the color saturation or intensity of color, a* represents hue of red/purple (+a) to blue/green (−a) on horizontal axis and b* represents hue of yellow (+b) to blue (−b) on the vertical axis. Data were subjected to regression analysis (analysis of variance where necessary and means were compared using LSD at $P = 0.05$; SAS Inst., Inc., Cary, NC).

3. Results and discussion

3.1. Dose response to Prohexadione-Ca

Petunia and impatiens plants grown under the A_{FR} film, without Prohexadione-Ca treatment, were ≈15% shorter than those grown under the clear film (Fig. 1A and C). In petunia, main stem length decreased in a quadratic pattern as the concentration of

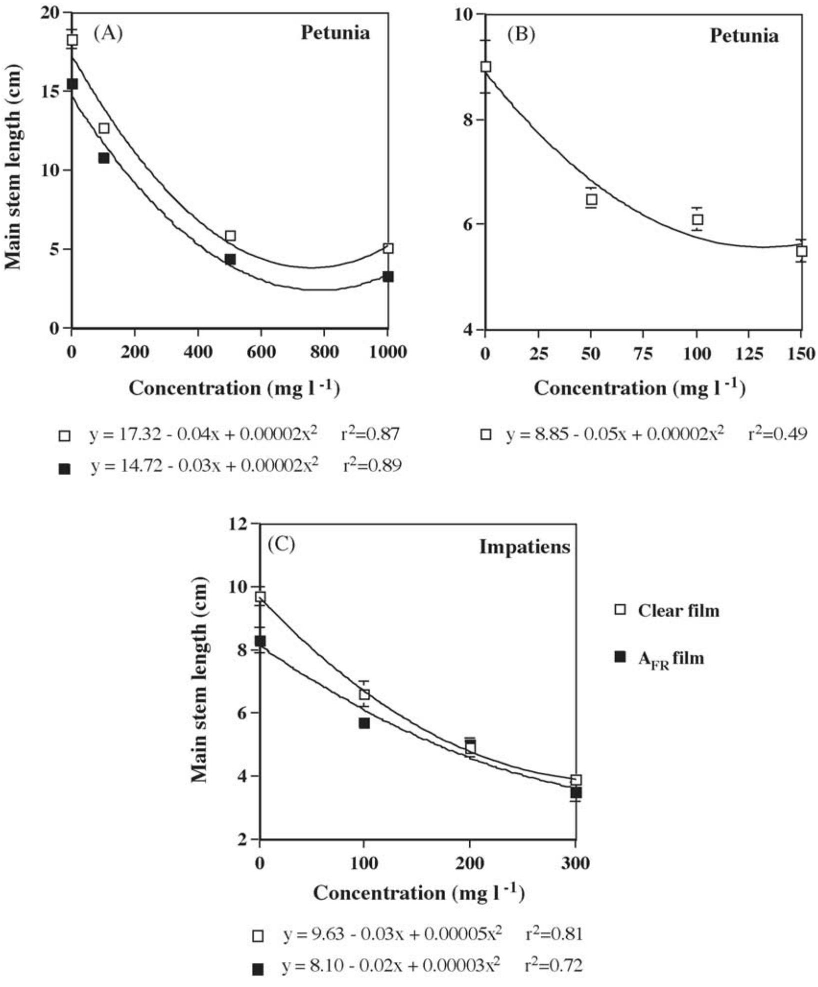

Fig. 1. Effect of Prohexadione-Ca dose on the main stem length of petunia (A) 0, 100, 500, or 1000 mg l^{-1}; (B) 0, 50, 100, or 150 mg l^{-1} (clear only) and impatiens (C) 0, 100, 200, or 300 mg l^{-1} plants grown inside clear and FR-absorbing (A_{FR}) film chambers. Plants were sprayed to run off three times at 2-week intervals beginning 3 weeks after germination. Vertical bars indicate standard error (S.E.; $n = 24$ for petunia and 12 for impatiens). If vertical bars are not seen, S.E. was smaller than the symbol.

Prohexadione-Ca increased from 0 to 1000 mg l^{-1} (Fig. 1A). There was no significant interaction between film and Prohexadione-Ca treatment. There was no difference in height between 500 or 1000 mg l^{-1} prohexadione treatments. Under both films, 100 mg l^{-1} Prohexadione-Ca resulted in ≈30% shorter petunia plants while high concentrations (500 and 1000 mg l^{-1}) resulted in excessively short plants (over 70%). Therefore, in a separate experiment, lower concentrations (0–150 mg l^{-1}) were evaluated to select optimum concentration of Prohexadione-Ca that can effectively control petunia height. Prohexadione-Ca at 50 mg l^{-1} reduced stem elongation ≈30% while higher concentrations (100 or 150 mg l^{-1}) had no significant further reduction in stem elongation (Fig. 1B).

In impatiens, stem length decreased in a quadratic pattern as the concentration of Prohexadione-Ca increased from 0 to 300 mg l^{-1} (Fig. 1C). Under both films, 100 mg l^{-1} Prohexadione-Ca resulted in ≈30% shorter impatiens plants. Higher concentrations resulted in further reduction in height (200 and 300 mg l^{-1} reduced height 40 and 60%, respectively).

Far-red light absorbing film delayed anthesis of petunia by 8 days (Table 1). Prohexadione-Ca at 100 mg l^{-1} delayed anthesis of petunia by 8 and 3 days under the clear film and the A_{FR} film, respectively (Table 1). Prohexadione-Ca at 500 mg l^{-1} resulted in further delay in anthesis under clear film (2 weeks). Under the A_{FR} film, 500 mg l^{-1} Prohexadione-Ca treated plants did not flower. Plants treated with 1000 mg l^{-1} Prohexadione-Ca did not flower under any film treatment. Prohexadione-Ca reduced flower size (length and diameter) and number of flowers. High concentrations of Prohexadione-Ca (500 or 1000 mg l^{-1}) caused excessive reduction in number of flowers and dry weight under both films. Leaves of Prohexadione-Ca treated plants were darker green than the leaves of non-Prohexadione-Ca treated plants. Leaf color intensity increased as the concentration increased.

Low concentrations of Prohexadione-Ca (50–150 mg l^{-1}) did not significantly affect anthesis of petunia (Table 2). Prohexadione-Ca at 50–150 mg l^{-1} did not affect the total number of flowers developed but decreased the shoot dry weight. However,

Table 1
Effects of Prohexadione-Ca dose (P-Ca; 0–1000 mg l^{-1}) on days to anthesis (DF), flower length (FL), flower diameter (FD), total number of flowers (NF, open and unopened buds), shoot dry weight (SDW), and leaf greenness (SPAD) of petunia plants grown inside clear and far-red light absorbing (A_{FR}) film chambers. Each number is the mean of 24 plants

Film (F)	P-Ca (mg l^{-1})	DF	FL (cm)	FD (cm)	NF	SDW (g)	SPAD
Clear	0	72	7.7	8.6	6.9	2.9	31.9
	100	80	6.1	7.2	6.4	2.3	33.6
	500	86	5.9	5.8	1.8	1.3	38.5
	1000	–[a]	–	–	0.5	1.0	39.5
L		***[b]	**	***	***	***	***
Q		***	***	***	***	***	***
A_{FR}	0	80 b	7.1 a	7.9 a	4.7	1.9	34.9
	100	83 a	6.1 b	6.6 b	3.7	1.6	34.5
	500	–	–	–	0.1	0.9	39.8
	1000	–	–	–	0	0.7	37.7
L					***	***	***
Q					***	**	***
ANOVA							
F		*	Ns	Ns	*	*	*
P-Ca			**	**	***	**	**
F × P-Ca					*	Ns	*

[a] Did not flower.
[b] Ns, *, **, *** non-significant or significant at $P = 0.05$, 0.01, or 0.001, respectively.

Table 2
Effects of Prohexadione-Ca dose (P-Ca; 0–150 mg l^{-1}) on days to anthesis (DF), number of flowers (NF, open and unopened buds), shoot dry weight (SDW), specific shoot dry weight (SSDW), and flower color characteristics (L*, a*, b* values and hue angle) of petunia plants grown inside clear film chamber

Film (F)	P-Ca (mg l^{-1})	DF[a]	NF[a]	SDW[a] (g)	SSDW[a] (g cm^{-1})	Petal color characteristics[b]				
						L*	a*	b*	C*	h°
Clear	0	49	5.1	1.6	0.18	14.1	40.1	−2.7	40.3	357
	50	50	4.1	1.3	0.20	27.2	54.7	−8.8	55.5	351
	100	51	4.8	1.2	0.20	38.9	56.8	−13.2	58.2	347
	150	50	5.2	1.2	0.21	47.8	50.0	−13.4	52.8	345
L		Ns[c]	Ns	*	Ns	***	***	***	***	***
Q		Ns	Ns	Ns	Ns	***	***	***	***	**

[a] Each number is the mean of 24 plants.
[b] Each number is mean of 20–30 flowers in a treatment using Minolta DP-301 chroma meter with CR-300 measuring head using CIELAB. L* = lightness, a* = bluish green/red purple hue component, b* = yellow/blue hue component, C* = chroma, and h° = hue angle.
[c] Ns, *, **, *** non-significant or significant at $P = 0.05$, 0.01, or 0.001, respectively.

Prohexadione treatment did not significantly influence the specific shoot dry weight, indicating that the smaller plants, not a reduction in photosynthesis, caused the dry weight reduction.

Prohexadione-Ca treatments significantly affected flower color development of petunia. Untreated petunia plants had dark burgundy flowers as indicated by the low lightness factor (*L*) and high hue angle (Table 2). As the concentration of Prohexadione-Ca increased, L* value increased suggesting that petal fading occurred. Hue angle decreased as Prohexadione-Ca concentration increased. Prohexadione-Ca treatment increased a* and C* values indicating that the flowers had red appearance. Prohexadione-Ca has resulted in loss of flower color, especially in crops with red coloration (Rademarcher, personal communication). In zinnia 'Dreamland Scarlet', application of Prohexadione-Ca to plants with visible flower buds resulted in yellow flowers instead of typical red color. It has taken over 2 months to produce normal red flowers following Prohexadione-Ca application. When applied to plants with both open flowers and flower buds, Prohexadione-Ca had no effect on petal color of already opened flowers (which were red) but petals of newly open flowers were yellow suggesting that Prohexadione-Ca interferes with anthocyanin production but does not destroy existing anthocyanin.

The response of petunia plants to Prohexadione-Ca was different between two experiments. For example, in the first experiment where high concentrations (up to 1000 mg l^{-1}) was tested, plants were generally taller than the plants in the second experiment where lower concentrations (up to 150 mg l^{-1}) were tested. Stem length leveled off at 500–1000 mg l^{-1} but in the second experiment it leveled off at 100–150 mg l^{-1}. Anthesis of petunia plants in the second experiment took place about 3 weeks earlier than the plants in first experiment. Prohexadione-Ca at 100 mg l^{-1} delayed anthesis by about a week in the first experiment but had no effect in the second experiment. The differential response between the two experiments can be attributed to the environmental conditions during the course of the experiments. First experiment was conducted during November to February where plants received $\approx 6.6 \pm 3.1$ mol day^{-1}

of PPF and a photoperiod of ≈10.5 ± 0.5 h. The second experiment with lower concentrations (50–150 mg l^{-1}) was carried out during February to April, where plants received 10.3 ± 4.8 mol day^{-1} of PPF and 12 ± 1 h photoperiod. Petunia is a quantitative long day plant and flowering is hastened during long photoperiods (Piringer and Cathey, 1960; Adams et al., 1998). Adams et al. (1999) reported that low light and low temperature prolonged the juvenile growth period of petunia. The environmental conditions during the first experiment with high doses were less inductive for flowering of petunia than the conditions during the second experiment with lower doses. Therefore, the vegetative growth period was longer in the first experiment and caused longer main stems than the plants in the second experiment. Inductive conditions during the second experiment caused petunia plants to flower early and less responsive to Prohexadione-ca in terms of delaying anthesis. Cerny et al. (2003) reported that floral primorida initiation and floral development of petunia was not affected by A_{FR} films during inductive photoperiods but was delayed up to 12 days by A_{FR} films during less-inductive photoperiods.

Far-red light absorbing film did not affect anthesis of impatiens (Table 3). Prohexadione-Ca at 100 mg l^{-1} delayed anthesis over 10 days under clear or A_{FR} film.

Table 3
Effects of Prohexadione-Ca (P-Ca; 0–300 mg l^{-1}) on days to anthesis (DF), number of flowers and buds (NFB), and flower color characteristics (L, a, b values and hue angle) impatiens plants grown inside clear and far-red light absorbing (A_{FR}) film chamber

Film (F)	P-Ca (mg l^{-1})	DF[a]	NFB[a]	Petal color characteristics[b]				
				L*	a*	b*	C*	h°
Clear	0	22 b	25.4	51.4 b	51.8 a	50.1 a	72.1 a	43.9 a
	100	35 a	19.1	79.0 a	13.1 b	7.1 b	14.9 b	29.1 b
	200	–[c]	0.9	–	–	–	–	–
	300	–	1.1	–	–	–	–	–
L			–*					
Q			Ns					
A_{FR}	0	22 b	21.4	52.2 b	51.7 a	50.9 a	72.6 a	44.5 a
	100	33 a	12.8	79.1 a	12.2 b	8.0 b	14.8 b	32.3 b
	200	–	2.9	–	–	–	–	–
	300	–	0.2	–	–	–	–	–
L			**					
Q			Ns					
Anova								
F		Ns[d]	Ns	Ns	Ns	Ns	Ns	Ns
P-Ca		*	**	***	***	***	***	***
F × P-Ca		Ns	Ns	Ns	Ns	Ns	Ns	Ns

[a] Each number is mean of 12 plants.
[b] Each number is mean of 20–30 flowers in a treatment using Minolta DP-301 chroma meter with CR-300 measuring head using CIELAB. L* = lightness, a* = bluish green/red purple hue component, b* = yellow/blue hue component, C* = chroma, and h° = hue angle.
[c] did not flower.
[d] Ns, *, **, *** are non-significant or significant at $P = 0.05$, 0.01, or 0.001, respectively.

Plants treated with 200 or 300 mg l^{-1} Prohexadione-Ca did not flower under both films. Prohexadione-Ca (100 mg l^{-1}) reduced the number of flowers under clear and A_{FR} film but the reduction was greater under A_{FR} film (25% versus 40% under clear and A_{FR} films, respectively). Greater concentrations (200 or 300 mg l^{-1}) caused excessive reduction (over 90%) in number of flower buds.

Prohexadione-Ca had a greater influence on petal color development of impatiens than the petunia (Table 3). Flowers of untreated impatiens plants were bright orange color as indicated by hue angle, a*, b*, and C* values. Prohexadione-Ca at 100 mg l^{-1} increased L* value and decreased a*, b*, and C* values, indicating significant petal fading. Flowers of treated plants were nearly white under both film treatments.

The transition from vegetative to reproductive growth is controlled by environmental and endogenous factors. Endogenous gibberellins control the stem elongation and flowering of some long day plants and cold-requiring plants. Exogenous gibberellins can substitute for long day requirement under less inductive photoperiods in many LD plants and for the cold requirement of plants that require a cold-requiring plants (Zeevaart, 1985). Prohexadione-Ca is a late stage gibberellin biosynthetic inhibitor that blocks

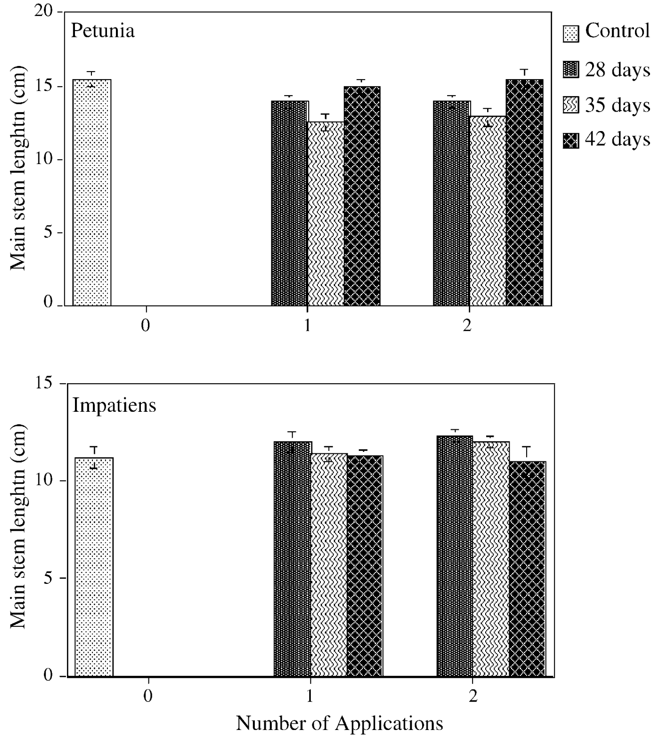

Fig. 2. Effect of developmental stage and number of Prohexadione-Ca (100 mg l^{-1}) applications on main stem height of petunia and impatiens. Plants were treated on 4, 5, or 6 weeks after germination. One week after each initial application, half of the plants received a second application of Prohexadione-Ca solution. Vertical bars indicate standard error ($n = 24$ for petunia and 12 for impatiens).

3β-hydroxylation causing a reduction in active gibberellins such as GA_1 and GA_4. It has also been shown to prevent catabolism of active gibberellins to inactive gibberellins such as GA_8 by blocking the 2β-hydroxylation (Brown et al., 1997). Therefore, it has been reported that Prohexadione-Ca can promote stem elongation and induce flowering due to reduced catabolism of gibberellins (Hisamatsu et al. (1998, 2000). However, our results show that Prohexadione-Ca delayed flowering significantly in petunia and impatiens and not effective in promote flowering under A_{FR} films.

3.2. Timing of applications

Time of application had a significant influence on stem elongation of petunia but not on impatiens (Fig. 2). One or two applications of Prohexadione-Ca, made 4 or 5 weeks after germination, reduced the height of petunia plants compared to untreated plants. However, the magnitude of height reduction was lower compared to previous experiments where the initial application was made 3 weeks after germination. When sprayed 6 weeks after germination, Prohexadione-Ca was not effective in height reduction. In impatiens, applications made 4 weeks after germination were not effective in controlling height.

In petunia, time and number of applications had no significant effects on anthesis or the number of flowers (Table 4). In general, applications made 4 or 5 weeks after germination resulted in flower fading (data not shown). Application made 6 weeks after germination had no effect on flower color showing that Prohexadione-Ca did not promote anthocyanin destruction.

In impatiens, applications made 4 or 5 weeks after germination delayed anthesis, but application after 6 weeks had no effect on anthesis. Prohexadione-Ca treated plants had significantly fewer number of flowers (or buds). Regardless of the time of application, flower fading occurred in treated plants (data not shown). Flower fading increased the

Table 4
Effects of time of application (days after germination) and the number of applications of Prohexadione-Ca on anthesis (DF) and number of flowers (NF: open and unopened) of petunia and impatiens plants

Time (*T*)	Number (*N*)	Petunia		Impatiens	
		DF[a]	NF[a]	DF[a]	NF[a]
(Weeks)					
–	0	31	6.1	32	47.8
4	1	26	9.0	38	23.8
	2	29	7.7	36	23.2
5	1	29	7.0	36	16.7
	2	28	7.9	35	21.4
6	1	28	7.4	34	20.7
	2	28	7.1	33	16.3
ANOVA					
T		*[b]	Ns	*	*
N		Ns	Ns	Ns	Ns
T × *N*		*	Ns	Ns	Ns

[a] Each number is the mean of 24 plants (petunia) or 12 (impatiens) plants.
[b] NS, *, **, *** non-significant or significant at $P = 0.05$, 0.01, or 0.001, respectively.

application was delayed. Flowers of plants treated 42 days after germination were nearly white. Unlike anthocyanins, results suggest that Prohexadione-Ca may be promoting the destruction of carotenoid pigments.

4. Conclusions

Our results show that Prohexadione-Ca was effective in reducing stem elongation of petunia and impatiens and that response varies with the environmental conditions. Concentration ranging from 50 to 100 mg l^{-1} can be used to achieve a 30% height reduction. However, petal fading occurred in both petunia and impatiens. Delayed application reduced petal fading of petunia but it was not effective in height control. In impatiens, delayed application was not effective in height control and resulted in petal fading. Although effective in height control, color loss would be a major limitation to the use of Prohexadione-Ca on ornamental crops.

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