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GROWTH, FLOWERING AND GAS EXCHANGE OF *Ruellia brittoniana* TREATED WITH DIFFERENT CONCENTRATIONS AND APPLICATION FREQUENCIES OF DAMINOZIDE

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ABSTRACT

Ruellia brittoniana is a fast growth plant with a strong tendency of vegetative growth that occurs at the expense of flowering capability, thus regular pruning for height control is necessary. Chemical pruning using suitable growth retardant may offer similar effects on stem and branch elongation, and flowering of the plants could be promoted. This study was carried out to evaluate the varying rate of daminozide and frequencies of application on the growth and flowering of *Ruellia brittoniana.* Results of a two factorial experiment involving two application frequencies (double and triple application) and five concentrations of daminozide (0, 500, 1000, 1500, 2000 mg L^{-1}) showed that different concentration of daminozide and application frequencies significantly affected the vegetative growth, photosynthesis, transpiration and chlorophyll content. Daminozide at 500 mg L^{-1} with triple application caused a reduction of 42% in plant height compared to control with less detrimental effects on flower number. Daminozide application had not affected on the stomatal conductance. Application of daminozide at 500 mg L^{-1} given in triple application was found to be effective in producing short, compact plants with more flowers.

Key words: *Ruellia brittoniana*, daminozide, growth retardant

INTRODUCTION

Ruellia brittoniana is one of the most frequently blooming shrubs. The plant grows quickly and frequent pruning is needed to maintain its stature. To control this problem, effort has been made to find a substitute technique in slowing the plant growth and consequently avoiding the need of frequent pruning to maintain its aesthetic formations for a long time. Plant response towards growth constraints varies not only between the type plants but also in response to various concentration and application technique of the growth regulators. The interest of horticulturist is always to develop a balanced growth of landscape plants by reducing the growth rate and maintaining its stature without affecting their flowering capability. Chemical pruning using suitable growth retardant may offer beneficial reducing effects on branching, thus producing short stature, compact plants with profuse flowers.

Growth retardants have an inhibitory effect on cell division and enlargement of cell in plants. Therefore, they are widely used for height control in floriculture (Pasian, 1999). Studies on zinnias as potted plants showed that the height of plants was reduced, producing high-quality marketable compact plants with the aid of growth retardants (Pinto, 2005). Daminozide is an effective height retardant labelled for use in azalea, pot chrysanthemum, gardenia, hydrangea and many bedding and foliage plants. Azalea treated with daminozide promoted early and more extensive flower-bud set and retarded vegetative shoot development. In general, it is not phytotoxic and has a short-term effect that seldom results in over stunting of treated plants (Latimer *et al*., 2001).

However, plant response to plant growth retardants may differ even within the same species and depends on the retardant type, dose and

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application method, frequency of application, age of plant, nutritional status and environmental conditions (Salachna & Zawadzinska, 2013, Sprzaczka & Laskowska, 2013, Ahmad *et al.,* 2015). Excessive concentrations may result in plants which are extensively retarded and often of poor quality. It is important to determine the optimum concentration levels and application frequencies, as well as the most economical and practical application methods in order to produce commercially acceptable plants. The study reported here was aimed at assessing the effect of different concentrations and application frequencies of daminozide on leaf photosynthesis, vegetative growth and flowering of *Ruellia brittoniana* in view of its application to inhibit the growth of the species and at the same time to promote its flowering.

MATERIALS AND METHODS

Plant materials and growth condition

The plant species, *Ruellia brittoniana* were obtained from local nursery at Sungai Buluh, Malaysia and the seedlings were planted in polyethylene bags with cocopeat. The plants were obtained after two months of the establishments using hardwood cutting at University's Agriculture Park nursery of Universiti Putra Malaysia. The plants were pruned to make them more uniform by cutting the shoots. Each plant was pruned to about the same height with an approximate height of 12 to 15 cm in length and having 2-3 nodes. The young leaves were fully emerged after 15 days following the trimming. The plants were fertilized a month after planting with a complete fertilizer (NPK-8.0N: $8.0P_2O_5$: $8.0K_2O$: 3MgO) about 6 g/plant every two weeks during the two months of experimental period.

Daminozide treatments

Daminozide was applied with two application frequencies (double and triple application). The plants were treated with a daminozide at five concentrations (0, 500, 1000, 1500, 2000 mg L^{-1}), 50 mL/plant through foliar spray. The plants were sprayed with a fine spray using hand sprayer. The control plants were treated with tap water.

Vegetative growth

Plant height was measured after daminozide treatment. Measurement of plant height was taken from the point of cutting to the highest shoot tip using a measuring tape. The internode length was determined by dividing the length of the stem with the number of nodes. Leaf areas were measured and recorded as total leaf area per plant using automatic leaf area meter (MODEL LI-300, LI-COR). The number of flowers emerged was counted. The whole plants were then separated into leaves, stems and roots and the dry weight of each part was determined after 72 hours at 75°C in a drying oven. Relative leaf chlorophyll content of three leaves per plot was measured at 45 days after treatment by using a Minolta-chlorophyll meter (SPAD-502 plus, KONICA MINOLTA OPTIC, Inc, Japan).

Net photosynthesis, stomatal conductance and transpiration

At 45 days of daminozide treatment, net photosynthesis rate, stomatal conductance and transpiration rate were measured on the selected of the third fully expanded leaves by using a portable close photosynthesis measurement system (Infra-Red Gas Analyzer, Li 6400, Licor, Lincoln, Nebraska, USA). The measurements were taken around 11:00 am using five measurements for each replication with an irradiance setting of 1000 µmol m^{-2} s^{-1}. Irradiance was provided by an LED RGB (Red Green Blue) light source (LI-6400-02B, Li-Cor Inc.).

Experimental design and data analysis

The treatments were arranged in a randomized complete block design (RCBD) with five daminozide treatments and two application frequencies with three blocks and each plot comprised of eight polybags. Data generated were subjected to analysis of variance (ANOVA) utilizing SAS software (Version 9, SAS Institute Inc. Cary, North Carolina, USA) and differences between treatments means were compared using Duncan's Multiple Range Test (DMRT) at $P \leq 0.05$ levels. Pearson correlation coefficient (r) was determined between the variables in each species at $P \leq 0.05$ levels.

RESULTS AND DISCUSSION

Vegetative growth

The result clearly shown that plant height was significantly affected ($P<0.01$) by daminozide concentrations (Table 1). Decrease in stem growth was recorded 64% at 2000 mg/L compared to control. The plant height was decrease with the increasing level of daminozide and application frequency. Increasing the frequency of applications has resulted in greater reductions in plant height for all levels of daminozide. This is an agreement to the result obtained by Poll and Allan (1990), where treatment of daminozide at 1250 and 2500 mg L^{-1} with triple application reduced plant height of the *Petunia hybrida.* Daminozide at 4250 mg L^{-1} was recommended for *Eucomis autumnalis* with double and three times reduced plant height compared to single application (Krause *et al*., 2003). The results

Table 1. Effects of the daminozide concentration and application frequency on plant height, internode length, number of flowers, leaf area, leaf, stem and root dry weight and relative chlorophyll content

TREATMENT	Plant height (cm)	Internode length (cm)	Number of flowers/ plant	Leaf area/ plant (cm^2)	Leaf dry weight (g)	Stem dry weight (g)	Root dry weight (g)	Relative chlorophyll content
Daminozide concentration (mg L^{-1})								
Control	49.67a	77.07a	7.5a	711.41a	5.05a	3.09a	1.71a	50.04d
500	30.67b	33.99b	4.0b	352.33b	2.85b	0.99b	1.45ab	51.41c
1000	24.67c	30.65b	3.33b	294.33bc	2.49bc	0.93b	1.42ab	53.21a
1500	20.17d	25.50c	1.83c	233.69cd	2.09cd	0.55c	1.26b	52.62ab
2000	17.83e	22.86c	1.17c	203.21d	1.87d	0.48c	1.16b	51.93bc
F-test	**	**	**	**	**	**	*	**
Application frequency								
Double application	29.93a	39.80a	3.00b	410.50a	3.14a	1.39a	1.49a	51.39b
Triple application	27.27b	36.23b	4.13a	307.49b	2.60b	1.02b	1.30a	52.29a
F-test	**	**	**	**	**	**	ns	**
Daminozide concentration x Application frequency	ns	ns	ns	ns	ns	ns	ns	ns

**Significant at 1% probability level, *Significant at 5% probability level, ns: Not significant.
Means in each column with the different letters within each factor indicate significant differences at P≤0.05% level according to DMRT.

obtained were as expected as plant height was associated with the process of cell elongation and cell division or in the association of decrease internode length. Daminozide at highest concentration (2000 mg L^{-1}) significantly reduced the length of internode by 70% compared to control (0 mg L^{-1}) (Table 1). Triple application had significantly lower by 9% in internode length compared to double application. The reduction in the internode length has been found have a positive correlation with plant height ($r=0.95$, $P<0.01$). Application of daminozide may also provide the control of main stem internodes elongation up to the end of the production cycle, keeping retardants at growth inhibiting levels as reported by Sachs and Hackett (1972). In term of flower numbers, increasing the daminozide concentration reduced the number of flowers (Table 1). *Ruellia brittoniana* plants treated with 2000 mg L^{-1} gave the lowest number of flowers. This result is similar to the behaviour of *Eucomis autumnalis* treated with daminozide by drenching method which produced significantly fewer flowers per inflorescence compared with the control (Salachna & Zawadzinska, 2017). Carlson *et al*. (2015) had demonstrated that retardants slightly delayed flowering in the pineapple lily 'Leia'. The significant effect ($P<0.01$) of daminozide given at different concentration and application frequency on leaf area was observed but there was no significant interaction between the factors (Table 1). The result indicated that daminozide reduced the leaf area of plant by 71% when the concentration of daminozide concentrations increasing from 0 to 2000 mg L^{-1}. Triple application of the growth retardant had significantly lower leaf area by 25% compared to double application. This was probably due to the reduction in leaf area retards the process of cell elongation and cell division caused by daminozide treatments. Results obtained in this study showed that leaf, stem and root dry weights were significantly reduced by daminozide treatments which in line with the results reported by Hashemabadi *et al.* (2012) (Table 1 and Figure 1). The reduction of leaf, stem and root dry weights following daminozide application is related to the reduced of plant size as reflected in reduction plant height, internode length and leaf area, as suggested by the coefficient of correlation. Daminozide treated leaves of plants appeared higher chlorophyll content than the control plants, and these values were significantly affected ($P<0.01$) by daminozide concentration (Table 1). Increasing the daminozide concentrations significantly increased the leaf chlorophyll content. Chlorophyll content for 1000 mg L^{-1} treatments was 9% increase compared to 0 mg/L treatments.

Net photosynthesis, stomatal conductance and transpiration

There was a significant effect of daminozide concentrations ($P<0.01$) and application frequencies ($P<0.05$) on leaf photosynthesis (Table 2). The effect of application frequency can be observed at every daminozide concentration (500, 1000, 1500 and 2000 mg L^{-1}). The decrease of photosynthetic rate at 500, 1000, 1500 and 2000 mg L^{-1} with triple

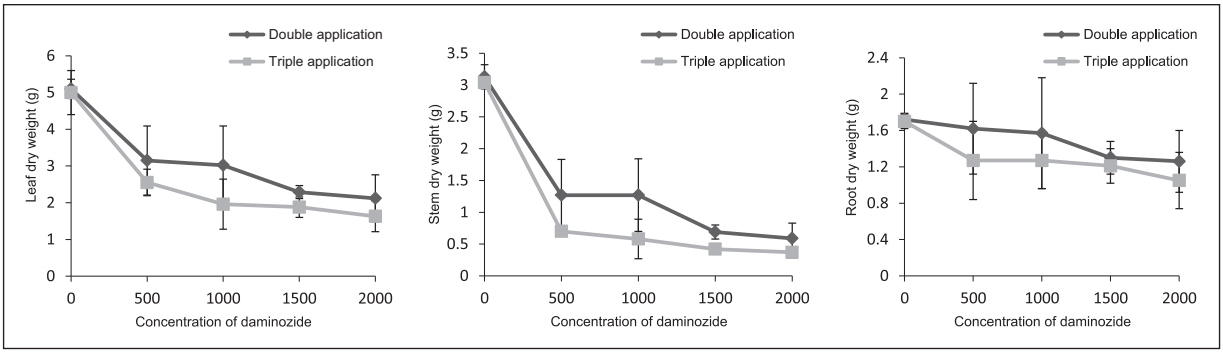

Fig. 1. Effects of daminozide concentrations and application frequency on leaf, stem and root dry weight of *Ruellia brittoniana.* Vertical bars represent standard error of means.

Table 2. Effects of the daminozide concentration and application frequency on photosynthesis, stomatal conductance and transpiration

Daminozide treatment	Photosynthesis (μmol $CO^2m^{-2}s^{-1}$)	Stomatal conductance (mol m^{-2} s^{-1})	Transpiration (mmol $m^{-2}s^{-1}$)
Daminozide concentration (mg L^{-1})			
Control	16.89a	0.42a	4.94a
500	14.99ab	0.37a	4.62ab
1000	13.38bc	0.33a	4.24bc
1500	13.14bc	0.32a	3.96c
2000	12.65c	0.65a	4.08bc
F-test	**	ns	*
Application frequency			
Double application	14.92a	0.51a	4.63a
Triple application	13.50b	0.33a	4.11b
F-test	*	ns	**
Daminozide concentration x Application frequency	ns	ns	ns

**Significant at 1% probability level, *Significant at 5% probability level, ns: Not significant.
Means in each column with the different letters within each factor indicate significant differences at P≤0.05% level according to DMRT.

application were 22%, 25%, 26% and 28%, respectively, while the corresponding decrease of photosynthesis of double application were 1%, 16%, 20% and 22%, compared to the control. Control plants had a higher photosynthetic of 16.92 μmol CO_2 $m^{-2}s^{-1}$ compared to daminozide treated plants. Overall, triple application of daminozide had significantly lower by 10% on leaf photosynthesis compared to double application. Different levels of daminozide and application frequency and their interaction did not affect stomatal conductance. The transpiration rate of leaves of plants was significantly affected by daminozide concentrations ($P<0.05$) and application frequencies ($P<0.01$) (Table 2). There was no significant interaction between daminozide concentrations and application frequencies on leaf transpiration. Moreover, the highest reduction of transpiration occurred at 500 mg L^{-1} daminozide with triple application frequency, which was 18%, while the reduction in transpiration under double frequency was 2% compared to control. The decreasing rate of transpiration could be attributable to thicker cuticles of the daminozide-treated leaves (Jaleel *et al.,* 2007).

CONCLUSION

Application of daminozide at a concentration of 500 mg L^{-1} reduced the plant height, internode length, leaf area, dry weight of leaves, stem and root, and photosynthesis rate but increased the relative chlorophyll contents. In addition, the increasing application frequency by triple application of daminozide significantly reduced the plant height, photosynthesis, transpiration rate and relative

chlorophyll contents compared to double application. Therefore, application of daminozide at 500 mg L^{-1} given in triple application should be sufficient to give a positive impact in producing short, compact plants with less inhibiting effects on flowering capability of the plants.

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