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Morphological and Physiological Response of *Syzygium myrtifolium* (Roxb.) Walp. to Paclobutrazol

(Respons Morfologi dan Fisiologi *Syzygium myrtifolium* (Roxb.) Walp. Terhadap Paklobutrazol)

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ABSTRACT

Syzygium myrtifolium *(Roxb.) Walp. is a popular landscape plant in Malaysia. This species is always planted as hedge plant and topiary. However, it requires frequent trimming to maintain its form and aesthetic functions as it grows vigorously. Conventional trimming technique by using trimming shear or trimming machine is currently practiced in landscape maintenance which is time consuming, labour intensive and costly. Therefore, a study was conducted to investigate the effects of a plant growth retardant, paclobutrazol, on the growth and physiological response of* S. myrtifolium. *The plants were planted in polyethylene bags sized 33 cm in height × 27 cm in diameter and filled with soil, organic matter and sand (3:2:1). Paclobutrazol at rates of 0, 1.25, 2.50 and 3.75 g L^{-1} were soil drenched at 30 days after the plants were trimmed and new shoots were produced. Paclobutrazol significantly reduced plant height and leaf area but increased the leaf area index. Reduced photosynthetic rates were recorded with the treated plants as compared to the control plants. In addition, paclobutrazol at 3.75 g L^{-1} significantly reduced the transpiration rate as compared with the control plants and plants treated with 1.25 g L^{-1} of paclobutrazol. However, stomatal conductance was not affected significantly by paclobutrazol. The growth inhibition effect of paclobutrazol reduced the trimming activity in* S. myrtifolium. *Chlorophyll fluorescence measurement indicated that physiological processes in the paclobutrazol-treated plants were not affected. Paclobutrazol is concluded to offer a better approach in maintaining the growth of* S. myrtifolium.

Keywords: Chlorophyll fluorescence; growth inhibition; landscape maintenance; plant growth retardant

ABSTRAK

Syzygium myrtifolium *(Roxb.) Walp. merupakan sejenis pokok landskap yang popular di Malaysia. Pokok ini sering ditanam sebagai pokok pagaran dan topiari. Walau bagaimanapun, pokok ini perlu kerap dipangkas kerana pertumbuhannya yang cepat. Teknik pemangkasan menggunakan gunting atau mesin pemangkas biasa diamalkan dan memerlukan jangka masa yang lama, tenaga kerja yang banyak serta kos yang tinggi. Oleh itu, satu kajian telah dijalankan bagi mengenal pasti kesan perencat pertumbuhan pokok, paklobutrazol terhadap pertumbuhan dan respon fisiologi pokok* S. myrtifolium. *Pokok ditanam di dalam beg politena yang berukuran 33 cm tinggi dan bergaris pusat 27 cm serta diisikan dengan campuran tanah, bahan organik dan pasir (3:2:1). Paklobutrazol dengan kadar 0, 1.25, 2.50 dan 3.75 g L^{-1} disiram ke media penanaman selepas 30 hari pokok-pokok itu dipangkas dan mengeluarkan daun-daun yang baru. Paklobutrazol didapati mengurangkan tinggi pokok dan keluasan daun, tetapi meningkatkan indeks luas daun berbanding pokok kawalan. Kadar fotosintesis yang rendah juga direkodkan bagi pokok yang dirawat dengan paklobutrazol. Paklobutrazol dengan kadar 3.75 g L^{-1} menurunkan kadar transpirasi berbanding pokok yang dirawat dengan kadar 1.25 g L^{-1} paklobutrazol dan juga pokok kawalan. Walau bagaimanapun, konduksi stomata tidak terjejas oleh paklobutrazol. Kesan perencatan pertumbuhan pokok oleh paklobutrazol ini dapat mengurangkan aktiviti pemangkasan bagi spesies* S. myrtifolium. *Pengukuran terhadap floresen klorofil menunjukkan proses fisiologi pokok yang dirawat dengan paklobutrazol tidak terjejas. Paklobutrazol merupakan satu pendekatan yang sesuai dalam menyelenggara pertumbuhan pokok* S. myrtifolium.

Kata kunci: Floresen klorofil; penyelenggaraan landskap; perencatan pertumbuhan; perencat pertumbuhan pokok

INTRODUCTION

Syzygium myrtifolium (Roxb.) Walp. is one of the popular ornamental species in Malaysia. This species belongs to the family of Myrtaceae. *Syzygium myrtifolium* is a medium size tree which grows up to 8 m in height when mature. It is always planted as hedge plant and topiary for its dense branching system and reddish new shoots. However, frequent trimming is required to maintain its shape and the aesthetic functions as it has a vigorous growth. As trimming activity is costly, time consuming and labour intensive, other options of maintaining the growth are needed. Application of plant growth retardants (PGRs), which offers a non-mechanical method of controlling plant growth, is thought to be a better alternative in extending trimming cycles of this species. Paclobutrazol has been found to be the most effective PGR in retarding growth of many plant species (Barrett 2001; Fletcher et al. 2000; Gilley & Fletcher 1997; Mann et al. 1995; Mansuroglu et al. 2009; Mohamed 2010). Paclobutrazol inhibits gibberellins (GAs) biosynthesis in plant (Rademacher 2000; Sponsel 1995) and hence, reduces cell elongation and retards the plant growth.

Measurement of chlorophyll fluorescence provides information about the function of the photosynthetic apparatus and is useful for stress detection in plants (Bolhàr-Nordenkampf & Öquist 1993; Yordanov et al. 1997). It works on the principle that photosynthesis is central to plant biosynthesis and provides an interactive link between the internal metabolism of a plant and the external environment (Baker 1991; Percival & Fraser 2001). The maximum photochemical efficiency of photosystem II (F_v/F_m) is considered to be a measure of PSII effectiveness in the primary photochemical reactions and is correlated with the efficiency of leaf photosynthesis (Buttler 1977, 1978; Gliožeris et al. 2007; Percival & Noviss 2008). This ratio can be a good indicator when plants are subjected to any environmental stresses (Bjorkman & Deming 1987).

This study aimed to determine the effects of paclobutrazol on the growth of *S. myrtifolium,* in an attempt to reduce trimming activities. The effects of paclobutrazol on photosynthetic rate, transpiration rate, stomatal conductance and chlorophyll fluorescence were also measured to determine the plant tolerance to the treatments.

Materials and Methods

STUDY LOCATION

This experiment was conducted at the Forest Research Institute Malaysia (FRIM), Kepong, Selangor, Malaysia (3°10'0" N/101°42'0" E). During the experimentation period, the mean daily temperature ranged from 21.1 to 34.2°C and the annual precipitation was 1914.8 mm.

PREPARATION OF TEST MATERIAL

The plants were purchased from a local nursery and planted in polyethylene bags sized 33 cm in height × 27 cm in diameter. The planting medium was a mixture of top soil, organic matter and sand (3:2:1). The plants were one year old when the experiment commenced. All plants were first trimmed with approximate height of 100 cm. They were then allowed to produce new shoots. From the observation, *S. myrtifolium* took about 30 days to recover from the trimming effect.

APPLICATION OF PACLOBUTRAZOL

Cultar-250 formulation, 250 g a.i. paclobutrazol per litre, was applied on the plants. Four rates of paclobutrazol (0, 1.25, 2.50 and 3.75 g L^{-1}) were studied and each rate was topped up to 1 L with tap water before being applied as soil drenching (each plant received 1 L solution). At the same time, control plants were applied with 1 L of tap water. The experiment was based on a Randomised Complete Block Design (RCBD) with ten replicates. The plants were watered twice daily, in the morning and late afternoon, depending on the weather. Nitrophoska Green, 15:15:15 (NPK Green) was applied monthly at a rate of 5 g per plant. Weeds in the polyethylene bags were controlled manually.

DATA COLLECTION

Morphological Response. Plant height (cm) was measured from the soil surface in the polyethylene bag to the highest shoot tip by using a telescopic height stick. Leaf area (cm^2) was determined with Leaf Area Meter (LI-3100 Nebraska, USA) by measuring the first three fully developed leaves of each plant. Plant Canopy Analyzer, model LAI 2000 (Nebraska, USA) was used to calculate leaf area index (LAI). One measurement was recorded outside the plant canopy and three measurements were recorded under the plant canopy for each LAI calculation. Plant height, leaf area and LAI measurements were done monthly throughout the study period.

Physiological Response. Portable Photosynthetic System, model Li-6400 (LICOR Nebraska, USA), was used to record the stomatal gas exchange. Measurement was made at ambient humidity and the temperature was maintained at 28°C. The measurements were taken when the internal light source was maintained at 1200 μmol photon $m^{-2}s^{-1}$ and the concentration of CO_2 was between 360 and 400 μmol $m^{-2}s^{-1}$. Photosynthetic (μmol $m^{-2}s^{-1}$), transpiration (mmol $m^{-2}s^{-1}$) and stomatal conductance (mol $m^{-2}s^{-1}$) of the plant were recorded from 9.00 a.m. to 11.30 a.m. under full sunlight.

Chlorophyll fluorescence was measured in the field at light saturation (I=100%) by using a Plant Efficient Analyzer (Hansatech Instruments Ltd., Kings Lynn, UK). Measurement was carried out at five-month after the application of paclobutrazol. Data were recorded from 9.00 am to 11.30 a.m. Before measurements were made, each leaf disc was dark adapted for 20 min in a leaf-exclusion clipped to the central region of the leaf surface. The excitation light for fluorescence was then given to the leaf disc at about 1500 μmol $m^{-2}s^{-1}$ for 5 s. Measurements of minimal fluorescence yield (F_0), maximal fluorescence yield (F_m) and variable fluorescence (F_v) were obtained from this procedure. A total of three fully expanded leaves from different shoot terminals of each plant were selected for the physiological measurements.

Statistical Analysis. The data obtained were subjected to ANOVA and means were compared using Tukey Studentized Range (HSD) test (p<0.05) to detect significant difference between treatments.

RESULTS AND DISCUSSION

MORPHOLOGICAL RESPONSE

At one month after the treatments, significant differences in plant growth were observed between paclobutrazol-treated plants and the control (Table 1). However, there was no significant difference in plant height among the plants treated with different rates of paclobutrazol. At five months after the treatments, the height increment of the control plant was about 21.1% while that of the plants treated with 1.25 g L^{-1} and 2.50 g L^{-1} was increased by 2.2% and 2.5%, respectively, whereas plant treated with 3.75 g L^{-1} paclobutrazol only had height increment of 1%. The treated plants were found to have more compact crown shape.

Paclobutrazol resulted in smaller leaves as compared with the control. Differences in leaf area became apparent as early as one month after the application of paclobutrazol (Table 2). The difference was also noted between the plant treated with 1.25 g L^{-1} and 3.75 g L^{-1} paclobutrazol at the second month after the application. At fifth month after the treatment, the leaf area of plants treated with 1.25 g L^{-1}, 2.50 g L^{-1} and 3.75 g L^{-1} was reduced by 54%, 56% and 59%, respectively, suggesting that as the paclobutrazol rates increased, the leaf area reduced.

There was no significant difference in terms of LAI for the first four months after the application of paclobutrazol (Table 3). At five months after the treatments, LAI of the control plant was only increased by about 15.1%, whereas those of the plants treated with 1.25 g L^{-1}, 2.50 g L^{-1} and 3.75 g L^{-1} were increased by 27.2%, 35% and 35.3%, respectively. Application rates of 2.50 and 3.75 g L^{-1} gave higher LAI values as compared with the control. However, all the paclobutrazol-treated plants under study were not significantly different in LAI.

TABLE 1. Effects of paclobutrazol on plant height in *S. myrtifolium*

Paclobutrazol (g L^{-1})	Plant height (cm)					
	Month after application					
	0	1	2	3	4	5
0 (control)	99.00a	110.75a	116.50a	119.00a	121.00a	125.50a
1.25	100.25a	101.50b	102.00b	102.25b	102.50b	102.50b
2.50	100.00a	102.00b	102.50b	102.50b	102.50b	102.50b
3.75	99.25a	100.25b	100.50b	100.50b	100.50b	100.50b

Means followed by the same letter(s) within column do not differ by Tukey's Studentized Range Test at $p<0.05$

TABLE 2. Effects of paclobutrazol on leaf area in *S. myrtifolium*

Paclobutrazol (g L^{-1})	Leaf area (cm^2)					
	Month after application					
	0	1	2	3	4	5
0 (control)	11.43a	11.06a	10.78a	11.09a	10.68a	11.18a
1.25	11.14a	8.81b	7.66b	6.50b	5.65b	5.18b
2.50	11.14a	8.14b	6.86c	6.02bc	5.42bc	4.85bc
3.75	11.02a	7.87b	6.64c	5.61c	4.90c	4.47c

Means followed by the same letter(s) within column do not differ by Tukey's Studentized Range Test at $p<0.05$

TABLE 3. Effects of paclobutrazol on leaf area index (LAI) in *S. myrtifolium*

Paclobutrazol (g L^{-1})	Leaf area index (LAI)					
	Month after application					
	0	1	2	3	4	5
0	2.20a	2.30a	2.73a	3.01a	2.81a	2.59a
1.25	2.30a	2.55a	2.65a	2.81a	2.91a	3.16ab
2.50	2.19a	2.34a	2.72a	3.08a	3.20a	3.37b
3.75	2.18a	2.41a	2.46a	2.91a	3.18a	3.37b

Means followed by the same letter(s) within column do not differ by Tukey's Studentized Range Test at $p<0.05$

Paclobutrazol is widely used as a retardant for controlling the vegetative growth of a wide range of angiosperm (De Jong & Doyle 1984; Mansuroglu et al. 2009; Mohamed 2010; Quinlan & Richardson 1984; Sterret 1985). It inhibits gibberellins biosynthesis in plant (Fletcher et al. 2000; Rademacher 2000). According to Rademacher (2000), proper use of paclobutrazol can restrict plant growth without side effects. Hamid and Williams (1997) stated that paclobutrazol effectively reduced the growth of *Swainsona formosa*. The same results were also obtained on *Ficus microcarpa* (Ahmad Nazarudin et al. 2003), *Veitchia merrilli* and *Syagras romanzoffiana* (Hensley & Yogi 1996). This evidence was also clear in *S. myrtifolium* in the current study. In this study, the height of the control plant at five months after the treatments affected its crown shape and it required trimming in order to restore its previous crown form while the crown shape of the treated plants were maintained with higher LAI. This result showed that the vegetative growth of the treated plant was effectively controlled by paclobutrazol. The difference in height was probably due to the inhibition effect of paclobutrazol on the cell elongation and hence, retarding the internodes length. The most noticeable effect of paclobutrazol is internodes compression resulting in compact and short plants (Berova & Zlatev 2000; Yeshitela et al. 2004). A similar effect of paclobutrazol was also observed in the leaf of the treated plants, which these leaves were found smaller than that of the control. This could be due to the inhibition effect of paclobutrazol on the cell elongation in the leaf. Tonkinson et al. (1995) noted that triazole decreased the size of wheat leaves by the reduction of cell length rather than cell number. Nevertheless, there was no abnormal leaf formation observed which could decline the aesthetic value of *S. myrtifolium* after the application of paclobutrazol. On the contrary, the inhibition effects of paclobutrazol seemed to enhance the crown compactness in plant expressed as higher LAI value in the treated plants as compared with the control. The low LAI in the control plant was due to the irregular crown shape which resulted in greater land coverage. In other words, the growth of the control plant was more vigorous as compared with the treated plant. This result suggested that the application of paclobutrazol enhanced the crown compactness of *S. myrtifolium*.

PHYSIOLOGICAL RESPONSE

At three-month after the application of paclobutrazol, the treated plants showed reduced photosynthetic rate as compared with the control. However, no significant difference in the photosynthetic rate was detected among the plants treated with different rate of paclobutrazol (Table 4). Paclobutrazol at rates of 3.75 g L^{-1} had lower transpiration rate as compared with the control. Reduced transpiration rate was also recorded between the plants treated with 1.25 g L^{-1} and 3.75 g L^{-1} paclobutrazol (Table 4). However, application of paclobutrazol at rates up to 3.75 g L^{-1} did not affect the stomatal conductance in plants (Table 4). The chlorophyll fluorescence values were also not significantly different among all the treatments (Table 5).

Photosynthetic rate and transpiration rate were reduced after treatment with paclobutrazol. Davis et al. (1988) reported that photosynthetic rate decreased as a result of inhibition of leaf expansion. Leaf is an important plant part as it contains mesophyll cells that are specialized as photosynthetic tissues. Gaussoin et al. (1997) also reported that plants treated with plant growth retardants often had a moderate restraining effect on carbon dioxide exchange rate, thus possibly reducing the photosynthetic rate.

TABLE 4. Photosynthetic rate, transpiration rate and stomatal conductance of *S. myrtifolium* at three months after treatment with paclobutrazol

Paclobutrazol (g L^{-1})	Photosynthetic rate (µmol $m^{-2}s^{-1}$)	Transpiration rate (mmol $m^{-2}s^{-1}$)	Stomatal conductance (mol $m^{-2}s^{-1}$)
0	6.93a	1.78a	0.13a
1.25	5.04b	1.77a	0.13a
2.50	4.27b	1.61ab	0.13a
3.75	3.70b	1.43b	0.11a

Means followed by the same letter(s) within column do not differ by Tukey's Studentized Range Test at $p<0.05$

TABLE 5. Chlorophyll fluorescence parameters of *S. myrtifolium* at five-month after treated with paclobutrazol

Paclobutrazol (g L^{-1})	F_0	F_m	F_v	F_v/F_m	F_m/F_0
0 (control)	111.00	675.50	564.50	0.84	6.18
1.25	72.25	437.75	365.50	0.84	6.08
2.50	82.25	495.25	413.00	0.83	6.04
3.75	87.50	502.50	415.00	0.83	5.75

ANOVA showed no significant difference at $p<0.05$

The reduction in photosynthetic rate would also imply reduction in the transpiration rate as both processes are associated with the opening and closing of stomata (Salisbury & Ross 1992). The decline in transpiration rate would then reduce the percentage of water released through stomata. Olsen and Andersen (1995) reported that reduction in transpiration rate would protect the plant against abiotic stress due to water restriction or drought period. In addition, paclobutrazol enhanced stress tolerance of plants by increasing the xylem pressure potential of the treated plants and thereby enhancing plant moisture status during drought period. According to Abod and Jeng (1993), the reduction in photosynthetic rate and transpiration rate was influenced by the stomatal activity and leaf area. However, in this study, stomatal activity was not the main factor in the reduction of photosynthetic and transpiration rate. The possible reason could be the reduction of leaf area, which further contributed to the reduction of the total leaf surface that absorbs the sunlight.

All plants showed optimal value of F_v/F_m of 0.83. Bjorkman and Deming (1987) and Johnson et al. (1993) reported that the physiological processes in plant were well performed if the F_v/F_m is above 0.8. Values lower than this is normally observed if the plant has been exposed to some kind of biotic or abiotic stress factor which reduced the capacity for photochemical quenching of energy within photosystem II. This result showed that the treated plants were not under stress as the ratio of maximal and minimal fluorescence yield (F_m/F_0) were above 3.0. According to Govindjee et al. (1981), values above 3.0 indicated that the water content in the plant was appropriate for the plants to perform its physiological processes. These results showed that *S. myrtifolium* was able to tolerate with the under study application rates of paclobutrazol.

CONCLUSION

The results obtained from this study showed that paclobutrazol had the ability to control the plant height, reducing the trimming activities of *S. myrtifolium*. It also reduced the leaf area but no abnormal leaf formation, which could affect the landscape aesthetic was observed. The species was well adapted to the treatments based on its physiological response. Therefore, paclobutrazol can be an optional procedure in maintaining vigorous landscape plants under tropical condition.

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