Sains Malaysiana 44(4)(2015): 483–489

Growth and Anatomical Responses in *Xanthostemon chrysanthus* as Influenced by Paclobutrazol and Potassium Nitrate

(Tindak Balas Tumbesaran dan Anatomi *Xanthostemon chrysanthus* yang Dipengaruhi oleh Paklobutrazol dan Kalium Nitrat)

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ABSTRACT

A study was conducted to determine the effects of a plant growth regulator (paclobutrazol, PBZ) and commercial fertilizer (Krista-K Plus) as a source of potassium nitrate (KNO_3) on the growth of Xanthostemon chrysantus. *It was also attempted to investigate the anatomical changes in the leaf and stem after the treatment. Nine treatments, i.e. control (no PBZ and Krista-K Plus application), 0 PBZ gL^{-1} + 100 g Krista-K Plus, 0 PBZ gL^{-1} + 200 g Krista-K Plus, 0.125 PBZ gL^{-1} + 0 g Krista-K Plus, 0.125 PBZ gL^{-1} + 100 g Krista-K Plus, 0.125 PBZ gL^{-1} + 200 g Krista-K Plus, 0.25 PBZ gL^{-1} + 0 g Krista-K Plus, 0.25 PBZ gL^{-1} + 100 g Krista-K Plus and 0.25 PBZ gL^{-1} + 200 g Krista-K Plus, were tested. PBZ was soil drenched at the commencement of the study while Krista-K Plus was applied at three-month intervals. Plant growth performances such as tree height, diameter at breast height, canopy diameter and leaf area were recorded monthly throughout the study period. Stem and leaf samples were collected before the application of treatments and after six months of treatments for anatomical observation by using electron microscope. Plant height, diameter at breast height, crown diameter and leaf area were significantly reduced with the application of PBZ. Palisade parenchyma thickness was increased by 33.83% with 0.25 PBZ gL^{-1} + 200 g Krista-K Plus, while only 2.44% increment recorded in the control tree. Xylem thickness in the stem was reduced by 21.81% after treated with the highest dosage of PBZ, while the control tree only had 1.78% increment. Spongy parenchyma thickness in the leaf was unaffected. However, palisade parenchyma was found the thickest after combined treatment with 0.25 PBZ gL^{-1} + 200 g Krista-K Plus. Micrograph images of the cross-section of leaf lamina and stem showed that the cells were tightly arranged in response to the application of PBZ.*

Keywords: Growth inhibition; ornamental plant; plant anatomy; plant growth regulator; scanning electron microscope

ABSTRAK

Suatu kajian telah dijalankan untuk mengenal pasti kesan pengawal atur pertumbuhan pokok (paklobutrazol, PBZ) dan baja dagangan (Krista-K Plus) sebagai punca kalium nitrat (KNO_3) terhadap tumbesaran pokok Xanthostemon chrysanthus. *Kajian ini juga bertujuan untuk mengkaji perubahan anatomi di dalam daun dan batang pokok selepas rawatan. Sembilan rawatan, iaitu kawalan (tanpa PBZ dan Krista-K Plus), 0 PBZ gL^{-1} + 100 g Krista-K Plus, 0 PBZ gL^{-1} + 200 g Krista-K Plus, 0.125 PBZ gL^{-1} + 0 g Krista-K Plus, 0.125 PBZ gL^{-1} + 100 g Krista-K Plus, 0.125 PBZ gL^{-1} + 200 g Krista-K Plus, 0.25 PBZ gL^{-1} + 0 g Krista-K Plus, 0.25 PBZ gL^{-1} + 100 g Krista-K Plus dan 0.25 PBZ gL^{-1} + 200 g Krista-K Plus, telah diuji. Aplikasi PBZ dilakukan secara siraman tanah pada permulaan kajian, manakala Krista-K Plus diberikan setiap tiga bulan. Tindak balas tumbesaran pokok seperti ketinggian pokok, garis pusat batang, garis pusat silara dan keluasan daun direkodkan setiap bulan sepanjang tempoh kajian. Sampel daun dan batang diambil sebelum rawatan dan selepas enam bulan rawatan untuk menilai perubahan anatominya menggunakan mikroskop elektron imbasan. Ketinggian pokok, garis pusat batang, garis pusat silara dan keluasan daun didapati berkurangan selepas rawatan PBZ. Ketebalan sel palisad parenkima dalam daun didapati meningkat sebanyak 33.83% selepas rawatan 0.25 PBZ gL^{-1} + 200 g Krista-K Plus, manakala hanya 2.44% peningkatan direkodkan pada pokok kawalan. Ketebalan xilem di dalam batang berkurangan sebanyak 21.81% selepas dirawat dengan PBZ pada kepekatan yang paling tinggi, manakala pokok rawatan hanya meningkat sebanyak 1.78%. Ketebalan sel parenkima span di dalam daun pula tidak terjejas. Walau bagaimanapun, sel palisad parenkima didapati paling tebal selepas dirawat dengan 0.25 PBZ gL^{-1} + 200 g Krista-K Plus. Imej mikrograf keratan rentas lamina daun dan batang pokok menunjukkan sel tersebut tersusun dengan padat akibat tindak balas terhadap rawatan PBZ.*

Kata kunci: Anatomi pokok; mikroskop elektron imbasan; pengawal atur pertumbuhan pokok; perencatan pertumbuhan; pokok hiasan

Introduction

Xanthostemon chrysanthus (F. Muell.) Benth. or known as golden penda is a medium-sized tree belonging to the family of Myrtaceae. Owing to its distinctive and bright yellow floral, this species becomes one of the popular ornamental trees in Malaysian cities. Native to tropical northern Australia, New Guinea, Indonesia and the Philippines (Sosef et al. 1998), it is widely planted for beautification of roadsides, urban parks, residential areas and golf clubs. However, the flowering of this species is inconsistent under local climate condition. Previous studies showed that plant growth regulator (PGR) successfully increased the flowering in some species such as *Lantana camara* (Matsoukis et al. 2001), *Lupinus varius* (Karaguzel et al. 2004), *Citrus aurantifolia* (Tripathi & Dhakal 2005) and *Camelina sativa* (Kumar et al. 2012). However, such study has never been reported for perennial species under tropical climate condition. Thus, a study was carried out to determine the effect of PGR, paclobutrazol (PBZ) and potassium nitrate (KNO_3) on the growth and anatomical changes of *X. chrysanthus* before this technique can be applied for flower induction of the urban tree in Malaysian landscapes.

Generally, PGR has several morphological effects on leaves and stems. It reduced leaf area (LA), but increased epicuticular wax, width and leaf thickness (Gao et al. 2011; Gopi et al. 2008). PBZ inhibits gibberellin biosynthesis in plants, reduces cell elongation and retards plant growth (Ahmad Nazarudin et al. 2012; Fletcher et al. 2000; Francescangeli et al. 2007; Mansuroglu et al. 2009; Rademacher 2000). PBZ was reported as the most persistent triazole in controlling the vegetative growth of various plant species. Reduced plant height was recorded in *Bougainvillea glabra* (El-Quesni et al. 2007; Karaguzel & Ortacesme 2002), *Rhododendron catawbiense* (Gent 2004), *Lilium* sp. (Francescangeli et al. 2007), *Consolida orientalis* (Mansuroglu et al. 2009) and *Syzygium myrtifolium* (Ahmad Nazarudin et al. 2012). According to Kishorekumar et al. (2006), the number of cells per unit area in the palisade spongy layers and chloroplast number per cells in the leaves of *Solenostemon rotundifolius* increased by PBZ treatment when compared to control leaves. Tekalign et al. (2005) concluded that the leaves of *Solanum tuberosum* treated with PBZ showed increased epicuticular wax layer, elongated and thicker epidermal as well as the palisade and spongy mesophyll cells. In addition, Bai et al. (2004) stated that the cambial growth of *Liquidambar styraciflua* and *Alnus glutinosa* was reduced following PBZ treatment.

In the present paper, the effects of PBZ and KNO_3 on the growth performance of a landscape tree *X. chrysanthus* were determined. In addition, the anatomy of the leaf and stem of *X. chrysanthus* was also studied under scanning electron microscope (SEM) in the attempt to investigate the changes of tissue structure in the leaf and stem after treatments with PBZ and KNO_3.

Materials and Methods

Study Location and Plant Materials

A study plot was established at Metropolitan Batu Recreational Park, Kuala Lumpur (latitude 3°12’49”N and longitude 101°40’43”E). A total of 81 trees aged about six years grown in the recreational park were used in the study. The average height and the average diameter at breast height (dbh) of these trees were approximately 6 and 12 cm, respectively. During the study period, the mean daily temperature ranged between 24 and 33°C and the annual precipitation was 2266 mm, with approximately 76% relative humidity.

The experiment was arranged in a CRD with nine replicates, i.e. T1 (control), T2 (0 PBZ gL^{-1} + 100 g Krista-K Plus), T3 (0 PBZ gL^{-1} + 200 g Krista-K Plus), T4 (0.125 PBZ gL^{-1} + 0 g Krista-K Plus), T5 (0.125 PBZ gL^{-1} + 100 g Krista-K Plus), T6 (0.125 PBZ gL^{-1} + 200 g Krista-K Plus), T7 (0.25 PBZ gL^{-1} + 0 g Krista-K Plus), T8 (0.25 PBZ gL^{-1} + 100 g Krista-K Plus) and T9 (0.25 PBZ gL^{-1} + 200 g Krista-K Plus). Cultar-250 formulation with 250 g a.i. PBZ per litre was used. PBZ was applied as soil drench (collar drench) at an application volume of 1 L per tree. Control plants were applied with 1 L of plain water. Application of PBZ was carried out once at the commencement of the study. Krista-K Plus (13.7:0:38.4) was applied quarterly using pocket system technique into the soil. The allocated amount of Krista-K Plus was equally applied in four holes for each tree. The holes of 15 cm in depth were dug under the drip-line of the tree canopy. Then, the holes were back-filled with the original soil to prevent runoff.

Data Collection and Analysis

Tree height (m) was recorded by using a LaserAce® Hypsometer, USA. The device was pointed and shot at the base of the tree and then at the shoot tip of the tree. The reading was then showed on the LCD screen. The dbh (cm) was measured at 1.3 m above the ground by using a diameter tape.

Canopy diameter (cm) is the mean of the widest and narrowest parts of the canopy viewed directly on the ground from below canopy. It was measured and recorded by using a measuring tape. The first three fully expanded leaves were randomly collected for leaf area (LA) measurement using a Leaf Area Meter CI 202 (CID. Inc., USA). LA was measured in cm^2. All data were collected on monthly basis for a year (April 2012 to March 2013).

Anatomical study was carried out at the commence of the study and six-month after the treatments. The first five fully expanded leaves from five trees of each treatment were collected for these measurements. Each leaf was cut into approximately 1 cm slices before they were cross sectioned, fixed in fixative, post-fixed in 1% cocodylate buffered osmium tetroxide for 2 h before dehydration through graded series of ethanol (30, 50, 70, 90, 95 and 100%) for 30 min each. The dehydration process using 100% ethanol was repeated twice. They were then subjected to critical point drying for 70 min, mounting on stubs and sputter coating in gold (Zakaria & Razak 1999). The specimens were then viewed under the SEM JSM-5610LV at an acceleration voltage of 15 kV. The same procedure was followed for the stem specimen preparation. The stem sample was obtained from the second internodes of the same plants. The measurements taken were the thickness of the palisade and spongy parenchyma of the leaf. For the stem, xylem thickness was measured. All measurements were recorded in µm.

Data obtained were subjected to ANOVA and the treatment means were then compared using Tukey's Honestly Significant Difference (HSD) test (p<0.05) to detect significant difference between treatments.

Results and Discussion

Growth Response

Tree height of *X. chrysanthus* was similar for the first four months after treatments (Table 1). In August 2012, the height was significantly reduced with T4 and T7 as compared to T1 and T2. At this stage, the height of trees treated with T4 and T7 was 6.5 m, while the control tree (T1) was measured at 7.02 m, showing a difference of 7.4%. At the end of the study period, T4 and T7 had tree height of 6.83 m, whereas the height of T1-treated tree was 7.51 m, giving a difference of 9.1%. Significant reduction in height of trees treated with T4 and T7 as compared to T1 and T2 were continuously observed until the end of the study period. T4 and T7 also recorded significant differences in height as compared to T3 from November 2012 to March 2013. Meanwhile, the tree height was not significantly differed amongst T4, T5, T6, T7, T8 and T9 for the last five months of the study period. These results showed that PBZ inhibited the tree growth. Previous research also reported that PBZ resulted in plant height reduction (Ahmad Nazarudin 2012; Gent 2004; Pinto et al. 2005; Taiz & Zeiger 2006; Williams et al. 2003).

Differences in dbh were not detected for the first four months of the study (Table 2). This parameter became apparent in August 2012, where T2 and T3 resulted in bigger dbh as compared to T4. At this stage, the dbh of T4-treated tree was 13.57 cm, while T2 and T3 increased

TABLE 1. Tree height of *X. chrysanthus* after the application of PBZ and KNO_3 (April 2012 – March 2013)

Trt	Tree height (m)											
	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Jan	Feb	Mar
T1	6.64a	6.73a	6.8a	6.92a	7.02a	7.07a	7.14a	7.22a	7.32a	7.39a	7.45a	7.51ab
T2	6.58a	6.66a	6.77a	6.88a	6.99a	7.04a	7.1a	7.15ab	7.22ab	7.29ab	7.37ab	7.42abc
T3	6.62a	6.7a	6.81a	6.85a	6.9ab	6.95ab	7.01ab	7.12ab	7.2ab	7.3ab	7.4ab	7.57a
T4	6.37a	6.4a	6.44a	6.47a	6.5b	6.55b	6.6b	6.62c	6.67c	6.7c	6.76c	6.83d
T5	6.4a	6.46a	6.54a	6.58a	6.62ab	6.66ab	6.7ab	6.75bc	6.83bc	6.89bc	6.96bc	7.03cd
T6	6.59a	6.23a	6.66a	6.71a	6.75ab	6.83ab	6.95ab	7.02abc	7.08abc	7.14abc	7.19abc	7.25abcd
T7	6.43a	6.45a	6.48a	6.49a	6.51b	6.55b	6.59b	6.63c	6.67c	6.72c	6.78c	6.83d
T8	6.46a	6.5a	6.55a	6.59a	6.65ab	6.69ab	6.73ab	6.78abc	6.87abc	6.91bc	6.98bc	7.03cd
T9	6.48a	6.52a	6.56a	6.61a	6.66ab	6.71ab	6.77ab	6.81abc	6.87abc	9.91bc	6.97bc	7.08bcd

Means followed by the same letter(s) within column do not differ (p<0.05) by Tukey's HSD Test; Trt (treatments)

TABLE 2. The dbh of *X. chrysanthus* after the application of PBZ and KNO_3 (April 2012 – March 2013)

Trt	dbh (cm)											
	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Jan	Feb	Mar
T1	12.35a	12.92a	13.46a	13.93a	14.42ab	14.84abc	15.4abc	15.77abc	16.31ab	16.88ab	17.35abc	18.05bc
T2	12.27a	12.9a	13.92a	14.34a	15.01a	15.65ab	16.13ab	16.62ab	17.16a	17.78a	18.61ab	19.19ab
T3	12.38a	13.01a	13.68a	14.34a	15.01a	15.74a	16.32a	16.98a	17.62a	18.27a	18.89a	19.76a
T4	12.32a	12.56a	12.91a	13.19a	13.57b	14.12c	14.65c	15.08c	15.51b	16.07b	16.49c	16.95c
T5	12.22a	12.62a	13.01a	13.58a	14.03ab	14.59abc	15.16abc	15.83abc	16.28ab	16.78ab	17.26bc	17.84c
T6	12.19a	12.61a	13.1a	13.63a	14.1ab	14.56abc	15.22abc	15.8abc	16.35ab	16.89ab	17.51abc	18.27abc
T7	12.23a	12.61a	12.97a	13.45a	13.85ab	14.25bc	14.78c	15.15c	15.5b	15.93b	16.38c	16.82c
T8	12.64a	12.91a	13.19a	13.52a	13.84ab	14.24bc	14.64c	15.05c	15.48b	15.84b	16.29c	16.87c
T9	12.21a	12.67a	13.04a	13.55a	13.96ab	14.32bc	14.85bc	15.24bc	15.57b	16.02b	16.44c	16.92c

Means followed by the same letter(s) within column do not differ (p<0.05) by Tukey's HSD Test; Trt (treatments) the dbh up to 15.01 cm, giving a difference of about 9.6%. It was also found that T3 and T6 resulted in no significant difference in dbh throughout the study period. It showed that the inhibition effect of 0.125 gL^{-1} PBZ was not obvious with the presence of 200 g Krista-K Plus. In another word, the existence of higher amount of KNO_3 in T6 could probably overcome the effect of lower concentration of PBZ, hence resulting in similar growth of dbh as T3. However, T6 also resulted in no significant differences in dbh as compared to T4, T5, T7, T8 and T9, suggesting that treatment with PBZ alone or PBZ with combination of KNO_3 gave similar growth effects to dbh throughout the study period. Growth of canopy diameter significantly differed in June 2012, July 2012 and March 2013 (Table 3). In June, T2 had 5.21 m in canopy diameter as compared to both T4 and T7, 4.52 and 4.45 m, respectively. Meanwhile, in March 2013 the canopy diameter of T2-treated tree was 9.78 m while T7 was measured at 8.92 m, showing 8.8% difference. It was again observed that there were no significant differences in canopy diameter amongst T4, T5, T6, T7, T8 and T9 with the presence of PBZ.

The differences in LA were also first noted in July 2012, where T2 had the highest LA (39.63 cm^2) and the lowest LA was measured in T6 (33.69 cm^2), showing a difference of 15% (Table 4). The changes in LA observed from August 2012 to February 2013 showed that T1, T2 and T3 had similar effects on the LA. In other word, KNO_3 did not have positive effects on the leaf growth. On the other hand, T4, T5, T6, T7, T8 and T9 had significantly smaller leaves in terms of LA but these PBZ treated trees did not differ significantly in LA, indicating that treatment with PBZ, or combination of PBZ and KNO_3 gave similar effect on leaf expansion of the species. These results showed that PBZ inhibited cell expansion in the leaf. In previous studies, PBZ was also reported to suppress leaf expansion in plants (Ahmad Nazarudin et al. 2007; Sebastian et al. 2002; Yeshitela et al. 2004).

ANATOMICAL STRUCTURE IN LEAF AND STEM

SEM micrograph images of the leaves showed that the palisade parenchyma cells of the PBZ treated trees were tightly packed as compared to those of the control tree

TABLE 3. Canopy diameter of *X. chrysanthus* after the application of PBZ and KNO_3 (April 2012 – March 2013)

Trt	Canopy diameter (m)											
	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Jan	Feb	Mar
T1	3.93a	4.47a	4.97ab	5.42ab	5.86a	6.43a	6.9a	7.53a	7.96a	8.55a	9a	9.35ab
T2	3.92a	4.59a	5.21a	5.59a	5.95a	6.46a	6.91a	7.45a	7.9a	8.6a	9.17a	9.78a
T3	3.85a	4.41a	4.85ab	5.51ab	5.96a	6.39a	6.83a	7.5a	8.03a	8.4a	9.02a	9.63ab
T4	3.82a	4.12a	4.52b	4.88ab	5.29a	5.84a	6.27a	6.79a	7.22a	7.79a	8.37a	9.01ab
T5	3.81a	4.09a	4.58ab	4.99ab	5.49a	5.98a	6.49a	6.94a	7.43a	7.89a	8.48a	9.03ab
T6	3.8a	4.22a	4.68ab	5.12ab	5.55a	6.05a	6.53a	7a	7.49a	8.02a	8.57a	9.14ab
T7	3.81a	4.07a	4.45b	4.83b	5.39a	5.76a	6.29a	6.77a	7.29a	7.8a	8.34a	8.92b
T8	3.81a	4.21a	4.66ab	5.34ab	5.77a	6.28a	6.74a	7.21a	7.71a	8.12a	8.66a	9.08ab
T9	3.81a	4.18a	4.63ab	5.12ab	5.61a	6.12a	6.59a	7.16a	7.56a	8.06a	8.54a	8.94b

Means followed by the same letter(s) within column do not differ (p<0.05) by Tukey's HSD Test; Trt (treatments)

TABLE 4. LA of *X. chrysanthus* after the application of PBZ and KNO_3 (April 2012 – March 2013)

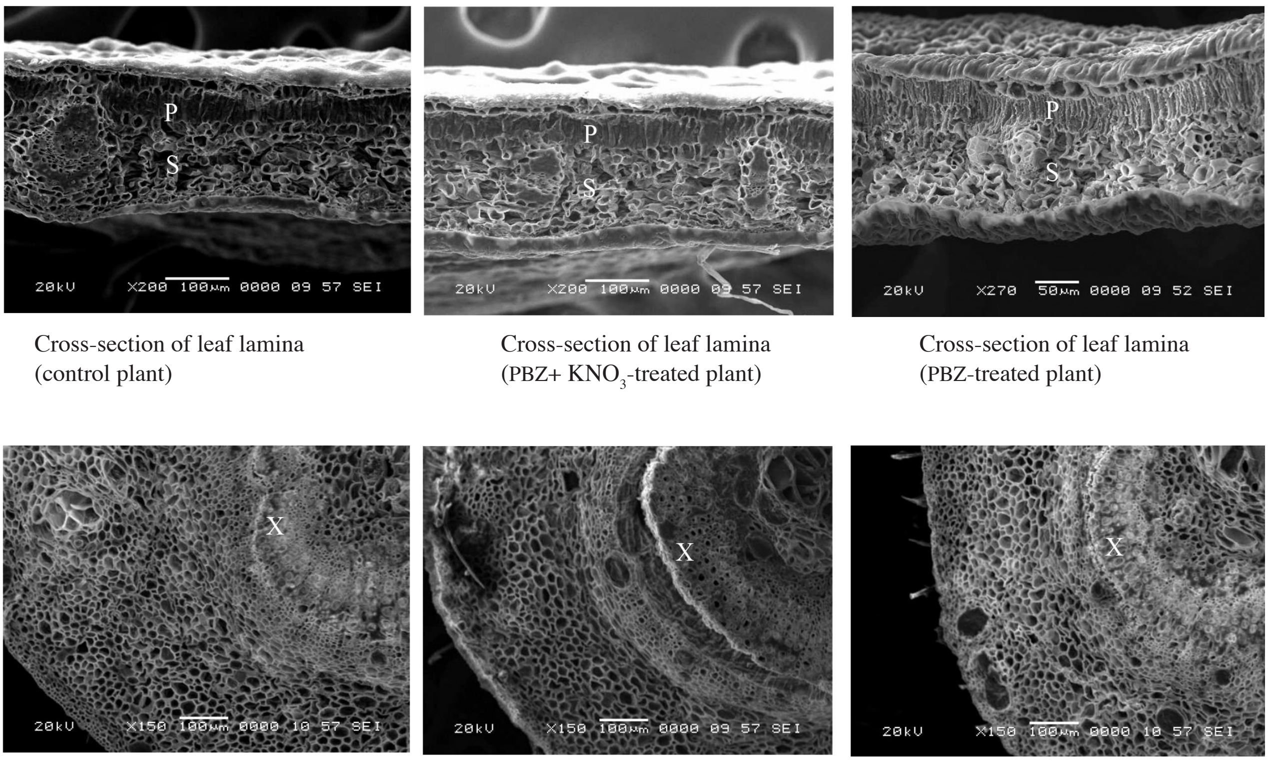
Trt	LA (cm^2)											
	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Jan	Feb	Mar
T1	41.67a	40.65a	40.2a	38.8ab	41.11a	41.97a	43.39a	44.64a	45.52a	46.06a	45.59a	48.45a
T2	40.42a	40.12a	39.74a	39.63a	40.18a	41.02a	42.78a	43.79a	45.01a	45.84a	44.65a	46.29ab
T3	37.22a	36.49a	35.96a	37.33ab	37.7ab	38.26a	39.83a	41.24a	42.43a	43.47a	43.33a	44.85b
T4	39.58a	37.44a	35.73a	34.19ab	33.02b	32.3b	31.46b	30.53b	29.49b	28.37b	27.55b	27.95c
T5	39.08a	39.85a	37.89a	34.99ab	34.11b	33.53b	32.34b	31.58b	30.69b	29.96b	29.19b	30.12c
T6	41.99a	36.73a	34.94a	33.69b	32.9b	32.3b	31.28b	30.43b	29.56b	28.78b	27.91b	28.62c
T7	41.24a	38.92a	36.76a	34.77ab	33.5b	32.79b	31.75b	30.75b	29.83b	28.51b	27.59b	28.02c
T8	39.96a	36.59a	34.66a	34.11ab	33.19b	32.56b	31.47b	30.91b	30.1b	29.29b	28.39b	28.98c
T9	39.48a	36.39a	34.56a	34.05b	33.23b	32.51b	31.68b	30.86b	30.11b	29.2b	28.26b	28.9c

Means followed by the same letter(s) within column do not differ (p<0.05) by Tukey's HSD Test; Trt (treatments)

(Figure 1). This could be due to the decreased leaf size forcing these tissues in such arrangement. The stem cross sections also showed that xylem thickness decreased as a response to these treatments. PBZ also suppressed leaf expansion in other tree species such as *Mangifera indica* (Yeshitela et al. 2004) and *Syzygium campanulatum* (Ahmad Nazarudin et al. 2007). The stem cross sections of *S. campanulatum* showed that xylem thickness decreased as a response to PBZ treatments (Ahmad Nazarudin et al. 2007). Jaleel et al. (2009) also found that PBZ increased leaf thickness in *Catharanthus roseus* by increasing the length of the mesophyll layers.

At the commence of the study and six months after the treatments, palisade parenchyma thickness in the control tree was measured at 50.08 and 51.33 µm (increment of 2.44%), respectively, while T9 significantly increased the thickness of the palisade parenchyma from 49.96 to 75.5 µm (increment of 33.83%) (Table 5). It shows that PBZ combined with KNO_3 increased the palisade parenchyma thickness, resulting in thicker leaf as compared to the untreated leaf. This finding was in line with the report of Gao et al. (2011) which indicated that PBZ increased leaf thickness. Nie et al. (2001) also found that PGRs increased mesophyll density and hence increased the leaf thickness. Increased thickness of the epicuticular wax layer and size of vascular bundles, epidermal, mesophyll and bundle sheath cells were amongst the responses in plants due to PBZ treatment (Ahmad Nazarudin et al. 2007; Gopi et al. 2008; Jenks et al. 2001; Sebastian et al. 2002; Yeshitela et al. 2004). In an ornamental plant, *S. myrtifolium,* reduced leaf area was noted after PBZ treatments, but no abnormal leaf formation which could affect the landscape aesthetic was observed (Ahmad Nazarudin et al. 2012). On the other hand, the spongy parenchyma thickness of *X. chrysanthus* in this study was unaffected.

Existence of PBZ also affected the anatomical structure in the stem of *X. chrysanthus* by reducing the xylem thickness. The xylem thickness was found reduced in T7 (21.81%) but increased by 1.78% as response to T1 (Table 5). A similar effect of PBZ on xylem thickness was also reported in *S. campanulatum* (Ahmad Nazarudin et al. 2007), *L. styraciflua* and *A. glutinosa* (Bai et al. 2004). These results showed that PBZ inhibited the cell elongation in xylem tissue, which was confirmed by the reduction in dbh. However, combined effects of PBZ and KNO_3 (T5, T6, T8 and T9) and KNO_3 alone (T2 and T3) showed thicker xylem layer as compared to single application of PBZ (T4 and T7). This result suggested that K plays important roles in plant growth and development. In *Gossypium hirsutum*, epidermal and mesophyll cells were more turgid, uniform, flaccid, symmetrical and structurally improved and leaf thickness increased with the application of K (Akhtar et al. 2009). Augmented tissue structures in the treated plant increased the water holding capacity which helped



Cross-section of leaf lamina (control plant)

Cross-section of leaf lamina (PBZ+ KNO_3-treated plant)

Cross-section of leaf lamina (PBZ-treated plant)

Cross-section of stem (control plant)

Cross-section of stem (PBZ+ KNO_3-treated plant)

Cross-section of stem (PBZ-treated plant)

FIGURE 1. Micrograph images of the anatomical structures in the leaf and stem of *X. chrysanthus* at six months after the treatments. P (palisade parenchyma); S (spongy parenchyma); X (xylem)

TABLE 5. Anatomical changes in the leaf and stem of *X. chrysanthus*

Trt	Leaf anatomy				Stem anatomy	
	Palisade parenchyma thickness (µm)		Spongy parenchyma thickness (µm)		Xylem thickness (µm)	
	At commence of study	6th month	At commence of study	6th month	At commence of study	6th month
T1	50.08 a	51.33d	124.00a	125.57a	399.87 a	407.13ab
T2	49.88 a	57.21c	123.49a	125.44a	400.12 a	416.60a
T3	50.57 a	59.06c	123.82a	125.21a	399.57 a	417.98a
T4	49.97 a	68.12b	123.60a	125.29a	398.91 a	326.48d
T5	50.57 a	72.00ab	125.77a	124.84a	400.58 a	387.43bc
T6	50.19 a	72.49ab	125.94a	125.86a	399.49 a	384.33bc
T7	49.83 a	68.47b	123.52a	124.35a	398.84 a	311.85d
T8	50.22 a	72.47ab	125.96a	124.68a	400.45 a	374.51c
T9	49.96 a	75.50a	125.95a	124.46a	400.32 a	365.81c

Means followed by the same letter(s) within column do not differ ($p<0.05$) by Tukey's HSD Test; Trt (treatments)

in enhancing their metabolic activities and increased production of photosynthesis like carbohydrates, proteins and their translocation to respective sinks. Moreover, Mengel (2007) attributed that K has a great regulatory role within plant cells and organs such as, activating enzymes, osmosis regulation and photosynthesis and loading and unloading of sugar in phloem. These many advantages of K may complement the plant in terms of growth and development.

CONCLUSION

The existence of PBZ inhibited tree height, dbh, canopy diameter and LA. In addition, PBZ altered the anatomical structure in the stem and leaf of *X. chrysanthus*. Palisade and spongy mesophyll cells in the leaf of the treated plants were tightly arranged as compared to that of the control plants. The palisade parenchyma thickness in the leaf was increased, while xylem thickness in the stem was reduced with PBZ application. Combination of PBZ and KNO_3, however, augmented the palisade parenchyma and xylem thickness as compared to PBZ alone. The combination of PBZ and KNO_3 may be useful in enhancing plant growth and development.

ACKNOWLEDGEMENTS

Thanks are due to the Kuala Lumpur City Hall (DBKL) for site permission. This work was supported by the Ministry of Agriculture and Agro-based Industry Malaysia (05-03-10-SF1030).

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Received: 26 September 2013
Accepted: 14 December 2014