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Pruning impacts on shoot-root-growth, biochemical and physiological changes of Bougainvillea glabra

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Abstract

The effects of different pruning of Bougainvillea glabra on chlorophyll a & b, quantum yield, sugar content, biomass, stomatal conductance, and flowering process factors were studied. Results on frequent pruning of Bougainvillea glabra plants gave the highest quantum yield, chlorophyll a and b, and maximum flower initiation per plant compared to those of non-pruning plants. The sugar content in pruned plants decreased, probably due to the prolonged phase and flowering process, developed into new flowering shoots. Minimum branch fresh weight and low potassium content were observed in the complete pruning plants. The observations also indicated that different pruning treatments modify the root and shoot initiation, which affects the physiological and biochemical content of plants. In the case of the completed pruning, plant maintain a minimum shoot growth rate due to the low availability of potassium, sugar nutrient and lower root growth. Tertiary branch initiation was found to be more effective in the non-pruning condition. The above findings have given a better understanding of the effects of different pruning on the growth and development of the Bougainvillea glabra. Hence, it is suggested that frequent pruning can be used to maintain plant growth and flower shoot initiation.

Keywords: Flowering process; pruning; quantum yield; stomatal conductance

Abbreviations: Higher Fluorescence (Fm), Lower Fluorescence (Fo), Relative Fluorescence (Fv), Quantum Yield (Fv/Fm), Relative Water Content (RWC), Fresh Weight (FW), Dry Weight (DW), Flower Blooming Cycle (FBC), Total soluble sugars (Ts)

Introduction

The genus bougainvillea plant has a wide variety of traits that make it a potential as a new ornamental plant for floriculture, horticulture, pharmaceutical, agriculture and environmental industries on account of large flexibility in different agro climatic regions of the world (Suexia et al., 2009; Simon et al., 2006; Saifuddin et al., 2009a; Saifuddin et al., 2009b; Kent et al., 2007). Considering its vast scope, introduction and alteration of flower color and size are highly desired ornamental plant all over the world. Recently this species is vastly recommended for plantation in heavy industrial area and traffic island to absorb the pollutants and green house gases from the environment (Kulshreshtha et al., 2009; Fatama et al., 2007; Saifuddin et al., 2010). Pruning is common and constructive technique for a variety of ornamental plants under natural exposed sunlight or greenhouse conditions (Sarkka and Erikson, 2003). Generally, the reason of pruning is to observe and control the plant growth to manipulate branching, flower and fruits production in different season or all year round. Many researcher and their studies have underlined the outcome of pruning on flowering and its effects on the subsequent fruit growth and quantity as well as its quality (Calatayud et al., 2007). But in the ornamental flowering plants, pruning usually was focused on initiation of new flowering shoot by creating or increasing the availability of metabolic sinks. Meanwhile, in the non-pruned plants, the less growth of shoots may be caused a short-term reduction of cell activity of older shoots, causing a decline in yield production (Zieslin and Mor, 1981). In addition, in the long term growing stage, the plant cannot improve its nutrient reserves, moderately or totally, and maintain their flower initiation again. Therefore, the level of plant recovery would rely on pruning, especially pruning-position, pruning-height, and the timing of plant phase (Li et al., 2009; Zieslin and Mor, 1981). Coder (1997) suggested that pruning can be done in a way so that lower branches will get sufficient light to utilize its leaves as a potential part too. In addition, pruning is carried out to maintain attractive plant shape, mostly on ornamental plants. This is accomplished through cutting and removing dead, broken, diseased and old branches even over-crowded branches. Though these techniques are more time consuming than chemical thinning, but the results recover flowering, and specially plant health as much as possible (Calatayud et al., 2007; Calatayud et al., 2009). The circulation and gathering of nutrient elements are affected by style of pruning or age of plant. Admasu and Struikb (2000) said that light penetration and allocation are enhanced by shoot pruning. Pruning can modify the light distribution within its canopy and change their photosynthetic capacity and quantum yield of leaves too (Stitt et al., 1990; Hossain et al., 2007). The quantum yield, the ratio of the number of photons emitted to the number of photons absorbed, is the sign of healthy photo system of plant leaf (Calatayud et al., 2009). Angeles et al. (2008) referred that low light intensity was belong to inner leaves before pruning. But after

pruning, inner and external leaves can receive the equivalent light intensity. Other effects on some physiological activities of a plant may be influenced by the age of leaf, leaf position within the canopy and leaf position in different directions of branches (Nabi et al., 2000). In addition, flower buds, sugar content and N, P, and K content in leaf were higher in pruned trees than unpruned peach trees (Hossain and Fusao, 2008). Pruning generally increased stem potential, induced canopy transpiration and improved water status. Therefore, even under dry condition a potential effect of pruning on fruit growth was observed. However, in wet and cloudy years the status may not be limited and carbohydrate supply and demand effects may be more dominant (Lin and Hsu, 2004; Manuel et al., 1998). Higher stem water potential was reported to be beneficial to increase flower number, fruit growth and quality (Roberto and Jonathan, 2000). In addition, leaf photosynthetic rate and water status can vary with potassium level in herbaceous and woody species (Bednarz and Oosterhuis, 1999; Egilla and Davies, 1995). $\dot{\text{CO}}_2$ assimilation rate and leaf respiration efficiency can be affected by potassium deficiency (Onguso et al., 2004). Thus, this experiment was aimed to study the effects of differing pruning on (1) the new shoot and root initiations, (2) the flowering process under exposed sunlight condition and (3) the content of chlorophyll a and b, quantum yield, biomass, sugar and potassium content of Bougainvillea glabra.

Materials and methods

Experimental site and plant materials

The experiment was carried out in the Plant Physiology Garden, Institute of Biological Sciences, University of Malaya, Malaysia. In this experiment, one-year-old (total of plants 12) bougainvillea plants were grown in a small polythene bag having 12 cm in diameter and 20 cm in length. The bags were placed under prevailing conditions (temperature 21-32°C, maximum PAR 2000 μ E m⁻² s⁻¹ and relative humidity of 60-90%). The plants were irrigated twice a day, to avoid water stress condition. The average height of each plant was 50 cm and having 50 cm row to row distance and 50 cm plant to plant distance. The plant consisted (control) of eight primary branches and five secondary branches. The nutrient, N: P: K (12:12:17), at the rate of 5 gm per plant was applied at a 15 day interval.

Pruning treatment

Effects of different pruning treatments were analyzed following a completely randomized design, having three replications per treatment. There were four treatments. The Control plant was not pruned within six months. Partial pruning was done by pruning all branches except one branch. Complete pruning was performed by completely cutting all primary and secondary branches. Finally frequent pruning was carried out by pruning all branches, 4 cm from shoot apex, at 30 days interval throughout the experimental period (Fig. 1).

Quantum yield measurements

Chlorophyll fluorescence was measured by using Plant Efficiency Analyser (Hansatech Instrument Ltd., England). A clip was affixed to leaves and reserved in dark for 30-45 minutes to



Fig 1. Different types of pruning have been executed at the beginning phase which has shown above. Non-pruning or Control plant (T1); Partial pruning (T2); Complete pruning (T3); and Frequent pruning (T4).

sustain dark adaptation. Then, the leaf clip was oriented with the shutter plate. When the light intensity was applied on the leaf, the fluorescence signal was counted for 3 seconds and observed the Quantum yield or Photosynthetic yield. The Higher fluorescence (Fm) and lower fluorescence (Fo) value was taken from the display pad of Plant Efficiency Analyser machine. The relative fluorescence (Fv) was calculated as Fm-Fo. Calculation of Quantum yield or Photo- synthetic yield was determined according to the equation Fv/Fm (Temperature = 28° C, Time range = $10 \,\mu s^{-3}$ sec).

Relative water content (RWC)

The three youngest fully expanded leaves per plant were taken randomly from each treatment. The leaves were weighed immediately to determine the fresh weight (FW). They were then floated in distilled water (in petri dishes) under a fluorescent lamp supplying compensation light intensity for 12 hours (Wealtherly and Slatye, 1962). Next, the leaves were blotted gently with tissue paper and weighed to determine the turgid weight (TW). The leaves were then oven dried (80 °C) to constant dry weight (DW). The RWC was calculated by RWC = [(FW –DW) / (TW – DW)] ×100.

Photosynthetic pigments

The photosynthetic pigments chlorophyll *a* and chlorophyll *b* were estimated after homogenizing 0.5 g of the fresh leaf sample in 80 percent ethanol. The absorbance was measured at 663 and 645 nm and the chlorophyll content was measured using the following formula (Asare-Boamah et al., 1986). Chl *a* (mg g⁻¹) = $[(12.7 \times A663) - (2.6 \times A645)] \times V$ mg/ gm Fresh Weight Chl *b* (mg g⁻¹) = $[(22.9 \times A645) - (4.68 \times A663)] \times V$ mg/ gm Fresh Weight



Fig 2. Bougainvillea plant structure and different type of branches. Number 1, 2 and 3 represents the primary, secondary and tertiary branch, respectively, at the end of experiments.

Estimation of total soluble sugars

In order to estimate the total soluble sugars, one gm of leaf was homogenized in 4 ml of 0.5 N of sodium hydroxide and grand in a mortar with a pestle and then centrifuged at 3,500 g for 20 min at 4 °C. The pellet was discarded and the supernatant was neutralized with 0.5 N Acetic acid. The resulting solution was made up to 40 ml and was stored at 4 °C till use. These extracts were then used in the estimation of total soluble sugars according to the Phenol-Sulphuric method (Dubois et al., 1956). One ml of the leaf extract was placed in a test tube and 1 ml of Phenol [5 % w/v] was added followed by 5 ml of concentrate Sulphuric acid and the contents were mixed and left at room temperature for 10 minutes. Spectrophotometers reading were taken at 490 nm absorbance. The sugar concentration was obtained by referring to the standard glucose graph. The assay for this standard glucose graph was carried out by adding phenol and sulphuric acid to a standard glucose solution with concentrations varying between 0-100 µg ml⁻¹. Total soluble sugars were expressed in mg/100 gm leaf fresh weight.

Stomatal conductance

Stomatal conductance was measured using a portable porometer (Leaf Porometer, Model SC-1, USA). A leaf clamber was attached to one of the leaf and kept in ambient temperature for 10-15 min to maintain sunlight adaptation. A stomatal conductance was measured in 3 replicates from different spots of a single leaf.

Potassium estimation

The most recent fully expanded leaves at the same age and relative position on the plant leaves were taken from each treatment. For potassium estimation, one gram leaf was homogenized in 5 ml distilled water in a motor with a pestle and then centrifuged at 3,500 rpm for 20 min. The 3 to 5 drops of the supernatant liquid were transpired onto the calibrated sensor pad (Cardy Potassium Meter, Model-2400, USA). A sampling paper was also be placed on the sensor and saturated with the liquid. After the value has stabilized (30 to 45 seconds), reading (ppm) was taken from the display pad.

Bract weight, length and plant biomass measurement

The weight of bract recorded at different (1st and 3rd) flower blooming cycles (FBC) and biomass was measured at the end of experiment using the weighing machine (Model-Mettle PJ3000, Japan) and bract length was measured by Vernier scale. Branch initiation was calculated by following formula: Branch Initiation (%) = (FBN – IBN)/FBN × 100.

Where, FBN is final branch number and IBN is initial branch number.

Statistical analysis

Statistical analysis was completed by SPSS software. Significant difference among the treatments was evaluated by one way ANOVA and LSD (p=0.05) was calculated using the error mean squares.

Results and discussion

The results showed that bract length, weight and bract numbers per plant were significantly decreased by complete pruning in the final season (Table 1). Bract length and weight was almost similar in all treatments in first season. In the final season, the highest bract length and weight was observed in the frequent pruning as compared with control plants. It was clear that frequent pruning had a positive effect on bract length and weight compared to nonpruning and other types of pruning. While complete pruning reduced the bract number per plant in both first and final seasons by 40 and 35 %, respectively. The influence of all treatments on branch initiation was observed throughout the experiments. All strategies were able to initiate primary and secondary branch with respect to experimental periods. The most effective strategies to increase 100 % secondary branches per plant were partial and complete pruning (Fig. 2). In the case of secondary branch initiation, lower value was observed in frequent pruning. However, the most effective strategy for increasing primary and secondary branch per plant was in the partial pruning. In this experiment, tertiary brunch initiation was observed only in nonpruning treatment. In contrast, the maximum branch wide was observed in partial pruning and the minimum was observed in complete pruning plants (Table 2). In addition, biomass measurements were taken at the end of this research. The influence of different styles of pruning on biomass was easily observed here and discussed in pruning studies. There has been a little effort to explain biomass responses to pruning on the basis of tree physiology. The results of above treatments showed that partial pruning has a great influence to increase on fresh weight and dry weight of branches. The fresh weight of complete pruned plants was decreased by 60% as a result of the lower branches present, smaller size and lower wide of the plant canopy. Another related cause was that complete pruning stimulated less vegetative growth. The new brunch initiation was particularly delayed and decreased by complete pruning. It might be due to the sudden shock to the plant and that is way, it might take time to initiate new branches (Table 3). The difference in plant shape among the different types of pruned and non-pruned treatments was evaluated (Table 4). It was perceptible that partial pruning improved root length as well as root weight than complete and frequent pruning. Root growth depends on the strategy of shoot distribution or

Table 1. Measurement of ornamental characteristics (bract length, weight and bract number) as affected by different type of pruning. Values are means of 3 measurements \pm SE. Values followed by different alphabets indicate the existence of significant differences according to LSD _{0.05} test. *Flower Blooming Cycle (FBC)

Treatments	Bract (mm)	Length	Bract (gm)	weight	Flower	number/ plant
	1 st FBC*	3 rd FBC	1 st FBC	3 rd FBC	1 st FBC	3 rd FBC
T1	44.33±0.42	41.6±0.32d	$0.53 \pm .006$	0.370±.02d	44.66±2ab	34.66±1.4b
T2	44.66±0.24	43.1±0.26ab	$0.53 \pm .006$	0.438±.02b	36.66±2bc	33.3±2.7bc
T3	44.83±0.14	42.7±0.37bc	$0.53 \pm .007$	0.403±.01bc	26.6±2.3d	22.66±1.7d
T4	44.53±0.35	44.4±0.52a	$0.53 \pm .007$	0.530±.01a	53.0±2.8a	50.33±1.4a

*means the value within column is not significant

Table 2. Different types of branch initiation were affected by pruning. Values are means of 3 measurements \pm SE. Values followed by different alphabets indicate the existence of significant differences according to LSD _{0.05} test.

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Treatment	Primary	Secondary	Tertiary	Branch
	Branch	Branch	Branch	wide
	Initiation	Initiation	Initiation	
T1	52%	80%	100%	45.9±1.30b
T2	91%	100%	0%	55.32±1.80a
T3	100%	100%	0%	28.84±0.90c
T4	38%	75%	0%	23±60±0.50d

pruning tactics or cutting position. Therefore, different types of pruning had shown different root length, though little similar results were observed between non-pruned and partial pruning. The highest reduction in root weight by 61% as a result of complete pruning may be attributed to lower root length or less vegetative growth (Table 3). These findings led to the proposal of root growth based on the remaining shoots or vegetative growth. The photosystem of leaves, measured by quantum yield referred lower values in the non-pruning leaves. Whereas, significant higher values of quantum yield were observed in all kinds of pruning leaves (Fig. 3). In frequent pruning, the quantum value increased by 4% in leaves of flowering branch. Among the pruned plants, the lower values were presented in complete pruning plant leaves. In respect to stomatal conductance, it was clearly observed that the initial conductance assessment was higher in control treatment (Fig. 4). Amongst the pruning treatments, frequent pruning leaves exhibited the highest value of stomatal conductance. The stomatal conductance was gradually increased throughout the observation except those leaves of the control or non-pruned. The difference in stomatal conductance between complete and partial pruning was perhaps associated to the fact that complete pruning reduced the stomata frequency, thus to maintain the physiological coherence, leaf age or due to a slower growth rate. That is why in a vegetative stage, leaf exhibited low stomatal conductance. In addition, Nabi et al. (2000) showed that the stomatal conductance was directly connected to age and position of leaf in a plant. With the increase of leaf age, the stomatal conductance had improved up to a certain value which was differed from plant species to species. In the case of complete and partial pruning, stomatal conductance increased rapidly beyond the second month of observation. The decline in relative water content (RWC) of complete pruning application was due to a lower plant growth and less capability to maintain photosynthesis process or less nutrient supply through root profile. The effect of partial pruning on (RWC) observed little higher than complete pruning. RWC was improved by frequent pruning by 15% (Fig. 5). Therefore, a higher quantum value and higher flower number per branch was observed in this pruning and similar result was proved by other researchers too (Roberto and Jonathan, 2000). The photosynthetic pigment chlorophyll b showed



Fig 3. Determination of quantum yield in all type of pruning plants leaf including control leaves. Values are means of 3 measurements \pm SE. Values followed by different alphabets indicate the existence of significant differences according to LSD 0.05 test.

a significant difference among the pruning treatments. The association of chlorophyll b was considerably higher in plants which underwent in the frequent pruning. Enhanced creation of chlorophyll dye by pruning treatment had before been stated too and it has been recommended that this accumulation was attributed to the increment of cytokinin action (Fig. 6) (Angeles et al., 2008). Similarly, chlorophyll a also showed a significant difference in both pruning and non-pruning treatments. The lowest quantity of chlorophyll was visualized in the control treatment. Hence, it can be referred that the frequent pruning, once per month, was the most favorable rate to keep the maximum stage of chlorophyll in bougainvillea. Thus, this finding agrees with the reports on the enhancement of the photosynthetic pigments

Table 3. Biomass measurement (of branches) at the end of experiments. Values are means of 3 measurements \pm SE. Values followed by different alphabets indicate the existence of significant differences according to LSD _{0.05} test.

Treatments	Total Branch Fresh Weight	Total Branch Dry	Ratio (Fw/Dw)
	(gm)	Weight (gm)	
T1	36.54±1.20a	21.45±0.90a	1.70±0.03
T2	34.65±1.05ab	18.84±0.34b	1.84 ± 0.08
Т3	14.43±1.03d	7.96±0.46d	1.90±0.24
T4	22.35±1.20c	11.72±0.37c	1.91±0.13

through pruning (Angeles et al., 2008). Potassium content was increased by frequent pruning. So frequent pruning could be one of the effective strategies to increase potassium in plant leaf. The potassium content was lower in partial pruning than in the control treatment. But the significant and effective treatment for reducing plant potassium concentration was complete pruning (Fig. 7). At the end of the test, total soluble sugars (Ts) were significantly higher in the non-pruning plants, whereas the contents of Ts were identical in partial and non-pruning plants. The Ts value was 50% higher in non-pruning plants. The minimum total soluble sugar was obtained in complete pruning compared to all pruning treatments and it was about 40% lower in frequent pruning (Fig. 9). In this work, the investigation of some physiological parameters on bougainvillea plants by pruning was exhibited. Many researches about pruning were carried out mainly on physiological parameters such as yield and carbohydrates content of many fruits plant. This research is focused on offering a wide range of physiological mechanism associated to pruning under natural sunlight growing condition of flowering plant. In this current study, it has been discovered that pruning is an effective method to increase quantum yield and chlorophyll (a & b), also to maintain bract length, bract number/plant and relative water content of leaf.

Quantum yield

Lower quantum (Fv/Fm) value was obtained in non-pruning plants. Fv/Fm reflects a responsive sign of plant performance and most plant species showed an optimal values of 0.8 (Calatayud et al., 2002; Johnson, 1993). In this experiment, around 0.88 has been exhibited in leaves of flower shoots in frequent pruning plants (Fig. 3). The value obtained by fluorescence, indicate that pruned plant promoted photosynthetic light accumulation (0.84). This matter was explained by Mediene et al. (2002) who described that photosynthetic rates was enhanced due to a higher radiation by remaining mature leaves or by the canopy.

Total soluble sugar and shoot length

The analysis of total soluble sugar (Ts) in different types of pruning and non-pruning plants was observed. Non-pruning plant leaves showed maximum Ts than pruned plants. It can be cleared in such a way that pruning perhaps fashion a great amount of metabolic trap by expanding new leaves or flower shoots (Fig. 8). Therefore, the reductions of stored carbohydrates of plant parts or shoots were observed due to the translocation of sugar to the new rising flowering shoots. Other assumption, pruning decreases the total number of photosynthesizing area (Fig. 2) and reduces the amount of new carbohydrate resources (Fig. 9) (Li et al., 2009). Therefore, the gathering of carbohydrates guides to both a reserving of photosynthesis and rising of respiration (Mediene et al., 2009; Stitt et al., 2002).



Fig 4. Stomatal conductance value was affected by different pruned treatments. Values are means of 3 measurements \pm SE. Values followed by a line indicates the existence of significant differences according to LSD _{0.05} test.



Fig 5. Relative water content of different treatments in bougainvillea plant leaves. Bars with different alphabets indicate significant difference according to LSD $_{0.05}$ test.

Table 4. Measurement of root biomass at the end of experiment. Values are means of 3 measurements \pm SE. Values followed by different alphabets indicate the existence of significant differences according to LSD _{0.05} test.

1	6	<u> </u>	100	
Treatment	Root Fresh Weight (gm)	Root Dry Weight (gm)	Fw/Dw	Root Length (cm)
T1	7.68±0.53a	4.63±0.37a	1.67±0.19	20.27±0.74a
T2	6.83±0.37ab	4.16±0.28ab	1.66±0.21	19.86±0.79ab
Т3	2.93±0.40cd	1.86±0.20d	1.56±0.06	13.21±0.38cd
T4	3.29±0.35c	2.44±0.17c	1.30±0.13	16.57±0.56c



Fig 6. The effect of different pruning treatments on photosynthetic pigment, chlorophyll a & b value of *Bougainvillea* sp. leaf. Values followed by different alphabets indicate the existence of significant differences according to LSD 0.05 test



Fig 7. Potassium concentration of different treated bougainvillea plants leaf. Values are means of 3 measurements \pm SE. Values followed by different alphabets indicate the existence of significant differences according to LSD _{0.05} test.

Root and shoot growth

With full expansion of the shoot and leaves, the first priority in the shoot is the deposition of carbohydrate behind the new buds. Shoot storage is always filled with carbohydrate from the tip downward. There should be a positive relationship both in shoot and root. Shoot and root obviously act together and distribute resources to make sure the plant success (Coder, 1997). Therefore, pruning plants has received an effort to balance shoot and root mass by controlling photosynthesis and nutrients consumption (Pinkard et al., 1998). Complete pruning has adjusted the mass of roots or shoots through the deficiency of potassium and stomatal conductance. Whereas, partial pruning showed extensive shoot range due to the availability for utilizing the remaining branch leaves in photosynthetic process and nutrient uptake by excessive root (Table 4). In the case of complete pruning treatment, shoot initiation and branch length was lower than in partial pruning and non-pruning. This decrease could have been due to the less root length to up take nutrient (potassium) or the lack of photosynthesis devices (leaf) or vice versa. Therefore, less root and shoot development was observed throughout the experimental. This hypothesis is that the shoot growth is dependent upon root length, water and mineral nutrient uptake systems, while root growth was depending upon the above shoot part or the plant for carbohydrates stored amount (Coder, 1997). Tertiary branch initiation as a result of non pruning may be attributed to high Ts or capability in photosynthesis through existing primary and secondary branches leaves (Table 2). Whereas, tertiary branch was not initiated by the pruned plants due to its need to re-grow and existing less number of shoot that was pruned off at the beginning of experiment.

Stomatal conductance

The effect of pruning on stomatal conductance was clearly significant throughout the experiment. Initially, stomatal conductance was higher in the non-pruned plants then the value was gradually decreased except those leaves of pruning plants. In the matured field grown vines, photosynthesis reached a maximum at 30 days after unfolding and then declined with leaf age (Poni and Intrieri, 1996). It was referred that leaves from the younger branch or middle age had a higher rate of photosynthesis and high stomatal conductance than the leaves of older branches (Nabi et al., 2000; Poni and Intrieri, 1996).

Relative water content, potassium, flower and chlorophyll

Potassium content was higher in frequent pruning plant than the non-pruned plants and potassium deficiency was reduced by complete pruning (Cheng and Fuchigami, 2000). These values might be caused by younger leaf of complete pruning. In addition,



Fig 8. Flower number per branch in different types of pruning.



Fig 9. Total soluble sugars (Ts) in leaves under different pruning treatment. Values are means of 3 measurements \pm SE. Bars with different alphabets indicate significant difference according to LSD _{0.05} test.

RWC was increased in pruning treatments. This might be due to the higher capability to accumulate chlorophyll a and b or photosynthetic pigment cell (Fig. 6) by pruning plants. Therefore, higher chlorophyll allowed more photosynthetic to take place and maintain higher RWC, producing new branches and leaves, allowing the plant to maintain same quality, size and number of its flower. Other researchers also stated similar outcomes which proved that as the water potential increased, the flower numbers, growth and quality also increased (Roberto and Jonathan, 2000).

Flower initiation

It has been revealed that pruning increased the supply of cytokinins from the roots, measured as increased concentration in the remaining above-ground tissue (Avner and Staden, 1983). The increase of hormone levels is probably responsible for stimulating cell division, new shoot formation and ultimately more flower per branch and frequent flower bud initiation. In this experiment, the frequent pruning took short period to induce flower by stimulating continuously cytokinins for bud formation and therefore flower number/branch, bract length, and weight was remain unchanged from the first to final season (Table 1). However, in the case of partial and complete pruning, the number of flower per branch was low in first and final seasons due to the prolonged vegetative stage for shoot initiation and lack of leaves to utilize in photosynthetic process or lack of contribution of cytokinins from root towards shoot (Calatayud et al., 2004).

Conclusion

In a conclusion, the experimental observations indicated that different pruning styles modified the root and shoot initiation, which ultimately affects the physiological and chemical contents of Bougainvillea glabra. In the case of complete pruning, plants maintain a minimum growth rate due to low availability of potassium, sugar nutrient and lower root growth. Flower initiations were found to be dependent on frequent pruning, high quantum yield and the availability of chlorophyll content. Tertiary branch initiation was found to be dependent on non-pruning condition. Therefore, it can be concluded that the frequent pruning is useful to induce flower and to maintain the growth of Bougainvillea glabra, which can be vastly used for landscaping and environmental beautification purposes. As a recommendation, for the further research, molecular technique is to be studied to genetically manage flowering process of Bougainvillea glabra flower.

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References

- Admasu T, Struikb PC (2000) Influence of repetitive transplanting and leaf pruning on dry matter and food production of enset (Ensete ventricosum Welw. (Cheesman)). Field Crops Research 68:61-74.
- Angeles C, Dolors R, Elisa G, Pedro FM (2008) Light acclimation in rose (Rosa hybrida cv. Grand Gala) leaves after pruning: Effects on chlorophyll a fluorescence, nitrate reductase, ammonium and carbohydrates. Scientia Horticulturae 111:152– 159.
- Asare-Boamah NK, Hofstra G, Fletcher RA, Dumbroff EB (1986) Triadimefon protects bean plants from water stress through its effects on ABA. Plant Cell Physiology 27:383-390.
- Avner C, Staden (1983) Role of roots in regulating the growth and cytokinin content in leaves. Plant Physiol 73:76-78.

- Bednarz CW, Oosterhuis DM (1999) Physiological changes associated with potassium deficiency in cotton. J Plant Nutr 22:303–313.
- Calatayud A, Iglesias DJ, Talon M, Barreno E (2004) Response of spinach leaves (Spinacea oleracea L.) to ozone measured by gas exchange, chlorophyll *a* fluorescence, antioxidant system, and lipid peroxidation. Photosynthetica 42:23–29.
- Calatayud A, Ramyrez JW, Iglesias DJ, Barreno E (2002) Effects of ozone on photosynthetic CO_2 exchange, chlorophyll *a* fluorescence and antioxidant systems in lettuce leaves. Physio Plant 116:308–316.
- Calatayud A, Roca D, Gorbe E, Martynez PF (2007) Light acclimation in rose (Rosa hybrida cv. Grand Gala) leaves after pruning: effects on chlorophyll a fluorescence, nitrate reductase, ammonium and carbohydrates. Scientia Horticulture 111:152–159.
- Calatayud A, Roca D, Gorbe E, Martynez PF (2008) Physiological effects of pruning in rose plants cv. Grand Gala. Scientia Horticultural 116:73–79.
- Cheng L, Fuchigami LH (2000) CO₂ assimilation in relation to nitrogen in apple leaves. *J. Hortic.* Sci Biotechnol 75:383–387.
- Coder KD (1997) Crown pruning effect on roots. [Online]. www.http://warnell.forestry.uga.edu/warnell/service/library/inde x.php3?docID=146
- Dubois M, Giles KA, Hamilton JK, Rebers PA, Smith F (1956) Colorimetric method for determination of sugars and related substances. Anal Chemistry 28:350-356.
- Egilla JN, Davies JFT (1995) Response of Hibiscus rosa-sinensis L. to varying levels of potassium fertilization: growth, gas exchange and mineral concentration. J Plant Nutr 18:1765–1783.
- Fatama EMQ, Magda MK, Mona HM (2007) Some studies on the effect of paclobutrazol on the growth and chemical composition of *Bougainvillea glabra* L. at nubaria. American Eurasian J Agric and Environ Sci 2:552-558.
- Ferree OC, Clayton KAG, Bishop B (1993) Influence of orchard management system on canopy composition, light distribution, net photosynthesis and transpiration of apple trees. J Hortic Sci 68:377-392.
- Hossain ABMS, Fusao M (2008) Determination of abscisic acid hormone (ABA), mineral content, and distribution pattern of 13C photoassimilates in bark-ringed young peach trees. Moj Int J Sci Tech 2:274-284.
- Hossain ABMS, Amru NB, Normaniza O (2007) Postharvest Quality, Vase Life and Photosynthetic Yield (Chlorophyll Fluorescence) of Bougainvillea Flower by Applying Ethanol. Australian Journal of Basic and Applied Sciences 1:733-740.
- Johnson GN, Young-Scholes AJD, Horton P (1993) The dissipation of excess excitation energy in British plant species. Plant Cell Environ 16:673–679.
- Kent DK, James MC, John G (2007) Bougainvillea, Ornamentals and Flowers. [Online] http://www.ctahr.hawaii.edu/oc/freepubs/pdf/OF-38.pdf".
- Kulshreshtha K, Rai A, Mohanty CS, Roy RK, Sharma SC (2009) Particulate Pollution Mitigating Ability of Some Plant Species. Int J Environ Res 3:137-142.
- Li KT, Lakso AN. Piccioni R, Robinson T (2009) Summer pruning reduces whole-canopy carbon fixation and transpiration in apple trees. J Hort Sci Biotech 78:749–754.
- Lin MJ, Hsu BD (2004) Photosynthetic plasticity of Phalaenopsis in response to different light environments. J Plant Physiol 161:1259–1268.

- Manuel R, Carmen DS, Roberto R (1998) Free polyamine content in leaves and buds of hazelnut (Corylus avellana L. cv. Negret) trees subjected to repeated severe pruning. Scientia Horticulturae 76:115-121.
- Mediene S, Jordan MO, Pages L, Lebot J, Adamowicz S (2002) The influence of severe shoot pruning on growth, carbon and nitrogen status in young peach trees (Prunus persica). Tree Physiol 22:1289–1296.
- Nabi G, Trought MC, Noor R, Samad A (2000) To study stomatal conductance at different leaf positions and xylem flow rate at different depths in the apple branch. Pakistan Journal of Biological Sciences 3:1634-1636.
- Onguso JM, Mizutani F, Hossain ABMS (2004) Effects of partial ringing and heating of trunk on shoot growth and fruit quality of peach trees. Bot Bull Acad Sin 45:301-306.
- Pinkard EA, Beadle CL, Davidson NJ, Battaglia M (1998) Photosynthetic responses of *Eucalyptus nitens* (Deane and Maiden) Maiden to green pruning. Trees - Structure and Function 12:119-129.
- Poni S, Intrieri C (1996) Physiology of grape leaf ageing as related to improved canopy management and grape quality. Proceedings 9th Australian Wine Industry Technical Conference, Winetitles, Adelaide, pp. 113-122.
- Roberto NE, Jonathan HC (2000) Selective pruning and crop removal increase early-season fruit production of carambola (Averrhoa carambola L.). Scientia Horticulturae 86:115-126.
- Saifuddin M, Hossain ABMS, Normaniza O (2010) Impacts of shading on flower formation and longevity, leaf chlorophyll and growth of *Bougainville glabra*. Asian Journal of Plant Sciences 9:20-27.
- Saifuddin M, Hossain ABMS, Normaniza O, Moneruzzaman KM (2009a) Bract size enlargement and longevity of *Bougainvillea spectabilis* as affected by GA₃ and phloemic stress. Asian Journal of Plant Sciences 8:212-217.
- Saifuddin M, Hossain ABMS, Normaniza O, Nasrulhaq BA, Moneruzzaman KM (2009b) The effects of naphthaleneacetic acid and gibberellic acid in prolonging bract longevity and delaying discoloration of *Bougainvillea spectabilis*. Biotechnology 8:343-350.
- Sarkka L, Erikson C (2003) Effects of bending and harvesting height combinations on cut rose yield in a dense plantation with high intensity lighting. Sci Hort 98:433–447.
- Simon A, Toth G, Duddeck H, Soliman H, Mahmoud I, Samir H (2006). Glycosides from *Bougainvillea glabra*. Natural product research 20:63-67.
- Stitt M, Muller C, Matt P, Gibon Y, Carillo P, Morcuende R, Scheible WR, Krapp A (2002) Steps towards an integrated view of nitrogen metabolism. J Exp Bot 53:959–970.
- Stitt M, Schaewen A, Willmitzer L (1990) Sink regulation of photosynthetic metabolism in transgenic tobacco plants expressing yeast invertase in their cell wall involves a decrease of the Calvin-cycle enzymes and an increase of glycolytic enzymes. Planta 183:40–50.
- Suxia X, Qingyun H, Qingyan S, Chun C, Brady AV (2009) Reproductive organography of *Bougainvillea spectabilis* Willd. Scientia Horticulturae 120:399-405.
- Wealtherly PE, Slatye RO (1962) Relationship between relative turgidity and diffusion pressure deficits in leaves. Nature 179:1085-1086.
- Zieslin N, Mor Y (1981) Plant Management of greenhouse roses. The pruning. Sci Hort 14:285–293.