

Floriculture Research in Sri Lanka

**Proceedings of the National Symposium
on Floriculture Research
2015**



**Department of National Botanic Gardens
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**Proceedings of the
National Symposium on Floriculture Research
(NaSFloR)**

2015

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Message from the Chairman

I have pleasure in sending this message on the proceedings of 8th National Symposium of the Floriculture Research and Development 2015. The research papers provide the direction for the future floriculture development of the country. The future economic development and standards of living are the direct results of the agricultural research undertaken at present. Thus the agricultural research plays a pivotal role in assuring food security and farmer lively hoods in the country.

Today and in the future, the potential for commercial floriculture expansion in Asia, including production for domestic and export sales of cut flowers, is greater than ever before. The elements for success needed to transform the Asian flower industry into the worldwide leader of commercial floriculture have been implied by reviewing the evolution of the world cut flower market place.

There is considerable opportunity in both export and domestic markets for cut flowers from southern Asia. In case of export markets, it seems reasonable to set short, middle, and long term goals with respect to entering these markets. Simultaneously, efforts to develop domestic markets within and among countries in Southeast Asia should be undertaken.

The National Committee on Floriculture Research and Development of SLCARP has achieved commendable heights by developing this valuable document. This is a result of the collective effort of the scientists in the National Agricultural Research System. I would like to express my since appreciation to the members of the National Committee on Floriculture Research and Development, Editorial board of the proceeding and the Scientists in the National Agricultural Research System.

Dr. S. S. B. D. G. Jayawardane
Chairman
Sri Lanka Council for Agricultural Research Policy

Message from Director

It is indeed a great pleasure for me to extend my sincere wishes to all the stakeholders of the proceedings of the 8th National Symposium of Floriculture Research -2015. As this Symposium was held for 8th time, I believe it shows enthusiasm and the hard work of its organizers and also of the researches who have contributed to the symposium. As the Sri Lanka Council for Agricultural Research Policy, it is inspiring to observe that the research culture has taken an upward trend, where scientists in the National Agricultural Research System (NARS) and academics are actively engaged in floriculture research which would ultimately help the development of the country.

This document itself is not an end; we continually explore, create and adopt new innovations to keep the Symposium every year. This process will keep SLCARP at the forefront in the arena of agricultural research. Leadership has been provided by the Ministry of Agriculture, the strength of our staff and the support we receive from the Department of National Botanic Gardens are instrumental in delivering the results.

The proceedings of the 8th National Symposium of Floriculture Research is no doubt a valuable document for the floriculture sector and I take this opportunity to congratulate the Chairman and the members of the Committee on National Floriculture Research and Development, Members of the Editorial Board and researchers who share their knowledge in this Symposium.

Dr. J. D. H. Wijewardana
Secretary
Sri Lanka Council for Agricultural Research Policy

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Preservation of *Polyscias guilfoylei* and *Chlorophytum comosum* for floral arrangements

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ABSTRACT

Value addition for cut foliage is a good tool to increase its demand among the customers who regularly enjoy floricultural products throughout the world. In this experiment, we practiced floral dyeing and drying to increase the appearance and to preserve foliage for different occasions. Food colouring (mL) and cochineal (g) were used as foliage dyeing agents and it was mixed with water (mL) in different ratios; 1:0.5, 1:1, 1:2 and 0.5:1 colouring agents to water respectively. Their colour intensities were ranked using a scale 0-4 (colour is; 0-not changed, 1-less changed, 2-moderately changed, 3-highly changed, and 4-very highly changed) according to the floral colour absorption. Two plant species *Polyscias guilfoylei* and *Chlorophytum comosum* were used as cut foliage. Microwave 80 power range for two minutes was used for foliage drying. Statistical analysis was performed using SAS software (version 9.1.3). Cochineal gave the best results compared to food colouring for floral dyeing. Higher colour intensity was observed in ratio of cochineal 0.5 g: 1 mL water. It was also observed that, *Polyscias guilfoylei* effectively absorbed dye compared to *Chlorophytum comosum*.

Keywords: *Polyscias guilfoylei*, *Chlorophytum comosum*, foliage, dyeing, drying

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INTRODUCTION

Cut flowers as well as cut foliage play a major role in the floriculture industry. There is a substantial growth in the floriculture industry in Sri Lanka. If this potential is fully met, the turnover can be doubled. The industry's global growth rate is about 15 percent annually, however in Sri Lanka, it has been 4-5 per cent over the last few years (Kumara *et al.*, 2007). Value addition for cut foliage is a good way to increase its demand to meet the commercial market.

Fresh cut foliage has a short shelf life due to their perishable nature. There is a high demand for cut foliage throughout the year which can be substituted by everlasting dry foliage. Dry foliage products and botanicals hold tremendous potential since they are cheaper, easily available, eco-friendly and biodegradable (Vishnupriya and Jawaharlal, 2014). Dry foliage has a good demand in both domestic and international market. Dyed dry foliages provide more colourful value added product of high quality to the customers during season and offseason.

Different types of drying methods are used for foliage drying such as air drying, microwaving, use of desiccants, glycerin drying, freeze drying and water drying. Microwave drying, which takes only a few minutes in the oven, provides material that looks fresher and more colourful than that obtained by other methods. Since flowers that are air-dried often become deformed in the process, a good alternative is to use a desiccant. The main advantage of using a drying agent is that, it absorbs water

from the flowers while still allowing the flowers to retain their shape and colour (Brown, 2013). This study was conducted to find out effective colouring agent (Food colouring and Cochineal), the ratio of their dilution with water and to check the best floral drying techniques for two species *Polyscias guilfoylei* and *Chlorophytum comosum*.

MATERIALS AND METHODS

Polyscias guilfoylei and *Chlorophytum comosum* were used for the experiment. Food coloring (mL) and Cochineal (g) were used as dyeing agents with water (mL) in different dilution ratios (1:0.5, 1:1, 1:2 and 0.5:1 colouring agents to water respectively). Cut foliages were dipped in food colouring and cochineal solutions separately, which consists with different ratios. Colour changes were observed in foliage after 24 hours and colour intensities were ranked using a scale (Table 1). Then, they were dried using a microwave 80 power range for two minutes (Perera and Dahanayake, 2014). Thereafter foliage were also ranked using scale given in the Table 1. Experiment was comprised four treatments and each treatment was replicated thrice. Statistical analysis was performed the using SAS software (version 9.1.3).

Table 1: Ranking method to assess the change of color of cut foliage.

Observations	Scale
Not changed	0
Less	1
Moderate	2
High	3
Very High	4

RESULTS AND DISCUSSION

Cochineal showed better colour formation than the food colouring in both cut foliage, *Chlorophytum comosum* and *polyscias guilfoylei*. All the concentrations did not yield the same colour intensity. In cochineal (g) to water (mL), the ratio of 0.5:1 yielded the best colour intensity for *polyscias guilfoylei*. The lowest colour intensity for both types of foliage was observed in cochineal (g) to water (mL) at 1:0.5. Food colouring did not show any colour change in *Chlorophytum comosum* and the ratio of 1:2 and 0.5:1 produced a lower colour intensity in *polyscias guilfoylei* (Table 2). After microwave drying, there were no significant differences in appearance of dried foliages.

After drying, colour was retained in *Polyscias guilfoylei* which were dipped in Cochineal solution and the colour did not retain in the foliage which were dipped in food colouring (Fig. 1). Dye is absorbed by leaves with water molecules and retains on the surface of the leaves and flower petals after water is evaporated by drying. In foliage dying, cochineal was more effective than the food coloring. It has yielded a higher colour intensity compared to food colouring and colour intensity was different depending on the concentration. Ratio of cochineal (g) to water (mL), 0.5:1 was the most effective while the ratio of cochineal (g) to water (mL), 1:0.5 was less effective in colouring of both species.

Table 2: Ranking the colour of cut foliage according to the scale

Ratio of Colouring agent to water	Before Drying				After Drying			
	Food Colouring		Cochineal		Food Colouring		Cochineal	
	C. <i>comosum</i>	P. <i>guilfoylei</i>	C. <i>comosum</i>	P. <i>guilfoylei</i>	C. <i>comosum</i>	P. <i>guilfoylei</i>	C. <i>comosum</i>	P. <i>guilfoylei</i>
1:0.5	0 ^e	0 ^e	1 ^d	1 ^d	0 ^e	0 ^e	1 ^d	1 ^d
1:1	0 ^e	0 ^e	3 ^b	2 ^c	0 ^e	0 ^e	3 ^b	2 ^c
1:2	0 ^e	1 ^d	2 ^c	3 ^b	0 ^e	1 ^d	2 ^c	3 ^b
0.5:1	0 ^e	1 ^d	3 ^b	4 ^a	0 ^e	1 ^d	3 ^b	4 ^a

Column values followed by the same letter are not significantly different as determined by Duncan's multiple range test ($P=0.05$).



Figure 1: Colour absorption in fresh foliage. Under food colouring; a, *Chlorophytum comosum* and b, *Polyscias guilfoylei*. Under cochineal; c, *Chlorophytum comosum* and d, *Polyscias guilfoylei*.

Polyscias guilfoylei effectively absorbed the dye compared to *Chlorophytum comosum* and produced a good colour. In drying process, *Polyscias guilfoylei* retained its colour. Silica gel, borax mixtures, and expanded clay cat litter work well for drying of flowers, out of which silica gel is the most preferred substance (Brown, 2013). However, colour changes can be occurred during the drying process using these chemicals: Borax may turn pink flowers to mauve, red generally becomes purple or bluish, pure blue acquires lavender or purplish color, magenta turns to lavender and yellow and orange are usually well-preserved and possibly intensified (Brown, 2013). Microwave drying, which takes only a few minutes, provides material that looks fresher and more colourful than that obtained by other drying methods.

CONCLUSION

Cochineal (g) to water (ml), 0.5:1 ratio yielded the highest colour intensity. *Polyscias guilfoylei* effectively absorbed the dye than *Chlorophytum comosum*.

REFERENCES

- Brown C., (2013). Changing the Color of Flowers, All Science Fair Projects.com [online] http://www.all-science-fair-projects.com/print_project_1548_50, accessed on 05.05.2015.
- Kumara G.K.K.P., Angunawela, R.K. and Weerakkody W.A.P. (2007). Plant selection, pre-harvest treatments and post-harvest management to prolong the vase-life of shoot cuttings of *Codiaeum variegatum* (croton), J. Natn. Sci. Foundation Sri Lanka, 35(3): 153-159.

Perera P.C.D. and Dahanayake N., (2014). Evaluation of effective time and microwave power levels for quick flower drying with minimum colour change. National Symposium on Floriculture Research (NaSFLOr): 3.

Vishnupriya K. and Jawaharlal M., (2014). Glycerinization of foliages for dry flower products making, An International Quarterly Journal of Life Sciences: 507-511.

Leaf yellowing and browning problem in aquatic plant *Echinodorus bleheri*

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ABSTRACT

Echinodorus bleheri (*E. bleheri*) is a one of the popular aquatic plants which is commonly known as 'Amazon sword' plant. This plant is cultivated for growing in artificial aquatic habitats. Cultivation of this plant has been easy due to non-occurrence of diseases in this species. However, very recently browning of leaves was recorded in *E. bleheri* mother plant cultivations in Sri Lanka and causal agent of this problem has not been identified yet. Therefore, this study was carried out to find the causal agent/s of browning of *E. bleheri* leaves. Leaves with symptoms were cultured on Potato Dextrose Agar (PDA) and Nutrient Agar (NA) media. Fungus was identified by morphological features and molecular methods. A gram positive bacterium was identified on NA medium while on PDA media fungi such as *Colletotrichum* spp, *Nigrospora* spp, *Botryodiplodia* spp, *Pestalotia* spp were identified. There were around 20 unidentified mycelium spp. Through molecular means, *Daldinia* sp were identified.

Keywords: Aquatic plant, *Echinodorus bleheri*, leaf browning

INTRODUCTION

Aquatic plants are plants that are adapted to aquatic environments, salt water or fresh water. These plants require special adaptations for living submerged in water or at the water surface. The most common adaptation is having aerenchyma, but floating leaves and finely dissected leaves are also common.

Echinodorus bleheri (*E. bleheri*), which is commonly known as Amazon sword plant, is one of the popular aquatic plants for their attractive form and general hardiness. This plant belongs to family Alismataceae and distributed in the Western Hemisphere from the Central United States to Argentina. This plant is cultivated for and used in ponds and artificial aquatic habitats. Mainly *in-vitro* propagation methods are developed to increase the production of *Echinodorus* spp., including *E. bleheri* (Dissanayake et al, 2007). Cultivation of this plant is very easy mainly because of non-occurrence of disease problems. However, very recently browning of leaves was recorded in *E. bleheri* mother plant cultivations in Sri Lanka. Causal agent of this problem has not been identified yet. According to the previous studies a leaf spot disease was recorded in water lily, which caused by *Colletotrichum* spp. (Chowdappa et al, 2012 and Johnson et al, 1997). However, little information is available pertinent to association of fungi with aquatic plants. Therefore, this study was carried out to find the causal agent/s of this problem and to expand the knowledge about diseases associate with aquatic plants.



Figure 1. Leaf yellowing and browning problem in *Echinodorus bleheri*

MATERIALS AND METHODS

Isolation of the pathogen

Leaf species with yellowing and browning symptoms were surface sterilized with 3% sodium hypochlorite solution, washed by sterile distilled water and placed in Potato Dextrose Agar (PDA) and Nutrient agar (NA) medium containing petri dishes separately. Those PDA plates were incubated at 28 °C for 5-7 days and NA plates were incubated at 28 °C for 24 hours. Bacterial colonies were sub cultured and identified microscopically. Morphologically unique fungal colonies were sub cultured and purified using standard techniques.

The fungus species were identified and characterized based on their morphological characters and microscopic analysis by using taxonomic guides and standard procedures. The following morphological characters were evaluated; colony color, colony growth, spore shape and spore length.

Molecular detection

Genomic DNA extraction procedure was done for selected fungi samples to extract fungal genomic DNA. The PCR amplification was done using universal primers ITS1 (5'TCC GTA GGT GAA CCT GCG 3') and ITS4 (5' TCC TCC GCT TAT TGA TAT GC 3') (Tuyama et al, 2013). The thermocycler conditions were initial denaturation 94 °C for 5 minutes, followed by 35 cycles, denaturation at 94 °C for 30 seconds, annealing at 55 °C for 30 seconds, and extension 72 °C for 1 minute with final extension at 72 °C for 5 minutes. Then PCR products were analyzed (Figure 03). Then PCR product was sent for sequencing ('Macrogen'- Korea). Sequenced results were analyzed using BLAST search programme.

RESULTS AND DISCUSSION

According to the Gram's staining and KOH test, Gram positive *Bacillus* spp. were identified from the infected leaf parts.

Isolation and characterization of fungal strains

A total of 14 fungal isolations were obtained from the analysis. All fungal isolates were obtained in pure cultures by using standard procedure. The photomicrographs of all the fungal isolates were taken to assist in identification of fungal isolates. The cultural characteristics and sporulating

structures of these isolates are represented in Figure 2. Four fungal isolates were identified as *Colletotrichum* sp., *Nigrospora* sp., *Botryodiplodia* sp., and *Pestalotia* sp. There were around 10 unidentified mycelium spp.

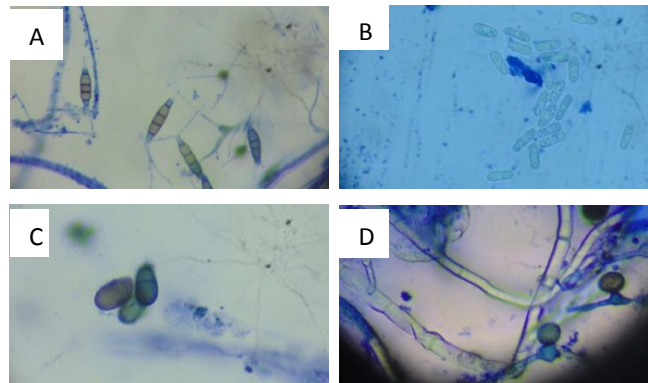


Figure 2. Sporulating structures of identified fungal species. A- Spores of *Pestalotia* sp., B- spores of *Colletotrichum* sp., C- Spores of *Botryodiplodia* sp. and D- spores of *Nigrospora* sp.

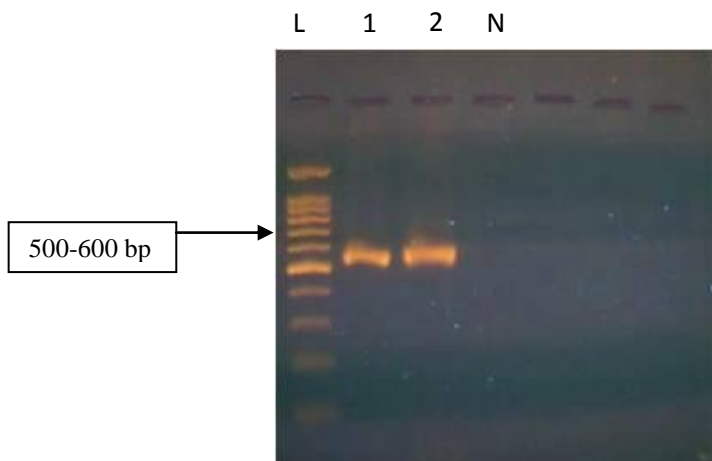


Figure 03. Agarose gel electrophoresis of amplified gene product after PCR (ITS1 and ITS4) L-1Kb ladder, 1 and 2 are two fungal samples and N is the negative control

According to the BLAST results, two fungal samples showed 94% similarity to the *Daldinia eschscholtzii* which can act as an endophyte. In addition, they have antimicrobial and nematocidal activity. Further confirmation of the causal agent is being carried out. This is the first record of fungal infection in aquatic plants in Sri Lanka.

CONCLUSION

From the present investigation, it is concluded that a total of 14 fungal species were isolated from the *E. bleheri* yellowing and browning leaves. Four fungal spp. belonging to the phylum of Ascomycota were identified based on their morphological characters and microscopic analysis namely *Colletotrichum* sp., *Nigrospora* sp., *Botryodiplodia* sp., and *Pestalotia* sp. Through molecular means *Daldinia eschscholtzii* were identified.

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REFERENCES

- Chowdappa P., Chethana C.S., Bharghavi R., Sandhya H. and Pant R.P. (2012). Morphological and molecular characterization of *Colletotrichm gloeosporoids* (penz) sac. isolates causing anthracnose of orchids in. India.Biotechnol.Bioinfo.Bioerg. 2(1):567-572.
- Dissanayake C., Hettiarachchi m. and Iqbal M.C.M. (2007). Sustainable use of *Cryptocoryne wendtii* and *Echinodorous cordifolius* in the aquaculture industry of Sri Lanka by micropropagation. Sri Lanka .J.quat.sci.12:89-101.
- Johnson D.A., Caris C.M. and Rogers J.D. (1997). Morphological and molecular characterization of *Colletotrichm nymphaeae* and *C. nupharicola* sp. nov. on water lilies (*Nymphaea* and *Nuphar*). Mycol.Res.101(6):641-649.
- Harnngton T.C., steimel J., Workneh F. and yang X.B. (2000). Molecular identification of fungi associated with vascular discoloration of soybean in the North Central United States. Plant Diseases.84:83-89.
- Tuyama K.T., Pereira J., Maki C.S. and Ishikawa N.K. (2013). *Daldinia eschscholtzii* (Ascomycota, Xylariaceae) isolated from the Brazilian Amazon: taxonomic features and mycelia growth conditions. Acta Amazonica. 43(1):1-8

Effect of Gibberellin and Cytokinin on lateral shoot formation of *Anthurium*

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ABSTRACT

Anthurium is a major cut flower species in tropical and subtropical countries and an economically important genus in the family Araceae. However, non-availability of quality planting materials is a major problem in *Anthurium* cultivation. Therefore, present study was carried out to induce lateral shoot formation of topped *Anthurium andreanum* with Gibberellin (GA₃) in combination with Cytokinin (BAP). Two varieties of *Anthurium andreanum*, i.e. 'Tropical red' and 'Safari' were selected for the study. Six months old plants were potted in 4 cm diameter plastic pots containing leaf mould, cattle manure and sand (4:2:1) with ¼ inch brick pieces. Before decapitation, plants were kept for two months in a plant house of the National Botanic Gardens Peradeniya. Application of different concentrations of GA₃ (125, 250, 375 and 500 ppm) were applied in combination with constant level of BAP (250 ppm) at 10 days interval. Number of lateral shoots, length of lateral shoots, number of leaves per sucker, leaf length, number of roots and time taken for sucker formation were recorded after the hormone treatment at 10 days interval. Among different treatments tested, *Tropical red* recorded the highest number of new suckers, shoot length and number of roots per sucker than *Safari*. However *Safari* recorded the highest average value of leaf length than *Tropical red*. Number of leaves per plant did not increase remarkably due to application of hormones in both varieties. However, *Tropical red* performed better in all treatments tested. Overall results showed that the application of 375 ppm Gibberellin in combination with 250 ppm of Cytokinin was the most effective treatment to induce lateral shoots as well as to improve growth performance in both varieties. Furthermore, this study can be extended to induce sucker formation of foliage *Anthuriums* as well.

Keywords: *Anthurium*, Tropical red, Safari, Gibberellin (GA₃), Cytokinin (BAP), lateral shoots

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INTRODUCTION

Anthurium is a major cut flower species in the tropical and subtropical countries and an economically important genus in the family Araceae (Anon, 2000). Even though Anthuriums have a high demand in the local market as well as in the export market, and large numbers of small scale growers are also involved in *Anthurium* cultivation, lack of availability of quality planting materials badly affect the industry. Therefore, it is essential to produce quality planting material consistently in order to maintain continuous production of Anthuriums (Yakandawela *et al.*, 2000). *Anthurium* can easily be multiplied by division of offshoots with portion of aerial roots from the main stem (Ekanayake and Hagen, 1977). Cultivars belonging to *Anthurium andreanum* generally produce 14-15 offshoots per plants in a year (Singh, 2006). Propagation by tissue culture technique appears as an alternative to increase the production and propagation of *Anthurium*, and it is commercially used by

growers. However, tissue culture is not a cost effective method (Martin *et al.*, 2003). Furthermore, *Anthurium* can be reproduced through a lateral shoots (suckers) which arise around the base of the stem. However, many *Anthurium* cultivars do not easily produce suckers with large numbers (Godigamuwa, 2010). *Tropical red* and *Safari* varieties are cut flowers that have very high demand in the export market due to their attractive flowers. *Tropical red* and *safari* plants naturally produce very few numbers of suckers in their life cycle. Plant growth regulators such as Gibberellins and Cytokinin are used to increase the plant growth and to obtain more suckers production (Godigamuwa, 2010). Cytokinin is used for multiplication of Anthuriums (Upamalika, 2003). It has been reported that after adding hormones i.e. Gibberellins and Cytokinin, the sucker formation in vegetatively propagated Anthuriums was enhanced (Godigamuwa, 2010). Thus, it is also envisaged that sucker formation can be increased in *in-vivo* propagated plants of *Tropical red* and *safari* with application of Gibberellins and Cytokinin. Hence this study was carried out to investigate the lateral shoots induction of topped *Anthurium andreanum* with Gibberellins and Cytokinin.

MATERIALS AND METHODS

The experiment was carried out at the Floriculture Research Unit of the National Botanic Gardens Peradeniya from January 2013 to October 2013. Plants were grown under net house condition as a pot experiment.

Design of the Experiment: The experiment was laid out in a Completely Randomized Design (CRD) with three replicates for each treatment and each replicate consisted of 04 individual plants.

Selection of varieties: Two *Anthurium andreanum* varieties, *Tropical Red* (dark green heart shaped leaves and large red colour spathe) and *Safari* (long reddish green leaves and red colour spathe), maintained under similar conditions, were selected for the experiment.

Plant management: Six-month old potted *Anthurium* plants were kept for two months in a plant house in preparation for decapitation. All cultural practices were done according to the DOA recommendations; i.e. weed control, shading, fertilization and watering etc. A weekly Cu Fungicide – Copper Oxychloride, treatment was applied in order to control spread of bacterial blight disease.

Topping of plants: Plants were topped two months after keeping in a plants house. Plant stems were cut with the basal section having at least 2-3 leaves and upper section having a minimum of 1-2 adventitious roots. Topping is the manual removal of the terminal portion. Slanted cuts using sharp secateurs were made and instruments were disinfected by dipping in a fungicide when using from plant to plant. Cutting surfaces were also disinfected using Mancozeb in a paste form.

Application of Gibberellin (GA₃) and Cytokinin (BAP): Plants were treated twice with 250 ppm BAP in combination with different concentrations (125, 250, 375 and 500 ppm) of Gibberellin. Topped *Anthurium* plants were sprayed with BAP after topping on the first day. GA₃ was sprayed on the following day. Hormonal treatments were done in the morning. Second hormone application was done after 10 days of the first application.

Data collection: Data were recorded just after initiation of lateral shoots 10 days after the second hormone application. Number of shoots per plant, length of shoots, leaves per shoots, leaf length, and roots per shoots as well as time duration for higher sucker formation were recorded.

Statistical Analysis: Data were tabulated and analyzed by using Analysis of Variance (ANOVA) procedure of Statistical Analysis System (SAS). Duncan's New Multiple Range Test (DNMRT) was used

to compare the differences among the treatment means at $p=0.05$. Linear Correlation Analysis was performed to determine the strength of the relationships between measured parameters.

RESULTS AND DISCUSSION

Effect of Gibberellin (GA_3) in combination with Cytokinin (BAP) on sucker formation

Newly developing lateral shoots (suckers) were clearly visible in 10 days after the second hormone application. Number of lateral shoots per plant increased with increasing concentration of GA_3 (125, 250, 375 and 500 ppm) in combination with 250 ppm of BAP compared to the control in both varieties. *Tropical red* produced the highest number of lateral shoots per plant 20 days after planting while *Safari* produced the highest number of lateral shoots 30 days after hormone application. The highest number of lateral shoots was recorded in plants treated with 250 ppm BAP with 375 ppm GA_3 in both varieties. Furthermore, production of lateral shoots per plant was significantly different in all treatments compared to the control ($p=0.05$). According to the study findings, there was a significant difference ($p=0.05$) between treatments in both varieties, i.e. *Tropical red* and *Safari* (Table 1).

Table 1: Effect of Gibberellin and Cytokinin on new sucker formation of *Tropical red* and *Safari*

Treatment	Number of new suckers	
	<i>Tropical red</i>	<i>Safari</i>
T ₁	1.42 ^c	1.25 ^c
T ₂	1.61 ^c	1.5 ^c
T ₃	2.42 ^b	2 ^b
T ₄	3.75 ^a	2.58 ^a
T ₅	2.17 ^b	1.51 ^c
LSD	0.3	0.45

Note : Means of each category with the same letters are not significantly different at $p=0.05$.

The tendency for shoot formation depends on the variety (Upamalika, 2003). *Tropical red* recorded the highest mean value of suckers than *Safari*. The success rate of shoot induction of *Anthurium* in this study was similar to that of Godigamuwa (2010).

Effect of Gibberellin (GA_3) and Cytokinin (BAP) on time taken for sucker formation of *Anthurium andreanum*:

Newly developed lateral buds were observed 10 days after the second hormone application. Maximum number of lateral shoots for *Tropical red* (4) was observed after 20 days and for *Safari* (3), it was observed after 30 days. Plants treated with 250 ppm BAP in combination with 375 ppm GA_3 recorded the maximum number of suckers (lateral shoots) within a short period of time when compared to the control in both varieties tested (Fig. 1).

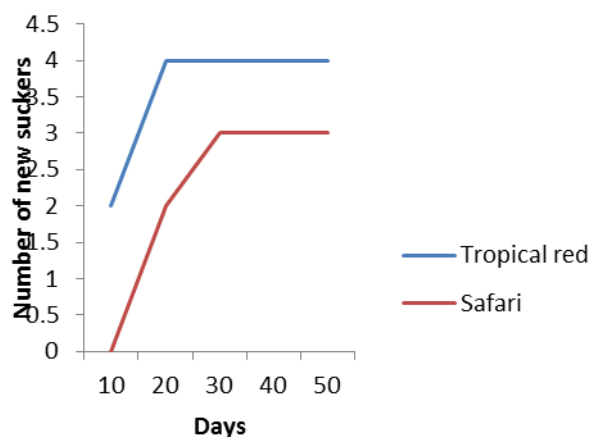


Figure 1. Effect of Gibberellin and Cytokinin on time taken for sucker formation

Effect of Gibberellin and Cytokinin on growth parameters of new suckers

(a) Shoot length of new suckers: Statistical analysis indicated that there was a treatment effect in the experiment ($p=0.05$). According to the results obtained, tallest shoots were observed in plants treated with 375 ppm GA_3 in combination with 250 ppm BAP (T4) in both varieties. They were significantly different compared to the control. Comparatively, *Tropical red* recorded the highest mean number of shoot length (6.5 cm) than *Safari* (4 cm) as shown in Table 2.

Table 2: Effect of Gibberellin and Cytokinin on Shoot length (cm) per sucker

Treatment	Shoot length(cm)	
	<i>Tropical red</i>	<i>Safari</i>
T ₁	3.65 ^d	2.05 ^c
T ₂	4.23 ^c	2.73 ^d
T ₃	6.21 ^b	4.11 ^b
T ₄	6.56 ^a	4.38 ^a
T ₅	5.62 ^c	3.64 ^c
LSD	0.37	0.35

Note : Means of each category with the same letters are not significantly different at $p=0.05$.

(b) Number of leaves per sucker: There is no significant difference between treatments compared to the control in both varieties. *Tropical red* recorded the highest number of leaves per sucker (2) when compared to the *Safari*. Both varieties showed a significant difference ($p=0.05$) between treatments as shown in Table 3. Plants were affected with bacterial blight during the study and it may have led to a decrease in further growth and development of plants. This may have directly affected the decrease of leaves in both varieties.

(c) Leaf length of new suckers: There was a significant difference between treatments ($P=0.05$). The highest mean value of leaf length per plant was observed in plants treated with 375 ppm GA_3 in combination with 250 ppm BAP (T4) in both varieties. Comparatively *Safari* recorded the highest leaf length (5.32 cm) than *Tropical red* (4.1 cm) as shown in Table 4.

(d) Number of new roots per suckers: Among different treatments tested, the highest number of roots per plant was recorded in plants treated at 375 ppm GA_3 in combination with 250 ppm BAP (T4) in both varieties. They also were significantly different compare to the control. Comparatively *Tropical red* recorded the highest number of roots (8) than *Safari* (6) as shown in Table 5.

Table 3: Effect of Gibberellin and Cytokinin on number of new leaves per sucker

Treatment	Number of new leaves	
	<i>Tropical red</i>	<i>Safari</i>
T ₁	1.96 ^a	1.46 ^a
T ₂	1.87 ^b	1.29 ^a
T ₃	1.49 ^b	1.25 ^a
T ₄	1.68 ^{ab}	1.66 ^a
T ₅	1.75 ^{ab}	1.33 ^a
<i>LSD</i>	0.34	0.27

Note: Means of each category with the same letters are not significantly different at p=0.05.

Table 4: Effect of Gibberellin and Cytokinin on leaf length (cm) per sucker

Treatment	Leaf length(cm)	
	<i>Tropical red</i>	<i>Safari</i>
T ₁	2.25 ^d	2.44 ^e
T ₂	2.73 ^c	3.91 ^d
T ₃	3.56 ^b	4.73 ^b
T ₄	4.51 ^a	5.32 ^a
T ₅	3.05 ^c	4.2 ^c
<i>LSD</i>	0.34	0.27

Note: Means of each category with the same letters are not significantly different at p=0.05.

Table 5: Effect of Gibberellin and Cytokinin on number of roots per sucker

Treatment	Number of new roots	
	<i>Tropical red</i>	<i>Safari</i>
T ₁	3.00 ^b	3.00 ^c
T ₂	3.00 ^b	3.00 ^c
T ₃	6.00 ^a	5.50 ^{ab}
T ₄	8.00 ^a	6.50 ^a
T ₅	6.00 ^a	5.00 ^b
<i>LSD</i>	2.29	1.15

Note: Means of each category with the same letters are not significantly different at p=0.05

Roy et al., (2004) stated that the highest number of roots and the longest roots per plant in rose explants were observed in 1 mg/L 0.5 mg/L NAA. Wernner and Motyka (2001) found that BAP was involved in controlling both root growth and the generation of new root meristem.

Correlation Analysis

Correlation analysis for growth parameters of new suckers of *Tropical red*:

When correlation analysis was performed for the overall data set, number of suckers showed a highly significant ($p < 0.0001$) positive correlation with shoot length and leaf length of sucker (Table 6). Numbers of suckers were not significantly correlated with number of leaves and number of roots. Shoot length showed a negative correlation with number of leaves.

Table 6: Linear Correlation Coefficients between Number of Suckers (NS), Shoot length (SL), Number of Leaves per sucker (NLVS), Leaf length (LVFL), and Number of roots (RT) per sucker in *Tropical red*

	NS	SL	NLVS	LVFL	RT
NS	-	0.86***	-0.43 ^{ns}	0.96***	0.38 ^{ns}
SL	-	-	-0.68*	0.88***	0.41 ^{ns}
NLVS	-	-	-	-0.55*	0.03 ^{ns}
LVFL	-	-	-	-	0.26 ^{ns}
RT	-	-	-	-	-

Note: ns- non Significant at $p = 0.05$; * Significant at $p < 0.05$; ** Significant at $p < 0.01$; *** Significant at $p < 0.0001$

Correlation Analysis for growth parameters of new suckers of *Safari*

When correlation analysis was performed for the overall data set, number of suckers showed moderately significant ($p < 0.01$) positive correlation with shoot length and leaf length of suckers (Table 7). Number of suckers showed significant ($p < 0.05$) positive correlation with number of roots per sucker and did not show a significant correlation ($p > 0.05$) with number of leaves. Shoot length showed a highly significant ($p < 0.0001$) positive correlation with leaf length and did not show significant correlation with number of leaves per sucker.

Table 7 Linear Correlation Coefficients between Number of Suckers (NS) , Shoot length (SL), Number of Leaves per sucker (NLVS), Leaf length (LVFL), and Number of roots (RT) per sucker in *Safari*

	NS	SL	NLVS	LVFL	RT
NS	-	0.76**	0.32 ^{ns}	0.74**	0.51*
SL	-	-	0.06 ^{ns}	0.92***	0.28 ^{ns}
NLVS	-	-	-	0.03 ^{ns}	0.46 ^{ns}
LVFL	-	-	-	-	0.30 ^{ns}
RT	-	-	-	-	-

Note: ns- non Significant at $p = 0.05$; * Significant at $p < 0.05$; ** Significant at $p < 0.01$; *** Significant at $p < 0.0001$

CONCLUSION

Among different treatments tested, *Tropical red* recorded the highest number of new suckers, shoot length and number of roots per sucker than *Safari*. However *Safari* recorded the highest average value of leaf length than *Tropical red*. Number of leaves per plant did not increase remarkably due to application of hormones in both varieties. However, *Tropical red* performed better in all treatments tested. Overall results showed that the application of 375 ppm Gibberellin in combination with 250 ppm of Cytokinin was the most effective treatment to induce lateral shoots as well as to improve growth performance in both varieties. Furthermore, in the present study, six months old *in vitro* plants were treated with different concentrations of Gibberellin and Cytokinin. However, this could be stressful for plants. Therefore, it can be suggested to use at least one year old *Anthurium* plants to induce lateral shoot formation, in order to reduce the stress associated with the hormone application. Furthermore, this study could be extended to induce sucker formation of foliage *Anthuriums* as well.

REFERENCES

- Anonymous (2000). Sri Lanka Export Development Board special *Anthurium* project. 1:2-7.
- Ekanayake, D.T and Hagen, P. W. (1977) Hand book for cut flower growers. Western Agricultural team collaboration with Royal Botanical Gardens, Peradeniya. Sri Lanka.
- Godigamuwa, G.R.C.N.K. (2010). Effect of GA₃ in combination with BAP on lateral shoot induction of topped *Anthurium in-vivo*.
- Martin, K.P.D, Joseph J, Madassery and V.J.Philip. (2003). Direct shoot regeneration from lamina explants of two commercial cut flower cultivars of *Anthurium andreaeanum* Hort. *In-vitro* cell. Dev.Biol-plant.39:500-504.
- Singh, A.K. (2006). Flower Crops Cultivation and Management. No.2, 23-34.
- Upamalika, H.S. (2003). Effect of Cytokinin on *in-vitro* multiplication of *Anthurium andreaeanum*.
- Werner, T., Motyka V. Strned, M. and Schmulling, T. (2001). Regulation of Plant growth by Cytokinin. Institute for plant breeding research. Cologne. Germany.
- Yakandawela,G., Peiris S.E. and Yakandawela, Y.L.N. (2000). Transient expression of *uida* reporter gene in regenerable callus tissues of *Anthurium andraeanum* Lind. by agrobacterium mediated transformation. Tropical Agriculture. p 20-25.

Evaluation of suitable rooting media for *Bougainvillea* (*Bougainvillea spectabilis*) stem cuttings and frequency of flowering

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ABSTRACT

Bougainvillea spectabilis is primarily propagated by stem cuttings. However, problems associated with rooting of stem cuttings are obstacles for vegetative propagation. Therefore present study was conducted to identify proper maturity stage of stem cuttings and appropriate medium for *Bougainvillea spectabilis* for rooting of cuttings and optimum flowering. The experiment was carried out in a plant propagator at the Floriculture Research Unit of the National Botanic Gardens Peradeniya by using three different stem cuttings (Soft wood, Semi hard wood and Hard wood) planted in three different media (Sand + Coir dust + Compost, Sand + Coir dust, and Coir dust only). The experimental design was two factor factorial laid out with 9 treatments each replicated thrice. Data were collected at 21 days after propagation and subsequent data were gathered at 10 days interval, i.e. number of new leaves, number of roots, number of shoots and height of the plant. Data were tabulated and analyzed by using Analysis of Variance (ANOVA) procedure of Statistical Analysis System (SAS). Duncan's New Multiple Range Test (DNMRT) was performed to compare the differences among treatment means at $p=0.05$. In the first experiment, Semi hard wood cuttings performed best in the Coir dust + Sand (1:1) rooting medium. In the second experiment selected cutting type i.e. Semi hard wood and the best medium i.e. Coir dust + Sand was used to study the effect of different fertilizer applications on flowering frequency of *Bougainvillea*. The second experiment was arranged in a Completely Randomized Design (CRD) with five treatments, i.e. Maxi crop (control), Maxicrop + NPK (10:11:18), Maxicrop + NPK (10:52:10), Maxicrop + NPK (20:20:20), Maxicrop + NPK (6:30:30). According to the overall study findings, the best medium for rooting was the mixture of Sand + Coir dust in equal proportions while Maxicrop + NPK (6:30:30) applied at weekly intervals was the best treatment for optimum flowering frequency of *Bougainvillea spectabilis*.

Keywords: *Bougainvillea spectabilis*, rooting media, stem cuttings, fertilizer, flowering frequency

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INTRODUCTION

Bougainvillea (*Bougainvillea spectabilis*) is one of the most popular flowering plant belongs to family Nyctaginaceae (Hammad, 2009). Special characteristics of *Bougainvillea*, i.e. high variation in foliage, production of many flowering inflorescence and continuous blooming with short production cycle has been very useful in ornamental industry (Kobayashi et al, 2007). *Bougainvilleas* are primarily propagated by stem cuttings, i.e. soft wood, semi hard wood and hard wood stem parts can be used for propagation. However, lack of competence to form adventitious roots by cuttings occurs

routinely and is an obstacle for the vegetative propagation (Celine *et al.*, 2006). *Bougainvillea* has extremely fine root system. Therefore it should be planted in well drained soil. Studies have shown that the ability of plants to form roots is influenced by physiological state of the stock plant, propagating environment, and the treatment applied to the cuttings prior to rooting (Hackett *et al.*, 1972). Adventitious root formation is a key step in vegetative propagation and problems associated with rooting of cuttings frequently result in significant economic losses (De Klerk *et al.*, 1999). In the light of this situation it is important to increase the rooting frequency of *Bougainvillea* to induce adventitious root formation and enable rooting of cuttings with minimal economic loss. Therefore present study was conducted to identify proper maturity stage of stem cuttings and appropriate medium and fertilizer ratio for rooting of *Bougainvillea spectabilis* as well as for optimum flowering.

MATERIALS AND METHODS

Experiment I

Evaluation of a suitable rooting medium for *Bougainvillea* stem cuttings: The experiment was carried out in a plant propagator at the Floriculture division of the National Botanic Gardens, Peradeniya from January 2013 to October 2013. Planted *Bougainvillea* stem cuttings were kept in the propagator (10×2') covered with nylon mesh to intercept 50% of the incident radiation.

Preparation of rooting media: Three types of rooting media, i.e. coir dust + sand + compost, coir dust + sand, coir dust were prepared. In all media, equal parts of materials were mixed and filled into 12 cm height black polythene covers.

Selection, preparation and planting of stem cuttings: Stem cuttings were selected from a *Bougainvillea spectabilis* variety which has purple flowers. Cuttings were collected in the morning and kept moist. Stem cuttings at three different maturity stages, i.e. Soft wood (SW), Semi-hard wood (SHW) and Hard wood (HW) were planted in three different rooting media. Each stem cutting was dipped in a hormone solution (Rapid Root®) before planting. All cuttings were kept in a polythene propagator for 21 days. Coir dust was used as a bedding material in the floor of the propagator and watered.

Design of the Experiment: The experimental design was two factor factorial laid out with 9 treatments, each replicated thrice. Each replicate consisted of twelve cuttings as given below. T₁ - Sand + Coir dust + Compost (Semi hard wood), T₂ - Sand + Coir dust + Compost (Soft wood), T₃ - Sand + Coir dust + Compost (Hard wood), T₄ - Coir dust + Sand (Semi hard wood), T₅ - Coir dust + Sand (Soft wood), T₆ - Coir dust + Sand (Hard wood), T₇ - Coir dust only (Semi hard wood), T₈ - Coir dust only (Soft wood), T₉ - Coir dust only (Hard wood).

Data collection: Data were collected at 21 days after propagation and subsequent data were gathered at 10 days interval, i.e. number of new leaves, number of roots, number of shoot and plant height.

Statistical Analysis: Data were tabulated and analyzed by using Analysis of Variance (ANOVA) procedure of Statistical Analysis System (SAS). Duncan's New Multiple Range Test (DNMRT) was performed to compare the differences among treatment means at p=0.05.

Experiment II

Effect of fertilizer on flowering of rooted cuttings: In the second experiment, selected cutting type i.e. Semi hard wood and the best medium i.e. Coir dust + Sand was used to study the effect of different fertilizer applications on flowering frequency of *Bougainvillea*.

Design of the experiment: The second experiment was arranged in a Completely Randomized Design (CRD) with five treatments, i.e. Maxicrop (control), Maxicrop + NPK (10:11:18), Maxicrop + NPK (10:52:10), Maxicrop + NPK (20:20:20), Maxicrop + NPK (6:30:30).

Data collection: Data were collected 41 days after propagation and subsequent data were gathered at 10 days interval, i.e. number of shoots, number of flowers and number of leaves.

Statistical Analysis: Data were tabulated and analyzed by using Analysis of Variance (ANOVA) procedure of Statistical Analysis System (SAS). Duncan's New multiple Range Test (DNMRT) was used to compare the differences among treatment means at $p=0.05$. Linear Correlation Analysis was used to determine the strength of the variables.

RESULTS AND DISCUSSION

Experiment I: Evaluation of a suitable rooting media for planting of *Bougainvillea* stem cuttings

Effect of different rooting media on leaf formation of *Bougainvillea* stem cuttings:

Average number of leaves produced by cuttings under different rooting media ranged from 2.95 to 6.8. Semi hard wood cuttings produced the highest number of leaves as 6.8 in the M2 (Coir dust + Sand) medium and the second highest 5.5 was in the M1 (Coir dust+ Sand + Compost). Hard wood cuttings produced similar number of leaves in the rooting media M2 and M3 (Table 1) while soft wood cuttings produced the lowest number of leaves in all three media.

Table 1: Effect of different treatments on number of leaves of *Bougainvillea* stem cuttings

	M1	M2	M3
SHW	5.53±0.07	6.80 ± 0.39	5.03± 0.25
SW	3.21± 0.15	3.30 ± 0.17	2.95 ± 0.08
HW	5.35± 0.27	5.06 ± 0.23	5.06 ± 0.17

Note: Measurements are the means of 15 replicates ± SE. SHW - Semi hard wood, SW - Soft wood, HW - Hard wood, M1 - Coir dust + Sand + Compost, M2 - Coir dust + Sand, M3 - Coir dust only

Effect of different rooting media on root formation of *Bougainvillea* stem cuttings: The overall result about root formation showed that, semi hard wood (SHW) produced more number of roots in media 1 (Coir dust + Sand + Compost), 2 (Coir dust + Sand) and 3 (Coir dust only) respectively. The lowest number of roots was observed in soft wood cuttings planted in the rooting medium 1 (Coir dust + Sand + Compost). Soft wood cutting produced less number of roots in all media. The second highest number of roots was produced in hard wood cuttings planted in the rooting medium 3 (Coir dust only) (Table 2).

Table 2: Effect of different treatments on number of roots of *Bougainvillea* stem cuttings

	M1	M2	M3
SHW	3.03± 0.07	4.98± 0.40	3.08± 0.17
SW	1.43± 0.05	2.40± 0.13	1.71± 0.32
HW	2.61± 0.18	3.06± 0.15	3.20± 0.25

Note: Measurements are the means of 15 replicates ± SE. SHW - Semi hard wood, SW - Soft wood, HW - Hard wood, M1 - Coir dust + Sand + Compost, M2 - Coir dust + Sand, M3 - Coir dust only

In contrast to the results of present study, Hudson *et al.* (1997) reported that soft wood cuttings generally root easier and quicker than semi hard wood and hard wood of *Woodfordia fruticosa*.

Furthermore, studies on *Alamanda cathartica* (Singhe,1980) and *Memecylon umbellatum* (Senarathne and Yakandawala, 2004), soft wood cutting recorded the highest rooting percentage.

Effect of different rooting media on shoots formation of Bougainvillea stem cuttings Semi hard wood stem cuttings produced the highest number of shoots (5.1) in the M2 (Coir dust + Sand). The second highest number of shoots was produced in hard wood stem cuttings planted in the rooting medium 2 (Coir dust + Sand). Hard wood stem cuttings in the rooting media M1 and M2 produced equal number of shoots accounted to 3.26. The lowest numbers of shoots were produced by soft wood stem cuttings planted in the M1 (Coir dust + Compost + Sand). It was clear from the study that semi hard wood cuttings of *Bougainvillea* produced the highest number of shoots (Table 3).

Table 3: Effect of different treatments on number of shoots of Bougainvillea stem cuttings

	M1	M2	M3
SHW	2.95± 0.18	5.13± 0.46	3.16± 0.20
SW	1.38± 0.11	2.10± 0.43	1.96± 0.24
HW	3.26± 0.15	3.28± 0.29	3.26± 0.05

Note: Measurements are the means of 15 replicates ± SE. SHW - Semi hard wood, SW - Soft wood, HW - Hard wood, M1 - Coir dust + Sand + Compost, M2 - Coir dust + Sand, M3 - Coir dust only

Effect of different rooting media on height of Bougainvillea plants

Semi hard wood cuttings planted in the medium M2 were significantly taller (10.1 cm) compared to all the other treatments while soft wood cuttings produced shorter plants than all the other types of cuttings irrespectively of rooting medium. Height of the hard wood cuttings planted in the medium M1, M2 and M3 were 9.44 cm, 8.58 cm and 8.80 cm respectively (Table 4).

Table 4: Effect of different treatments on plant height (cm)

	M1	M2	M3
SHW	9.69 ± 0.30	10.1 ± 0.51	8.73± 0.38
SW	6.62± 0.29	6.59 ± 0.00	6.70± 0.20
HW	9.44 ± 0.15	8.58 ± 0.11	8.80± 0.15

Note: Measurements are the means of 15 replicates ± SE. SHW - Semi hard wood, SW - Soft wood, HW - Hard wood, M1 - Coir dust + Sand + Compost, M2 - Coir dust + Sand, M3 - Coir dust only

Experiment II

Effect of fertilizer on flowering of rooted cuttings

The highest number of flowers were recorded from treatment 5 (Maxicrop + NPK 6:30:30) while the lowest number flowers were recorded in treatment 3 (Maxicrop + NPK 10:52:10). There was no significant difference ($p>0.05$) between treatment 1 (Maxicrop), treatment 2 (10:11:18 N: P: K + Maxicrop), treatment 3 (Maxicrop + NPK 10:52:10), and treatment 4 (Maxicrop + NPK 20:20:20) (Table 5). Flowering is a complex process in plant life for which the plants requires optimum growth and nutrients and thus the media containing more nutrients, produced higher number of flowers. More Dahlia flowers were obtained previously in Sand + Silt + Leaf mould (Kiran et al., 2007). The similar results were observed by Ahmad (1989) who reported that the number of flowers was higher for roses grown in the mixture of leaf mold garden soil and sand.

Correlation Analysis

When Linear Correlation Analysis was performed for number of leaves (NL), number of roots (NR), number of shoots (NS), length of shoots (LS), length of roots (LR) and plant height (HP), a highly

significant positive correlation ($p < 0.0001$) was observed with flowering. The number of flowers (NF) showed a significant negative correlation ($p = 0.05$) with number of leaves (NL) and plant height (PH). On the other hand, there was no significant relationship ($p = 0.05$) between number of roots (NR), number of shoots (NS), length of shoots (LS) and length of the roots (LR) with number of flowers (NF) of *Bougainvillea spectabilis* (Table 6)

Table 5 Effect of different fertilizer applications on number of flowers per plant during the study period

Fertilizer	Treatments	51DAP	61DAP	71DAP	81DAP
Maxicrop (MC)	T1	4.00 ^b	5.33 ^b	3.66 ^b	2.66 ^c
MC + 10:11:18	T2	5.00 ^b	6.00 ^b	3.00 ^b	4.33 ^{cb}
MC + 10:52:10	T3	2.66 ^b	4.00 ^b	3.66 ^b	5.00 ^{cb}
MC + 20:20:20	T4	2.67 ^b	5.00 ^b	5.33 ^b	6.33 ^b
MC + 6:30:30	T5	9.00 ^a	9.66 ^a	12.00 ^a	12.66 ^a

Note: Means with the same letter/s are not significantly different at $p = 0.05$

Table 6: Linear Correlation Coefficient between growth parameters of number of leaves, number of roots number of shoots, length of roots, length of roots and number of flowers for the overall data set

	NL	NR	NS	LS	LR	PH	NF
NL	-	-	-	-	-	-	-
NR	0.97***	-	-	-	-	-	-
NS	0.94***	0.95***	-	-	-	-	-
LS	0.71***	0.70***	0.68***	-	-	-	-
LR	0.95***	0.92***	0.94***	0.75***	-	-	-
PH	0.69***	0.60***	0.63***	0.47***	0.64***	-	-
NF	-0.12*	-0.08 ^{ns}	-0.04 ^{ns}	0.08 ^{ns}	-0.04 ^{ns}	-0.11*	-

Note: ***= Significant at $p < 0.0001$, *Significant at $p < 0.05$; NS= non significant NL- number of leaves, NR- number of roots, NS- number of shoots, LR- length of root, LS- length of shoots, PH- Plant height NF- number of flowers

CONCLUSION

The best maturity stage of cuttings of *Bougainvillea spectabilis* for efficient rooting was Semi hard wood while Semi hard wood cuttings performed best in the Coir dust + Sand (1:1) rooting medium.. Optimum flowering frequency of *Bougainvillea spectabilis* plants was observed when they are grown on Sand + Coir dust in equal proportions with the application of Maxicrop + NPK (6:30:30) at weekly intervals.

REFERENCES

- Ahmad, K.K., (1989). Effect of different potting media on different rose cultivars under plastic tunnel, M.sc. Hons Thesis, NWFP. Agriculture University, Pesawar, Pakistan.
- Celine S., Luc N., Thierry B., Helene C., Marie-Pierre J., Marlene D., Goran S., Michel Z., Catherine B., (2006). Proteomic Analysis of Different Mutant Genotypes of Arabidopsis Led to the Identification of 11 Proteins Correlating with Adventitious Root Development. Plant Physiol. 140: 349 – 364.

- De Klerk G.J., Van Der Krieken W.M., De Jong J.C. (1999). The formation of adventitious roots; new concepts, new possibilities. *In Vitro Cell Dev Biol.* 35: 189- 199.
- Hackett, W. P., Scachs, R. M. and Debie J. (1972). Growing Bougainvillia as a flowering pot plant. *California. Agriculture.* 26 (8): 12 – 13.
- Hammad, I. (2009). Genetic variation among *Bougainvillea glabra* cultivars (Nyctaginaceae) detected by Rapid markers and Isozymes patterns. *Journal of Agriculture and Biological sciences* 5 (1): 63-71.
- Hudsen, T.H., E. K., Dale, T.D., Fred and L. G. Robert (1997). Techniques of propagation by cuttings. *Plant propagation principles and practices.* eds, J. Williams, C. Cobb and B.Cappuccio, 329 – 338.
- Kiran, M, Baloch, J.U.D. and Waseem, M. (2007). Effect of different growing media on the grown and development of Dahilia (*Dahlia pinnata*) under the Agro-climatic condition of Dera Ismail Khan. *Department of Horticulture, Faculty of Agriculture, Gomal University* 10 (20): 4140 – 4143.
- Kobayashi, D.K., McConnell, J., Griffis, J. (2007). Bougainvillea. Available from the Department of Tropical Plant and Soil Sciences, published by the College of Tropical Agriculture and Human Resources.
- Senarathne, M.P.I. and Ykandawala, K. (2004). Propagation and Landscape Potential of Korakaha (*Memecylon umbellatum* Burm. F). *Faculty of Agriculture and Plantation Management, Wayamba University,* 326 – 336.
- Singhe, S.P. (1980). Mist Propagation of *Allamanda cathartica* by different types of stem cuttings, *Plant Science,* 42 – 43.

Identification of a suitable fertilizer combination for commercial cultivation of *Dracaena sanderiana* cv. 'White'

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ABSTRACT

Dracaena sanderiana is a widely cultivated floricultural crop on commercial scale in Sri Lanka. It is mainly cultivated to obtain un-rooted top cuttings for exports to the international market. This study was carried out to find out a good fertilizer combination for the best vegetative growth and good quality products of *D. sanderiana* cv. 'white'. Six combinations of treatments, AS+TSP+SOP-(N:P:K-3:1:2), Urea+TSP+MOP-(N:P:K-3:1:2), AS+TSP+MOP-(N:P:K-3:1:2), AS+TSP+SOP-(N:P:K-3:1:2) + Organic Liquid Fertilizer, N : P₂O₅ : K₂O : MgO -16% : 9% : 18% : 2% and AS+TSP+SOP-(N:P:K-3:1:2) + Dolomite were arranged in a CRD. The crop was grown for 6 months and plant height (cm) number of leaves per plant, average leaf length (cm) and average leaf area (cm²) were measured at monthly intervals. Quality of the crop after six month was also evaluated. Data were analyzed statistically using Minitab. *D. sanderiana* cv. 'white' provided with the fertilizer combination N : P₂O₅ : K₂O : MgO -16% : 9% : 18% : 2% resulted in significantly higher plant height while the lowest height was observed in plants provided with the AS+TSP+MOP-(N:P:K-3:1:2) fertilizer combination. Furthermore, the highest number of leaves per plant, highest leaf length and leaf area were also recorded in plants provided with the fertilizer combination N : P₂O₅ : K₂O : MgO -16% : 9% : 18% : 2% and the lowest was from the AS+TSP+MOP-(N:P:K-3:1:2) fertilizer combination. In quality analysis, plants provided with fertilizer combination AS+TSP+SOP-(N:P:K-3:1:2) + Organic Liquid Fertilizer received the highest score. Therefore it is concluded that N : P₂O₅ : K₂O : MgO -16% : 9% : 18% : 2% was the most suitable fertilizer combination for vegetative growth of *D. sanderiana* cv. 'white' at a commercial scale cultivation and organic liquid fertilizer is recommended to increase of quality of plants.

Keywords : *Dracaena sanderiana* cv.white, suitable fertilizer combination, vegetative growth

INTRODUCTION

Dracaena sanderiana L. is a popular foliage plant commercially grown in many parts of the island to produce several products for the export market. It is mainly grown to produce un-rooted and rooted top cuttings with several lengths for export trade. In 2014 Sri Lanka earned 14.86 US \$ million by exporting cut foliage and it represented 56.5 % of the total income from floriculture export products (Anon., 2014). Gampaha is a prominent district in commercial cultivations of *D. sanderiana*. Annual rain fall in the Gampaha district is about 2500 mm and average temperature is about 27.9 °C (Anon., 2015). Approximately 80 acres of new land were brought under cultivations of *D. sanderiana* in the Gampaha district through a project implemented by the Department of National Botanic Gardens in 2012/2103 and the highest percentage of the *D. sanderiana* cultivations was represented by *D.*

sanderiana cv. 'White' (Subasinghe *et al.*, 2014) Therefore *D. sanderiana* cv. 'White' was selected for this experiment. The first harvest of *D. sanderiana* is expected at 6 month after planting and 2 or 3 harvested cuttings are expected within a year. Therefore farmers try out various fertilizer combinations to fulfill their targets. This study was carried out to find out the most effective fertilizer combination among six fertilizer combinations identified at the field and used by farmers for commercial *D. sanderiana* cultivations for the best vegetative growth and good quality products.

MATERIALS AND METHOD

The experiment was conducted in a 60 % shade house at the Botanic Gardens, Gampaha from December 2013 to June 2014. Rooted cuttings (15 cm in length) of *D. sanderiana* cv. 'white' were planted in plastic pots (15 cm diameter and 15 cm height) filled with potting medium consists of top soil: sand: coir dust -3:1:1 ratio (volume basis). Fertilizer application was started one week after planting. Seven different fertilizers, Urea, Ammonium sulphate (AS), Tripple super phosphate (TSP), Muriate of potash (MOP), sulphate of potash (SOP), Organic Liquid Fertilizer and dolomite were used to form fertilizer combinations. Six fertilizer combinations were, Treatment 1 (T1)- AS+TSP+SOP-(N:P:K-3:1:2), Treatment 2 (T2) -Urea+TSP+ MOP-(N:P:K-3:1:2),Treatment 3 (T3) – AS+TSP+MOP-(N:P:K-3:1:2),Treatment 4 (T4) – AS+TSP+SOP-(N:P:K-3:1:2) + Organic Liquid Fertilizer, Treatment 5 (T5) - N : P₂O₅ : K₂O : MgO -16% : 9% : 18 % : 2% , Treatment 6 (T6) – AS+TSP+SOP-(N:P:K -3:1:2) + Dolomite. Treatment 1, which was frequently used by growers, was considered to be the control. Fertilizer combination used in T5 was a commercially available mixture and all the other fertilizer combinations were prepared by mixing each fertilizer according to the weight to obtain the correct N:P:K ratio. All treatments were arranged in a Complete Randomized Design with 6 replicates and each replicate consisted of 6 plants. Fertilizer application was done once a month. One plant was treated with 3 g of fertilizer mixture. The crop was grown for 6 months. Measurements taken were plant height (cm) at monthly intervals, number of leaves per plant after 6 months, average leaf length (cm) and average leaf area (cm²) after 6 months. A panel evaluation was arranged at the Botanic Gardens, Gampaha to asses quality of plants belong to deferent treatments at 6 month after planting. Data were analyzed statistically by ANOVA using Minitab statistical package. Mean separation was done by Turkey Pairwise Comparisons test and non-parametric data were analyzed using Kruskal-Wallis Test.

RESULTS AND DISCUSSION

Plant height

Table 1 shows that the highest plant height (55.46 cm) at the 6 month after planting when plants were provided with the fertilizer combination (T5) N : P₂O₅ : K₂O : MgO -16% : 9% : 18 % : 2%. The lowest plant height (39.35 cm) was in plants provided with the AS+TSP+MOP-(N:P:K-3:1:2) fertilizer combination T3, and T1 (AS+TSP+SOP-N:P:K-3:1:2). Plant height rapidly increased after the third month in T5 and T4 (Fig. 1).

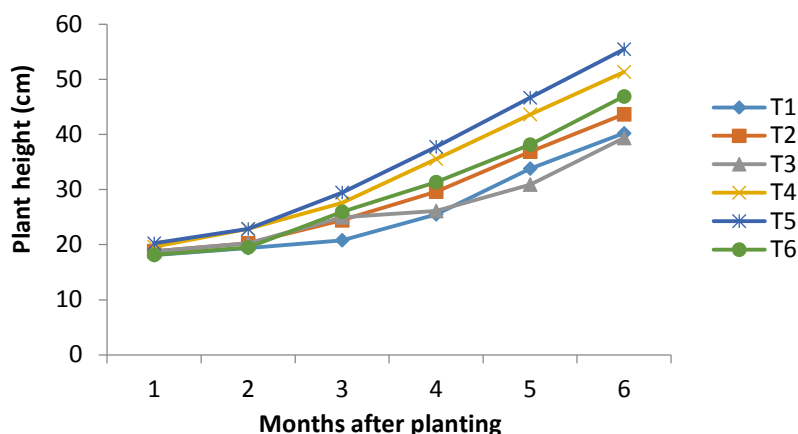


Figure 1: Increase of plant height with time

Table 1: Mean values of plant height, number of leaves, leaf length and leaf area of *D. sanderiana* in different fertilizer treatments at 6 month after planting.

Treatments	Plant height (cm)	Number of leaves/Plant	Leaf length (cm)	Leaf area (cm ²)
Treatment1(T1) – AS+TSP+SOP-(N:P:K-3:1:2),	40.21 ^e	18.88 ^d	19.59 ^{ab}	49.60 ^{ab}
Treatment2-(T2)- Urea+TSP+MOP – (N:P:K-3:1:2)	43.65 ^d	21.76 ^c	21.72 ^a	50.46 ^a
Treatment3(T3)– AS+TSP+MOP-(N:P:K-3:1:2)	39.35 ^e	17.86 ^d	18.51 ^b	47.26 ^b
Treatment4(T4) – AS+TSP+SOP-(N:P:K-3:1:2) + Organic Liquid Fertilizer	51.34 ^b	25.65 ^{ab}	21.56 ^a	49.64 ^{ab}
Treatment 5 (T5) –N : P ₂ O ₅ : K ₂ O : MgO - 16% : 9% : 18 % : 2%	55.46 ^a	27.91 ^a	22.57 ^a	52.35 ^a
Treatment6(T6) – AS+TSP+SOP-(N:P:K -3:1:2) + Dolomite	46.89 ^c	23.17 ^{bc}	20.87 ^{ab}	50.18 ^{ab}
R ² (adj.)	96.80 %	88.61 %	74.27 %	96.23 %

*Means that do not share a letter along the column are significantly different at P=0.000.

Number of leaves per plant

The highest number of leaves per plant was observed in plants treated with T5 (N : P₂O₅ : K₂O : MgO -16% : 9% : 18 % : 2%) but was not significantly different from plants treated with T4 (AS+TSP+SOP-N:P:K-3:1:2) + Organic Liquid Fertilizer). The lowest number of leaves per plant was observed in plants treated with T3 (AS+TSP+MOP-N:P:K-3:1:2).

Leaf length and leaf area

The highest leaf length and leaf area were recorded in plants treated with T5, but was not significantly different from T1, T2, T4, and T6. The lowest leaf length and leaf area were observed in plants treated with T3.

The best performance of all the parameters were recorded in plants treated with T5 (N : P₂O₅ : K₂O : MgO -16% : 9% : 18 % : 2%) which is a commercially available fertilizer mixture. This may be due to

the product consisting of Urea, Di-Ammonium phosphate (DAP), Sulphate of ammonia, Muriate of potash (MOP) and Dolomite.

Quality analysis of plants

Scores obtained for plants of different treatments in the quality evaluation was statistically analyzed and significant differences (Kruskal-Wallis Test, $P=0.000$) were obtained between the scores (Table 2).

Table 2: Quality analysis of plants (Kruskal-Wallis Test, $P=0.000$)

Treatments	Median	Ave Rank	Z
Treatment 1 (T1) - AS+TSP+SOP- (N:P:K-3:1:2),	84.13	23.5	0.43
Treatment 2- (T2)- Urea+TSP+MOP – (N:P:K-3:1:2)	66.10	9.2	- 2.66
Treatment 3 (T3) – AS+TSP+MOP- (N:P:K-3:1:2)	74.44	15.5	-1.29
Treatment 4 (T4) – AS+TSP+SOP- (N:P:K-3:1:2) + Organic Liquid Fertilizer	93.84	38.2	3.59
Treatment 5 (T5) – N : P ₂ O ₅ : K ₂ O : MgO -16% : 9% : 18 % : 2%	90.63	32.0	2.26
Treatment 6 (T6) – AS+TSP+SOP- (N:P:K -3:1:2) + Dolomite	87.86	28.3	1.47

The quality analysis indicated that plants treated with T4 – AS+TSP+SOP-(N:P:K-3:1:2) + Organic Liquid Fertilizer obtained highest average rank and Z value. Plants treated with T2- Urea+TSP+MOP – (N:P:K-3:1:2) received the lowest average rank and Z value. It is evidenced that the quality of plants had increased with the use of organic liquid fertilizer. This may be due to trace elements in the organic liquid fertilizer.

CONCLUSION

Commercially available mixture of N : P₂O₅ : K₂O : MgO -16% : 9% : 18 % : 2% was the best fertilizer mixture among the fertilizer combinations used by growers in the Gampaha district for a good vegetative growth of *D. sanderiana* cv. 'white' to obtain the required length to harvest top cuttings for export market at sixth month after planting. Organic liquid fertilizer or other fertilizer containing trace elements along with the main fertilizer is necessary to increase the quality of harvesting products of *D. sanderiana* cv. 'white'.

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REFERENCES

- Anon. (2105) Floriculture Export Performance. Export Development Board, Sri Lanka [on line]. [Accessed on 02.12.2015] Available at <http://www.srilankabusiness.com/floriculture/floriculture-export-performance.html>
- Anon. (2015). Wikipedia, the free encyclopedia [on line]. [Accessed on 02.12.2015] Available at <https://en.wikipedia.org/wiki/Gampaha#Climate>
- Srikrishnah, S., Peiris, S.E., and Sutharsan, S. (2011). Influence of shading on growth and quality of *Dracaena* varieties (*Dracaena sanderiana* L.). Proceedings of the National Symposium on Floriculture Research. 1-5.
- Subasinghe, S.A.A.U., Wickramasinghe, M.C. and Krishnarajah, S.A. (2014) Survey on the foliage Cultivations in the Gampaha District after implementation of Divi Naguma Programme by Department of National Botanic Gardens. Proceedings of the National Symposium on Floriculture Research.

***In-vitro* establishment of *Cassia fistula* L.**

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ABSTRACT

Experiments were carried out to develop a protocol to establish *Cassia fistula* under *in vitro* conditions to produce a large number of uniform planting materials. *C. fistula* is mainly propagated by seeds. However, germination is low due to the hard seed coat and low seed quality. Because of these reasons, required seedling demand could not be achieved. Micropropagation provides a best tool for large scale production of planting materials. Yet a protocol of *in vitro* propagation of *C. fistula* does not exist. Therefore, a method for *in vitro* propagation of *C. fistula* is required to make this attractive tree available for landscaping. A protocol to establish *C. fistula* sterile *in vitro* cultures using nodal explants collected from a field grown mother plant was investigated. The nodal segments were exposed to 0, 0.5, 1, and 2% concentrations of Redoaxyl Metalaxil™ fungicide solutions with 20 and 40 minute duration. Thereafter, it was sterilized with 10, 20, and 30% concentrations of sodium hypochlorite two times in 10 minute duration. The sterilized nodal explants were established *in vitro* on Murashige and Skoog's (MS) medium supplemented with 1.5 mgL⁻¹ BAP. *C. fistula* explants exposed to 1% fungicide for 40 minutes and sterilized with 30% sodium hypochlorite twice in 10 minute duration produced the highest percentage (60%) of contamination-free cultures in *in vitro* establishment.

Keywords: *Cassia fistula*, contaminations, *in vitro* establishment, nodal explant

INTRODUCTION

Cassia fistula L., (Golden Shower) belongs to Fabacea family, is native to tropical regions of Asia. It is naturally distributed across India, Indo-China, Malaysia, Thailand and Sri Lanka and considered as the national tree and national flower of Thailand (Rocas, 1969).

C. fistula L. is widely used for landscaping as a specimen plant and shade tree around houses, on the edges of roads, and in the streets, parks, and gardens of towns. (Edward *et al.*,1993). It is a perfect tree for city landscaping and for air pollution termination (Davison, 2004). *C. fistula* is the most beautiful among the cassia spices because of its pendulous showy flowers. It is an excellent tree for the tropical and subtropical landscaping projects. Therefore, the demand for *C. fistula* as an ornamental tree is very high than other cassia species. As the demand for *C. fistula* increases, growers try to increase their production. Good quality healthy planting materials are required for continuous production. *In vitro* propagation is the best solution to meet increasing demand by producing large number of good quality new plants in a short time.

The trees grow well in dry climates, under full sunlight on well-drained soil. It is moderately drought and salt-tolerant and it has strong, heavy and very durable, hard wood. Hence it provides good quality timber also.

Medicinally it has various pharmacological activities such as anti-microbial, antifungal, antipyretic, analgesic, laxative, anti-inflammatory, antioxidant, anti-tumor, antineoplastic, hepatoprotective and hypoglucemic activities (Neelam et al., 2011). Therefore, demand for this as a herbaceous plant has been increased highly.

C. fistula seeds are poisonous. Therefore, distribution of seeds by animals is restricted. It is generally propagated only by seeds. However, germination is difficult due to the hard seed coat and low seed quality. Dormant seeds are also commonly found. Therefore, methods for rapid micropropagation and genetic improvement are urgently required.

No comprehensive micropropagation reports are available for *in vitro* regeneration of *C. fistula*. Hence, the development of a protocol to establish *C. fistula* under *in vitro* condition is a timely requirement.

MATERIALS AND METHODS

The experiments were carried out in the tissue culture laboratory of the Department of Crop Science, Faculty of Agriculture, University of Peradeniya. Planting materials were collected from the selected healthy mother plants that were located in Peradeniya. Single nodal segment of 1 cm length with an un-sprouted axillary bud was used as the explant for shoot initiation. They were thoroughly washed under tap water with few drops of liquid soap by rubbing. This process was repeated 3 times. Nodes were exposed to 0, 0.5, 1 and 2% concentrations of Redoxil Metalaxil™ for 20 or 40 minutes of time periods. After decanting the fungicide solution, nodes were washed with autoclaved water once. Thereafter, they were sterilized either with 10, 20, or 0% concentrations of sodium hypochlorite solution for 10 minute. Bleaching solution was decanted and washed three times with autoclaved water. This process was repeated with 10% bleach solution without Tween-20 for another 10 minute. These shoots were rinsed three times in sterile distilled water with each rinse lasting approximately one minute. One explant was cultured in one tube. Standard Murashige and Skoog's (MS) medium supplemented with 1.5 mg/L BAP and 30 g/L sugar, was used throughout the experiment. These explants were monitored for contamination and for shoot production. Data on number of uncontaminated explant and survival plants were recorded in each week. After six weeks, survival percentage was calculated.

RESULTS AND DISCUSSION

After observing the cultured explants for 6 weeks for growth and contamination, it was found that nodes exposed to one percent (1%) Redoxil Metalaxil™, for 40 minutes and sterilized with 30% sodium hypochlorite twice in two times 10 minute duration produced highest percentage (60%) of contaminations free cultures. According to the statistical analysis among 21 treatment combinations, 1% Redoxyl Metalaxil™ was significantly better than other concentrations. Increasing fungicide concentration above 1% produced more of dead shoots. According to the time level 40 minutes exposure for Redoxyl Metalaxil™ was more effective than 20 minutes for survival. Among the concentrations 30% showed somewhat effectiveness than others (Figures 1 and 2).

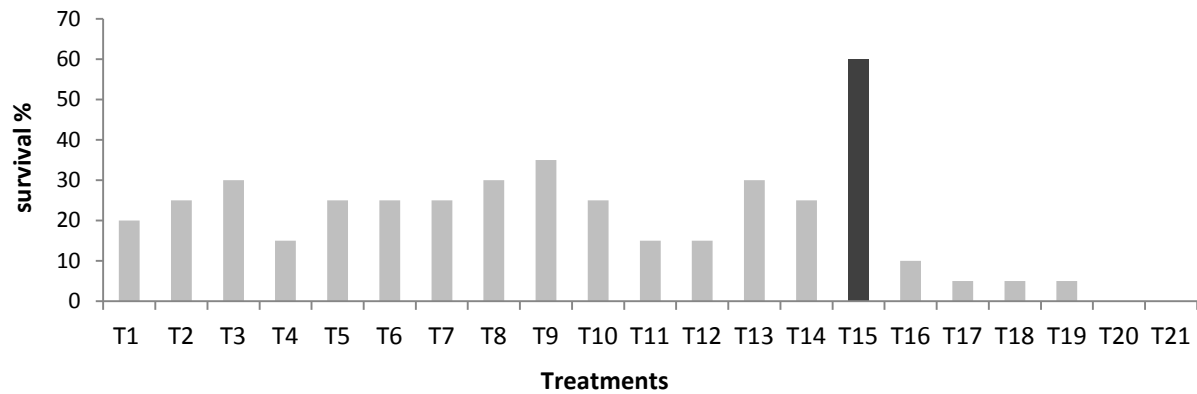


Figure 1: Effect of different concentration of fungicides and time period exposed under different bleach concentration on surface sterilization of *C. fistula* explants. The treatment combinations were identified as 0% fungicide with 0 time and 10, 20 and 30% clorox™ as T1, T2 and T3, 0.5% fungicide with 20 minutes and 10, 20 and 30% clorox™ as T4, T5 and T6, 0.5% fungicide with 40 and 10, 20 minutes and 30% clorox™ as T7, T8 and T9, 1% fungicide with 20 minutes and 10, 20 and 30% clorox™ as T10, T11 and T12, 1% fungicide with 40 minutes and 10, 20 and 30% clorox™ as T13, T14 and T15, 2% fungicide with 20 minutes and 10, 20 and 30% clorox™ as T16, T17 and T18, 2% fungicide with 40 minutes and 10, 20 and 30% clorox™ as T19, T20 and T21,

Treatment combinations with low concentration of fungicide and low level of bleach concentration showed a higher contamination percentage. Both bacterial and fungal contaminations were seen in cultured explants. Increasing fungicide concentration over 1%, showed an adverse effect on explants. Low levels of concentrations may be insufficient for surface sterilization of *C. fistula* nodes. Therefore, this might be the most effective combination for sterilization of the plant.

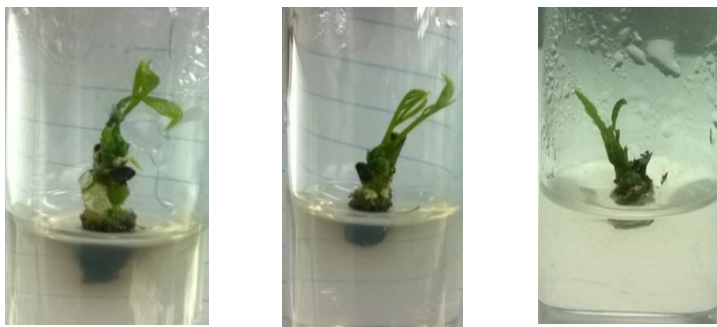


Figure 2: Response of axillary bud after five weeks of culturing

CONCLUSION

C. fistula exposed to one percent (1%) fungicide for 40 minutes and sterilized with 30% sodium hypochlorite two times of 10 minute duration produced the highest percentage (60%) of contaminations free cultures in *in vitro* establishment. This is a successful protocol for *in vitro* establishment *C. fistula* to produce large numbers of planting materials.

REFERENCES

- Amesh, K., Chandra, M.K. and Vijaya, T., (2009). *In vitro* Propagation and *Ex vitro* Rooting of *Tectona grandis* (L . f), APNBV-1 Clone. , *Botanical Review*. 25(2), pp.119–126.
- Anis, M. (2012). Advances in Micropropagation of a Highly Important Cassia species- A Review. *New Perspectives in Plant Protection*. 24:14-18
- Article, R., (2012). *In Vitro* Propagation of Medicinally Important Cassia Species – A Review. *Journal of Plant Biology*, 2(1), pp.1–11.
- Davison, E., (2004). *Cassia fistula* and *Delonix regia*. In: Trees for Future. 16(1), pp.1–8.
- Neelam, C., and Ranjan, B. (2011). Antimicrobial activity of *Cassia fistula* linn . *Legumes.*, 2(10), pp.100–102
- Pijut, P.M. (2012). *In vitro* propagation of tropical hardwood tree species - A review. *Propagation of Ornamental Plants*, 12(1), pp.25–51.

***Gypsophila paniculata*: a potential floricultural crop for Low Country Wet Zone**

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ABSTRACT

Baby's breath (*Gypsophila paniculata* L.) is a herbaceous perennial plant, often grown commercially as an annual crop. It is valued as a cut flower in floriculture industry and added as a filler to flower bouquets and several other floral arrangements. Although there is a high demand for this cut flower in the market throughout the country, cultivation is mainly confined to the upcountry areas. A study was carried out to explore the suitability of *G. paniculata* for Low Country Wet Zone (LCWZ) conditions through evaluation of growth and yield potential under protected cultivation. First experiment was conducted to identify the effect of Gibberellic acid (GA₃) on growth and yield of two commercial varieties; 'Million stars' and 'New love'. Results revealed that plant height was not influenced by the application of GA₃. In the 2nd experiment variety 'Million stars' was subjected to pinching, with and without application of GA₃. Results revealed that GA₃ and pinching individually affected the number of days to bloom. Mean number of days to bloom was extended with pinching by more than 2 weeks and application of GA₃ also delayed flowering by 7 days when compared to untreated plants. Pinching reduced the plant height, but increased the number of branches and flower yield significantly. The study revealed that both the varieties of *Gypsophila* can be successfully grown in LCWZ under protected house conditions.

Keywords: GA₃, growth, *Gypsophila paniculata*, pinching and yield

INTRODUCTION

Out of many *Gypsophila* species *Gypsophila paniculata* (family Caryophyllaceae) is the major one used in commercial cut flower production. The species is valued as a cut flower in floristry and widely used as a filler to fill the gaps with larger blooms especially with roses and carnations. In addition, it is also used as a dried flower. *Gypsophila* is popularly known as baby's breath due its soft odor.

It has a seasonal demand in Sri Lanka and catches high price during wedding seasons. The crop is mainly grown in up country areas under rain shelters. Growing crops under protected houses are becoming popular in other parts of the island with various national programmes implemented to uplift the rural livelihood. Introduction of potential crops for those houses is a timely needed activity for income generation and to make a greater contribution to economy. *Gypsophila* can be grown as a short term crop and planting can be planned to get the blooms in wedding season. However, effort has not been made to popularize *Gypsophila* cultivation in other areas of the country. Therefore, a study was conducted at Agriculture Research Station, Thelijjawila (LCWZ) to look for the possibility of introducing this important cut flower in to the area with following specific objectives; to identify a

suitable variety for low country wet zone area, to study the effect of GA₃ for growth and yield and to identify the combined effect of pinching and GA₃ application on growth and yield of *Gypsophila paniculata* L.

MATERIALS AND METHODS

Rooted cuttings of two *Gypsophila* varieties; 'New love' and 'Million stars' were kindly provided by Agriculture Research Station (Department of Agriculture) Sita Eliya, Nuwara Eliya.

Two factor factorial experiments were conducted inside an insect proof poly-tunnel in Complete Randomized Design at Agriculture Research Station, Thelijjawila during October to February, 2012/2013. Plants were maintained in the polytunnel under the following conditions. The average temperature inside the poly-tunnel was in the range of 29 – 30 °C in the morning (9.00 a.m.) and reached the maximum temperature of 35 °C at 1.15 p.m. The temperature was reduced to 29 - 32 °C at 4.00 p.m. whereas the average light intensity at plant level by 9.00 a.m., 1.15 p.m. and 4.00 p.m. were 17.2, 18.5 and 12.3 Klux respectively. Sand, compost and topsoil at 1:1:2 ratio (v/v) was used as the plating medium and pH was adjusted to 6.2 with Dolomite. The plating medium was treated with Thiophenate – methyl 50 % + Thiram 30 % WP 4 days prior to planting. Urea, Triple Super Phosphate and Muriate of Potash were mixed at 20:33:25 ratio w/w and 13 g from the mixture was added as the basal fertilizer and top dressing was done with urea and Muriate of Potash at 1:2 ratio at the rate of 2.2g/pot 1 month after planting. Plants were supported with sticks 3 weeks after planting to prevent stem damage due to bending. Pots were arranged in the polytunnel by giving 50 cm between rows and 30 cm between plants.

Experiment 1

In this experiment, effect of GA₃ on growth and yield parameters of two varieties; New love and Million stars was tested. Two forms of GA₃ (Pure GA₃ (Duchefa, G0907.0001) and commercial GA₃ containing 10% GA₃+6%Ca and 2%B) along with control were tested in the experiment which was laid as a 2 factor factorial design with 5 replicates. Four weeks after planting, the plants were sprayed with each formulation at the rate of 150 ppm which has been recommended in cultivation manual by Dan farm, Israel and KF Bioplants Pvt. Ltd, India (www.kfbioplants.com) to ensure uniform elongation of the shoots. Data were recorded on height of the plant (weekly, commencing from 1 week after planting to harvesting) and number of flowers/inflorescence.

Experiment 02:

This experiment was conducted with 2 weeks old rooted cuttings of variety; Million stars to study the effect of pinching and GA₃ application on growth and flower production characteristics. There were 4 treatment combinations with 5 replicates per treatment. GA₃ application and pinching were done in the morning before 9.00 a.m. one month after planting. Application of GA₃ and pinching were practiced only once during the crop growth. Height of the plant (weekly, from one week after spraying to fully formed inflorescence), number of branches/plant (weekly) and weight of the inflorescence were measured.

RESULTS AND DISCUSSION

Experiment 1

Data on plant height are illustrated in Table 1. It can be observed from the plant height data that the growth of main shoot is very rapid and it may be due to the high temperature prevailed under local conditions. One week after spraying of GA₃, plant height was in the range of 57-68 cm indicating that the plants are at bolting stage (rapidly growing main stem). Normally, Gypsophila is grown to a height 120 cm (<http://www.gardenershq.com>) and in this experiment plants have almost attained their full length 4 weeks after spraying of GA₃.

Table 1: Effect of different treatment combinations on plant height up to 4 weeks from spraying of GA₃

Treatment combination	Mean Plant height (cm)			
	1 week after spraying	2 weeks after spraying	3 weeks after spraying	4 weeks after spraying
New love + pure GA ₃	63.8	86.0	95.4	106.5
Million stars + pure GA ₃	58.72	74.8	90.08	100.7
New love + Nap Gibb	68.44	85.78	97.8	102.64
Million stars + Nap Gibb	61.72	76.8	89.02	98.92
New love + water	57.32	75.78	91.54	104.92
Million stars + water	64.3	82.74	93.5	100.1
CV%	13.39	14.07	7.95	7.47
	ns	ns	ns	ns

ns: Not significant at p = 0.05

However, the difference in the plant height among the treatment combinations was not significant in all 4 sets of data. Though it is known that gibberellins enhance elongation of internodes and plant growth in ornamental crops such as tulip (Rudnicki *et al.*, 1976) and dahlia (Khan and Tewari 2003) by increasing the cell division and enlargement, no pronounced effect was observed in the present study. The reason for not showing any effect of GA₃ on growth in the present study may be due to the fact that gibberellins (GAs) produce the effect of substituting the action of warm temperatures which are needed by the plant to respond to the inductive effect of long days (Shillo 1985). Therefore, the supply of GAs may not be required or its effect is hindered during warm months as plants are exposed to higher night temperatures when grown under LCWZ conditions. However, repeated application of GA₃ might lead to different results.

Table 2: Effect of variety on number of flowers/inflorescence

Variety	Mean no. of flowers/inflorescence
Million stars	2672.5 ^a
New love	1122.7 ^b
CV%	15.85

Different letters along the column shows significant difference at p = 0.05

Table 2 shows the results of the statistical analysis of data on number of flowers/ inflorescence. Number of flowers per inflorescence was significant only at variety level indicating that GA₃ has no effect on flower number. The significant difference between two varieties is attributed to the genetic variation. Variety Million stars produced greater no. of flowers than var. new love (Product

information, Amazon.com). In a review by Vieira *et al.*, (2010) they quoted the findings of Henny *et al.*, (1999) that GA application increased the number of flowers of *Syngonium podophyllum* Schott cv. 'White Butterfly' when treated with 80 ppm GA₃. However, no information was found on the temperature under which the study has been conducted. Application of gibberellins ensures flowering under cold temperatures (Shlomo *et al.*, (1985) especially when the night temperature is below 12 °C. Though GA₃ had no effect on flower number in the present experiment, it might have increased the flower yield in terms of weight which we have not assessed in this experiment.

Experiment 2

Statistical analysis of number of days to initiate flower bud opening was significant at both GA₃ treatment and pinching. Plants applied with GA₃ extended flower bud opening time by 7 days than untreated plants (Table 3). GA₃ is known to decrease in time to flower in ornamental crops such as rhododendron (Chang and Sung, 2000) and Aglaonema (Henny, 1983). However, the response of GA₃ can be varied with the concentration. As an example, GA₃ did not change the growth and flowering of chrysanthemum at low concentrations (Vieira, 2008). However, in the present study GA₃ delayed flowering and it may be attributed to weather conditions.

Pinching involves the removal of the head of the main stem at an early stage by breaking out the head of the cutting by bending at 8 to 10 pairs of leaves (internodes) on the plant. It is performed only once in the plants life cycle. Pinching is an essential operation in cultivation of gypsophila. If left unpinched, main stem continues to grow suppressing the emergence and elongation of the side shoots due to apical dominance. Therefore, pinching helps to balance the plant architecture with branches. Delay in flowering in pinched plants may be due to stimulated and extended vegetative growth due to removal of apical dominance (Table 4).

Table 3: Effect of GA3 on number of days to initiate flower bud opening

Treatment	Mean no. of days
With GA	76.2 ^a
Without GA	69.7 ^b
CV%	3.00

Different letters along the column shows significant difference at p = 0.05

Table 4: Effect of pinching on number of days to initiate flower bud opening

Treatment	Mean no. of days
With pinching	80.8 ^a
Without pinching	65.1 ^b
CV%	3.00

Different letters along the column shows significant difference at p = 0.05

Tables 5 shows that GA₃ application had no contribution towards plant height but pinching has a significant effect. The difference in plant height in all 4 sets of data was significant and at the beginning the difference between pinched and unpinched plant was 22 cm while at flowering it is about 18 cm.

Table 5: Effect of pinching on plant height up to 4 weeks from spraying

	Mean height of plant (cm)			
	1 week after spraying	2 weeks after spraying	3 weeks after spraying	4 weeks after spraying
Without pinching	64.3 ^a	88.6 ^a	100.75 ^a	110.5 ^a
With pinching	42.7 ^b	55.25 ^b	74.7 ^b	92.75 ^b
CV%	16.25	11.92	8.43	5.89

Different letters along the column shows significant difference at p = 0.05

Table 6: Effect of pinching on number of branches

Treatment	Mean no. of branches
Without pinching	0.1 ^b
With pinching	4.1 ^a
CV%	11.98

Different letters along the column shows significant difference at p = 0.05

Pinching induces formation of branches by removing apical dominance. The auxin inhibits the growth of lateral buds and so its removal enables the lateral buds to grow and the plant becomes fuller and bushier. Table 6 shows that number of branches was significantly different between two treatments. Unpinched plants hardly produced branches and the average is 0.1 branches per plant. Pinched plants produced 4.1 branches.

Table 7: Effect of pinching and GA₃ application on weight of the inflorescence cv. Million stars

Treatment combinations	Mean weight of flowers(g)/plant
Pinching + 150 ppm pure GA ₃	116.70 ^b
Pinching + Without GA ₃	119.06 ^a
Without pinching + 150 ppm pure GA ₃	115.32 ^b
Without pinching + Without GA ₃ (control)	107.3 ^c
CV%	4.53

Different letters along the column shows significant difference at p = 0.05

The yield of flowers was measured by weighing up to end of February through staggered harvesting. Table 7 shows that the weight of flowers was significantly affected by treatment combinations. The highest yield (119.06 g) was observed in pinched plants without GA₃ and the lowest yield (107.3 g) was recorded in control plants. GA₃ treated plants (with or without pinching) produced statistically similar yields. The number of panicles that could be harvested from pinched plants is higher than the unpinched plants due to higher number of branches. GA₃ has a marginal effect on increasing flower yield as the quantities of flowers were more than the untreated plants (control). Shlomo *et al.*, (1985) stated that GA₃ had insignificant promotive influence under fully inductive condition of long day and high temperatures. The results we obtained here are in par with their findings.

CONCLUSION

The results of our experiments revealed that both varieties of *Gypsophila* can be cultivated in Low Country Wet Zone under protected house conditions. Though application of GA₃ is not required, pinching can be recommended as it produces more number of branches and thereby increases the flower yield in cultivation of *G. paniculata*.

REFERENCES

- Chang, Y.S. and Sung, .F.H. (2000) Effects of gibberellic acid and dormancy breaking chemicals on flower development of *Rhododendron pulchrum*. Amsterdam. ScientiaHort. 83. P.331-337.
- Henny, R.J. (1983) Inducing *Aglaonema* to flower using Gibberallic acid treatment. Hortscience, Alexandria. 18. p. 374.
- Khan, F.U., and Tewari, G.N. (2003) Effect of growth regulators on growth and flowering of dahlia (*Dahlia variabilis* L.). Indian Journal of Horticulture, Bangalore. 2(60). p.192-194.
- Rudnicki, R.M., Nowak, J. and Saniewski, M. (1976) Effect of gibberellic acid on sprouting and flowering of some tulip cultivars. Amsterdam. ScientiaHort. 23.p. 387-397.
- Shlomo, E., Shillo, R. and Halevy, A.H. (1985) Gibberellin substitution for the high night temperatures required for the long-day promotion of flowering in *Gypsophila paniculata* L. ScientiaHort. 26.p.69-76.
- Shillo, R. (1985) *Gypsophila paniculata*, p. 83-87.In: A.H. Halevy (ed.). Handbook of flowering. vol. 3. CRC Press, Boca Raton, Florida.
- Vieira, M.R.S. (2008) Effect of gibberellic acid application on quality and biochemistry of chrysanthemum cv 'Faroe' (Máster in Agronomy) -Universidade Estadual Paulista, Botucatu. p. 134.
- Vieira, M.R.S., Lima, P.P., G, de Souza, V. and Alves, L.D.S. (2010) Use of gibberellin in floriculture. African Journal of Biotechnology 9(54). p.9118-9121.

Evaluation of plant extracts and essential oils for antibacterial activity against bacterial blight of Anthurium (*Anthurium andraeanum*)

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ABSTRACT

Bacterial blight caused by *Xanthomonas campestris* pv. *dieffenbachiae* has been a major economic concern among anthurium growers as the disease could cause substantial losses in anthurium. The disease is difficult to control because of the inoculum persisting in the field in the form of symptomless infection. Control of the disease is heavily depend upon the use of chemical pesticides, however misuse of synthetic chemicals cause health hazards on humans and animals, environmental degradation and resistance development. Hence, there is an urgent need to develop new, cheap, eco-friendly alternative methods of disease control. Thus, this study was aimed to evaluate antibacterial activity of plant extracts of Aloe (*Aloe vera*), Clove (*Syzygium aromaticum*), Lemon grass (*Cymbopogon citratus*), Neem (*Azadirachta indica*), Onion (*Allium cepa*), Turmeric powder (*Curcuma longa*) and essential oils of Citronella (*Cymbopogon nardus*), Neem (Neemox: i.e. a commercial product containing neem oil) were evaluated against *X. campestris* pv. *dieffenbachiae*. Plant extracts were obtained by macerating plant materials with water in a juice extractor. All solutions were mixed with surfactant (Teepol®) before spraying. *Anthurium andraeanum* cultivar "Akapana" was used as test variety. Spraying was started 7 days after artificial inoculation and five sprays were given with weekly intervals. Eight treatments as well as Mancozeb and water which were used as standard treatment and control were replicated 8 times. Percentage of infection was recorded before each spray and 7 days after fifth spray, the disease index was calculated. Data were subjected to analysis of variance (ANOVA) with mean separation by Tukey's test at 5% levels of significance using Minitab 17 statistical package. Results showed that lemon grass extracts and citronella oil were highly effective (90%) against bacterial blight compared to the control after 5 rounds of spraying. Next most effective treatment which had shown 88.8% control of the disease was neem leaf extract. Clove leaves extracts, onion extracts, turmeric powder and Neemox also effectively reduced the growth of bacteria and mean reduction percentages were 77.9%, 76.8%, 75.8% and 74.3% respectively. Treatments with mancozeb proved to be 72.7% effective and aloe extract was less effective (50.4%) against *X. campestris* pv. *dieffenbachiae*. This experiment showed that there is a potential for using plant extracts and essential oils as antibacterial agents considering their availability and environment friendliness when compared with synthetic chemicals.

Keywords: *Anthurium andraeanum*, *Xanthomonas campestris* pv. *dieffenbachiae*, plant extracts, essential oils

INTRODUCTION

Anthurium belongs to the family Araceae which includes more than 100 genera and 1500 species from the tropics. Anthurium cut flowers and potted plants have a growing demand both in local and global markets. Anthurium growers have been struggling with bacterial blight caused by

Xanthomonas campestris pv. *Dieffenbachiae* (Alvarez *et al.*, 2006; Higaki *et al.*, 1995; Nishijima *et al.*, 1985) which has been a major economic concern in Sri Lanka as well. Most anthurium cultivars are susceptible to bacterial blight, making it very difficult to manage the disease once introduced to a production area. Infected tolerant cultivars may provide inoculum for a long period if left in the field undetected and eventually succumb to the disease (Fukui *et al.*, 1999). Bacteria enter the leaf margins via pores (hydathodes) where guttation droplets form. The first visible symptoms of the disease are chlorotic, water soaked lesions along the leaf margins that grow rapidly to form necrotic V-shaped lesions. As the disease progresses, more leaf tissue is killed and systemically infected plants turn dark brown and the growing point deteriorates leading to death of the plant (Norman & Ali, 2006 and Nishijima *et al.*, 1985).

Good cultural practices help to reduce losses, but they are insufficient to control bacterial blight. At present quick and effective management of bacterial blight in anthurium is generally not achieved even with the use of synthetic pesticides. However, the continual and indiscriminate application of agro-chemical pesticides has caused health hazards on living beings and environment degradation due to their residual toxicity and accumulation in the food chain. Many pathogenic microorganisms and insect pests have developed resistance against chemical pesticides (Williams & Heymann, 1998). Therefore most farmers are reluctant to use commercial agrochemicals and there is an urgent need to develop new, cheap, safe, eco friendly and effective alternative methods to control bacterial blight in anthurium. Thus, this study aimed to develop an eco-friendly alternative to control anthurium blight with the use of plant extracts and essential oils which are able to produce anti bacterial substances.

MATERIALS AND METHODS

An experiment was conducted in a glass house of Botanic Gardens, Gampaha in 2015. Cultivar "Akapana" was selected as the test variety. Five-month old healthy tissue culture-derived anthurium plants were used for the trial. The pathogen was isolated from leaves with symptoms of water soaked spots at leaf margins bordered by chlorotic or necrotic zones in PDA media. After confirming the cultures, a bacterial suspension was prepared by incubating purified cultures for 24 hours by suspending the bacteria in sterile distilled water. Pre-injured second youngest leaves were used for inoculation since the youngest leaves were at different developmental stages. Plants were inoculated by rubbing leaves with cotton swabs dipped in to a suspension of the bacteria (Sathyanarayan *et al.*, 1998) and they were arranged in a glass house with completely randomized design (CRD).

Six plant extracts and two essential oils were tested against *X. campestris* pv. *dieffenbachiae* as shown in the Table 1. Plant extracts were obtained by macerating plant materials with water in a juice extractor. All solutions were diluted to the concentrations as given in Table 1, and they were mixed with surfactant (Teepol) before each spraying to make an emulsion.

Other additional treatments were the control (plants spray with water) and Mancozeb which were used as standard treatment to compare the effects of plant extracts and essential oils over the synthetic chemical. Spraying was started seven days after inoculation, as the symptoms appeared as small lesions around pinpricks on leaves and five sprays were given at weekly intervals. Scoring was

done using a disease scale given in Table 2. Percentage of infection was recorded before each spray and after the fifth spray, and disease index (DI) was calculated using the following formula.

$$DI = \frac{\text{Sum of individual scores}}{\text{Total leaves observed} \times \text{Maximum score}} \times 100$$

Table 1: Plant extracts and essential oils and their concentrations to be tested

Common name	Botanical Name	Family	Part to be tested	Concentration
Plant extracts				
Aloe	<i>Aloe vera</i>	Aloaceae	Leaves	100g/l
Clove	<i>Syzygium aromaticum</i>	Myrtaceae	Leaves	250g/l
Lemon grass	<i>Cymbopogon citratus</i>	Poaceae	Leaves	250g/l
Neem	<i>Azadirachta indica</i>	Meliaceae	Leaves	250g/l
Onion	<i>Allium cepa</i>	Liliaceae	Bulb	100g/l
Turmeric	<i>Curcuma longa</i>	Zingiberaceae	Powder of rhizome	1.5g/l
Essential oils				
Citronella	<i>Cymbopogon nardus</i>	Poaceae	Oil	2ml/l
Neem; (Neemox)	<i>Azadirachta indica</i>	Meliaceae	Commercial product contains neem oil	30ml/l

The experiment consisted of 10 treatments with 8 replicates per treatment and in each treatment, individual plants were arranged in a Completely Randomized Design (CRD). Data were subjected to analysis of variance (ANOVA) using Minitab 17 statistical package and comparisons of mean values were analyzed by Tukey's multiple range tests at 5% level of significance.

Table 2: Description of the disease score and infection percentage

Disease scale	Infection (%)	Extent of disease development
0	0	No infection
1	1-5	Lesions as pinpricks
2	6-25	Lesions as pinpricks along with yellowing of 1-2 leaves
3	26-50	Lesion of size 1.2×0.5cm along with yellowing of 1-2 leaves
4	51-75	Yellowing of all leaves with blackening of petiole of leaves
5	76-100	Complete death of the plant

(Dhanya & Mary, 2006)

RESULTS AND DISCUSSION

Results showed a significant difference among treatments ($p < 0.0001$), calculated disease index 7 days after inoculation at weekly intervals. As shown in Table 3, the reduction of development of *X. campestris pv dieffenbachiae* in anthurium compared with non treated control and standard treatment.

Results showed that lemon grass extracts and citronella oil demonstrated to have best antibacterial activity from the beginning and both controlled the disease by 90% compared to the control after 5 rounds of spraying. Efficacy of citronella and lemon grass in the present study confirms the results obtained by Lucas *et al.*, 2012. It is proved that essential oils of clove, citronella, tea tree, and lemongrass reduced disease severity of bacterial spot caused by *Xanthomonas vesicatoria* in tomato by causing severe damage to the structure of bacterial cell. Several studies had shown that active

compounds of oils act directly on the pathogen or induce host resistance through production of phytoalexins, synthesis of structural compounds, and biochemical plant defense (Lucas *et al.*, 2012).

Table 1: Effect of spraying plant extracts, essential oil and fungicide on management of bacterial blight of anthurium (*Anthurium andraeanum*)

Treatments	Disease index before each spray and week after final spray					
	0	1	2	3	4	5
Aloe extract	3.87 ^{ab}	4.62 ^b	5.62 ^b	7.53 ^b	9.5 ^b	12.2 ^b
Clove leaves extract	3.12 ^{cd}	3.93 ^c	4.75 ^c	5.25 ^c	5.42 ^c	5.42 ^d
Lemmon grass extract	2.43 ^f	2.56 ^e	2.56 ^d	2.56 ^d	2.3 ^d	2.4 ^e
Neem leaves extract	2.82 ^{de}	2.94 ^{de}	2.94 ^d	3.0 ^d	2.8 ^d	2.74 ^e
Onion extract	3.06 ^{cd}	3.67 ^c	4.42 ^c	4.56 ^{bc}	5.03 ^d	5.7 ^d
Turmeric powder	3.56 ^{ab}	4.25 ^{bc}	5.2 ^c	5.39 ^{bc}	5.46 ^c	5.95 ^d
Citronella oil	2.62 ^{ef}	2.69 ^e	2.69 ^d	2.69 ^d	2.52 ^d	2.42 ^e
Neemox	3.37 ^{bc}	4.12 ^{bc}	4.62 ^c	5.56 ^{bc}	5.74 ^c	6.32 ^c
Mancozeb	3.37 ^{bc}	3.56 ^{cd}	4.44 ^c	5.2 ^{bc}	6.06 ^c	6.7 ^c
Control (water)	4.06 ^a	5.66 ^a	7.37 ^a	10.2 ^a	16.2 ^a	24.6 ^a

Values followed by the same letter in each column are not significantly different at $p = 0.05$

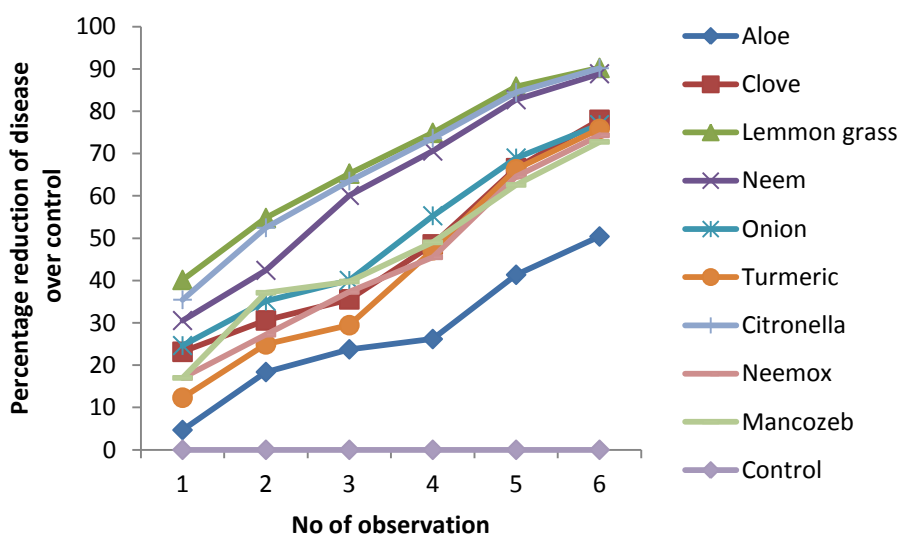


Figure 1: Effect of plant extract on percentage reduction of disease over control

After the second spray, neem leaf extracts also showed significant disease reduction (Figure 1) and 88.8% pathogen control of the disease was shown over the control treatment after five sprays. "Azadirachtin" containing in neem (*A. indica*) has antibacterial properties and Opara and Wokocho (2008) also reported that *A. indica* and *Allium cepa* was the most effective to control *Xanthomonas campestris* pv. *Vesicatoria*.

Clove leaf extracts, onion extracts and Neemox effectively reduce the growth of bacteria after third spraying and mean reduction percentages of disease after fifth spray were 77.9%, 76.8% and 74.3% respectively. Antibacterial activity of clove to control *X. vesicatoria* in tomato has already been described by Lucas *et al.*, 2012 and "eugenol" which is contained in clove has an important function in the membrane of microorganism, including the inhibition of specific processes or cellular enzymes (Walsh *et al.*, 2003). These results coincided with experiment conducted by Opara and Wokocho,

2008 which indicated that onion showed antibacterial activity against *Xanthomonas campestris* pv. *Vesicatoria*.

Antibacterial activity of turmeric developed with the third round of spraying and showed 75.8% suppression of disease after fifth spray. The efficacy of spraying turmeric powder impregnated in sodium bicarbonate (0.15%) in the control of anthurium blight has been demonstrated in a previous study (Dhanya and Mary, 2006). Chemical compound “curcumin” (diferuloylmethane) found in turmeric has antimicrobial properties (Leela, 2002).

Mancozeb which is a recommendation to control bacterial blight at present, proved to be 72.7% effective and all the other plant extracts and essential oils were more effective than standard treatment while aloe extract was less effective (50.4%) against *X. campestris* pv. *dieffenbachiae*.

This study showed that plant extracts and essential oils contain antibacterial agents which inhibit growth of pathogen. González-Lamothe et al., 2009 reported that plants accumulate antimicrobial secondary metabolites to protect themselves such as phytoalexins and phytoanticipins defending plants against bacterial aggressors. This study confirmed the utilization of plant products in phytoprotection in an environmentally friendly manner with low cost.

Further research should be done to study the effect of plant extracts on other anthurium cultivars. Future thrust and follow-up research is needed to develop these plant extracts and essential oils as formulations on a large scale addressing economics of applying them and to find out effective concentrations of active ingredient according to their mode of action.

CONCLUSION

Among the six plant extracts and two essential oils studied, most effective treatments were lemongrass extract and citronella oil. Plant extracts of neem, clove, onion, turmeric and commercial product; neemox also proved to be effective against bacterial blight of anthurium compared to the control.

REFERENCES

- Alvarez A.M., Toves P.J., and Vowell T. S. (2006) Bacterial Blight of Anthuriums: Hawaii's Experience with a Global Disease. Available at: <file:///P:/print/BacterialBlightAnthuriums.pdf>. [Accessed on Oct. 2014]
- Dhanya M.K. and Mary C.A. (2006) Management of bacterial blight of anthurium (*Anthurium andreanum* Linden.) using ecofriendly materials. *Journal of Tropical Agriculture*. 44 (1-2): 74-75.
- El-Hendawy H. H., Osman M. E. and Sorou N. M. (2005) Biological control of bacterial spot of tomato caused by *Xanthomonas campestris* pv. *Vesicatoria* By *Rahnella aquatilis*. *Microbiological Research*. Microbial Research.160 (4): 343—352
- Fukui R., Fukui H., and Alvarez A.M. (1999) Comparisons of Single Versus Multiple Bacterial Species on Biological Control of Anthurium Blight. *Phytopathology* . 89(5):366-73

- González-Lamothe, Mitchell G., Gattuso M., Diarra M., Malouin F. and Bouarab K (2009) Plant Antimicrobial Agents and Their Effects on Plant and Human Pathogens. *International Journal of Molecular Sciences*. 10(8): 3400–3419.
- Leela N., (2002) Chemical composition of essential oils of turmeric, (*Curcuma longa L.*), *Acta Pharmaceutica*. Available at: <http://220.227.138.214:8080/dspace/bitstream/123456789/255/1/Chemical+Composition.PDF>. [Accessed on July, 2014]
- Lucas G.C., Alves E., Pereira R. B., Perina F. J. and Souza R. M. D. (2011). Antibacterial activity of essential oils on *Xanthomonas vesicatoria* and control of bacterial spot in tomato. *Phytopathology*. Available at: http://www.scielo.br/scielo.php?pid=S0100-204X2012000300006&script=sci_arttext&tlng=es [Accessed on July, 2014]
- Nishijima, W.T. and Fujiyama, D.K. (1985). Bacterial blight of anthurium. Commodity-Fact-Sheet, Cooperative Extension Service. Available at: <http://www.ctahr.hawaii.edu/oc/freepubs/pdf/CFS-AN-4A.pdf> [Accessed on July, 2014]
- Norman D. J. and Ali G. S., (2008) *Anthurium* Diseases: Identification and Control in Commercial Greenhouse Operations. Available at: <https://edis.ifas.ufl.edu/pp292> [Accessed on July, 2014]
- Opara E. U. Wokocha R.C. (2008). Efficacy of some plant extracts on the in vitro and in vivo control of *Xanthomonas campestris pv. Vesicatoria*. *Agricultural journal*. 3: 163-170
- Sathyanarayana N., Reddy O. R., and Latha S. (1998). Interception of *Xanthomonas campestris pv. dieffenbachiae* on Anthurium Plants from the Netherlands. *Plant disease*. 82 (2): 262
- Satish S., Raveesha K. A., and Janardhana G. R. (1999) Antibacterial activity of plant extracts on phytopathogenic *Xanthomonas campestris* pathovars. *Applied Microbiology*. 28, 145–147
- Walsh S.E., Maillard J.Y., Russel A.D., Catrenich C.E., Charbonneau D.L., Bartolo R.G. (2003). Activity and mechanisms of action of selected biocidal agents on gram-positive and -negative bacteria. *Journal of applied microbiology*. 94. 240-247

Validation of integrated dwarfing (*gai*) gene in *Clerodendrum philippinum* 'schauer' using GUS assay and PCR method

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ABSTRACT

This study was conducted to validate the *gai* dwarfing gene integration in *Clerodendrum philippinum* which has been transferred in the previous studies with the intension of producing a dwarf plant marketable as a potted plant. The plasmid used to carry the gene was pJIT60 with CaMV 35S promoter and (GUS) reporter gene. In this study, β glucuronidase (GUS) assay and polymerase Chain Reaction (PCR) methods were used to detect whether the *gai* gene has been integrated into the plant genome or not. The study was carried out in Department of Crop Science, Faculty of Agriculture, University of Peradeniya. In the GUS assay, there were some plants in which, blue colour appeared in their veins. However, the results were consistent. Analysis from PCR showed that there were 6 plants which were positive to 35S promoter gene in this study, which shows that the *gai* gene has been integrated into six plants tested. As the results from GUS assay were not reliable, PCR method can be used to validate integration of the *gai* gene.

Keywords: *Clerodendrum philippinum*, *gai* gene, GUS assay, PCR, 35S promoter

INTRODUCTION

Clerodendrum philippinum belongs to family Lamiaceae which is a perennial shrub, up to 3 m height with attractive, scented and dense terminal heads. Flowers contain purple or red calyx and pale pink corolla. The plant is economically important as the plant has medicinal and pharmaceutical properties as well a special ornamental value due to its fragrance and the beauty of the flowers. However, the tall and spread architecture and high leaf-to-flower ratio hinder its use as an ornamental plant. An attempt is progressing to alter plant architecture towards producing a dwarf plant which makes the plant more desirable for floriculture industry.

With the purpose of altering plant architecture of *C. philippinum* plant, pJIT60 plasmid which has been constructed expressing *gai* under the control of the CaMV 35S promoter and with ampicillin resistance gene and GUS reporter gene as the selector and the marker has been transformed using particle bombardment gene transfer method in previous studies undertaken by Department of Crop Science, University of Peradeniya.

The *gai* is a semi-dominant mutation of *Arabidopsis*, which confers dark green, dwarf phenotype (Peng and Harberd 1993) because the *Arabidopsis gai* mutant allele confers reduction in gibberellin (GA) responsiveness. Gibberellins (GAs) are tetracyclic diterpenoid growth factors that are essential regulators of stem elongation and other plant developmental processes (Hooley 1994). It confers reduced GA responses and so increase endogenous GA levels (Talon *et al.* 1990). Comparison of *GAI*

and *gai* DNA sequences shows that the mutant protein (*gai*) lacks a short (17-amino-acid) segment of the GAI protein sequence. This structural alteration is responsible for the dominant, gain-of-function properties of *gai* (Jinrong *et al*, 1997).

Successive gene incorporation will express the characters of particular gene in genetically modified plant. To optimize the transformation process several tests are carrying out such as PCR, GUS assay and Ampicillin screening. GUS assay is used to analyze the expression of a marker gene either in a quantitative way or through visualization of its activity in tissues. As plant cells themselves do not contain any GUS activity, the production of a blue staining testing with X-gluc in particular cells indicates the transcription of *gus* A-chimeric gene in that particular cell. PCR is a very powerful technique, now used in many areas of biology, which allows *in-vitro* amplification of specific DNA sequences from undetectable quantities of target DNA (Innis *et al*, 1990).

MATERIAL AND METHODS

GUS assay was followed according to a method described by Patel (1990). Leaf samples were collected from nine *in-vitro* *C. philippinum* plantlets which were subjected to gene transfer. Wild *in-vitro* *C. philippinum* plant leaves were used as negative control. Two replicates from each were used for GUS activity. As the blue staining is difficult to visualize against dark green background of mature leaves, samples were taken out from GUS assay solution and placed in 70% ethanol until the chlorophyll is extracted into ethanol.

DNA Extraction and Purification

Plant samples used in GUS assay were selected for DNA extraction. Two replicates from each treatment were used. CTAB method was followed for DNA extraction according to Doyle and Doyle method (1987). To obtain the correct purity for PCR extracted DNA was allowed to ethanol purification. For that, 0.1 volume of 3M Sodium acetate (pH 5.2) and 2 volumes of absolute ethanol were added and incubated at -20 °C for overnight. Then samples were spinned at 5000 rpm for 15 minutes, drained and rinsed the pellet with 70% ethanol. Finally, pellets were air dried and dissolved in 30 µL of distilled water. To check the purity of samples, nano drop readings including DNA concentrations (ng/µL), wave length ratios of 260/280 and 260/230 were obtained.

Deciding Primer Oligonucleotides

To detect transgenic *C. philippinum* plants during PCR, a primer pair was used which have been used in a previous study by Vollenhofs *et al* (1999). This primer pair was designed that amplified a fragment covering the junction between the CaMV 35S promoter and the chloroplast transit peptides (CTP) sequence derived from *Petunia hybrida* 5-enol-pyruvylshikimate-3-phosphate-synthase (EPSPS) gene. As CaMV 35S gene promoter is naturally not present in any plant except Cauliflower, 35S promoter in transgenic *C. philippinum* plant with pJIT60 plasmid can be identified using primers of CaMV 35S promoter.

Gene sequence of 35S promoter was identified using the following link <http://www.pgreen.ac.uk/JIT/pJIT60.htm>, and checked the forward and reverse primer sequences with 35S promoter gene sequence. Primer pair was provided by Integrated DNA Technologies (<http://www.idtdna.com>) and provided by Avon Pharmo Chem (pvt) Ltd, Nugegoda. Sequences of primers are listed in the Table 1.

Table 1: Oligo nucleotide primers

Primer	Sequence
35SFZ1 (forward)	5'-CCG ACA GTG GTC CCA AAG ATG GAC-3'
35SFZ2 (reverse)	5'-ATA TAG AGG AAG GGT CTT GCG AAG G-3'

Polymerase Chain Reaction

Amplification reactions were carried out in a 20 μ L total volume. Final concentrations of PCR components were as follows: PCR buffer minus Mg^{++} ; 1 \times , dNTPs; 0.2 mM each, $MgCl_2$; 1.5 mM, Primers; 0.5 mM each, *Taq* DNA Polymerase 1 Units/ reaction. PCRs were performed using 2 μ L of extracted DNA. Cyclic condition for PCR was as follows; Initial denaturation at 94 $^{\circ}C$ for 3 minutes, Denaturation at 94 $^{\circ}C$ for 45 seconds, Annealing 55 $^{\circ}C$ for 45 seconds, Extension at 72 $^{\circ}C$ for 90 seconds, Final extension at 72 $^{\circ}C$ for 10 minutes. PCR amplification was performed for 35 cycles. One-percent agarose concentration was used for the electrophoresis of the PCR amplified products.

RESULTS AND DISCUSSION

The expected strength of blue color in the GUS histochemical assay did not appear in the tested plants of *C. philippinum*. But light blue color was observed in the veins of some samples.

According to the results of GUS assay, the correct blue reaction product did not appear in the tested plant tissues. According to this integration of *gai* gene, it may not be successful in some plants and some plant samples may have given false negative results. This *gusA* reporter gene encodes the enzyme β -glucuronidase (GUS) which cleave the chromogenic (colour generating) substrate X-Gluc (5-bromo-4-chloro-3-indolyl β -D-glucuronic acid), resulting in the production of an insoluble blue colour in those plant cells displaying GUS activity. β -glucuronidase activity is not detected in most plant species, therefore the detection of GUS activity in plant cells is excellent evidence that the *gusA* gene has been introduced and is being expressed. The lack of GUS expression in tested leaves and may be due to alteration or loss of GUS gene resulting from rearrangement of the coding sequence or methylation of the gene, as suggested by Batraw and Hall (1993).

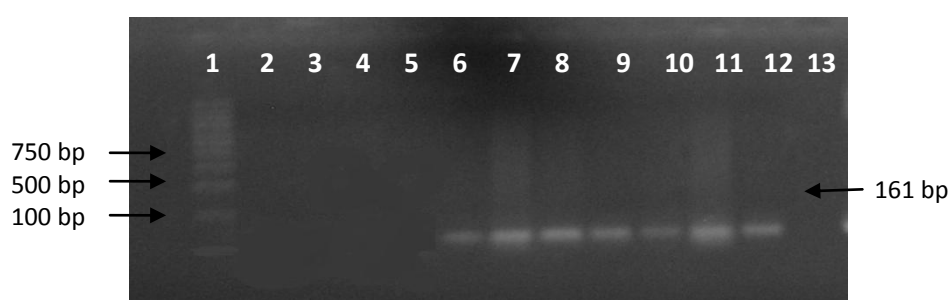


Plate 2: Agarose Gel Electrophoresis of PCR products of *C. philippinum* with 1kb DNA ladder (Lane 1; 1kb DNA ladder, Lane 3,4,5,6,7,8,9,10 and 11 are separate samples taken from gene integrated in-vitro plantlets of *C. philippinum*, Lane 12; *gai* gene containing pJIT plasmid as positive control, Lane 13; Wild *C. philippinum* plant as negative control.

The presence of ferri- and ferrocyanide in the incubation medium is a critical point for the visualization reaction in this procedure. It accelerates the formation of the final reaction product. It also protects the formed indigo from further oxidation, which would convert it to colourless or yellowish products. Optimal concentration of the ferri- and ferrocyanide should be tested for a given

specimen to find the best compromise between the intensity of staining and the precision of localization (Vitha, 2012). As the GUS protocol used in this study did not contain any ferri- and ferrocyanide in the incubation medium, it may be the reason to obtain negative results.

In PCR method, the primer pair specified for the CaMV 35S promoter yielded a PCR product of 161 bp size in the pJIT60 plasmid (positive control) containing lane. It indicates that the 161 bp size has replicated under the cyclic conditions used in PCR. It also showed the positive PCR results for six of the plants tested. According to the theory in genetics a promoter is a region of DNA that initiates transcription of a particular gene and promoters are located near the transcription start sites of gene. Thus, it can be concluded that if those plants are positive for the promoter, the *gai* gene has also been integrated into the plant genome.

It is very important to obtain pure DNA for PCR. Because endonucleases, polyphenols, or polysaccharides, which can all co-precipitate along with DNA during extraction, result in irreversible interactions with nucleic acids and then it can affect enzymatic reactions such as *Taq* DNA polymerase-mediated PCR (Wang *et al.*, 2012).

CONCLUSION

According to the study, PCR results showed that, the dwarf gene has successfully incorporated to six of the tested plants. As results from GUS assay were not reliable, PCR method can be used to validate integration of the *gai* gene.

REFERENCES

- Doyle J.J., and Doyle J.L. (1987) A rapid DNA isolation procedure for small quantities of fresh leaf tissue. *Phytochemical Bulletin* 19:11-15
- Peng, J., & Harberd, N. P. (1993). Derivative Alleles of the Arabidopsis Gibberellin Insensitive (*gai*) Mutation Confer a Wild-Type Phenotype. *The Plant Cell*.5:351-360
- Hooley, R. (1994) Gibberellins: Perception, transduction and responses. *Plant Molecular Biology* 26: 1529–1555.
- Talon, M., Koornneef, M. and Zeevaart, J.A.D. (1990) Accumulation of C19-gibberellins in the gibberellin-insensitive dwarf mutant *gai* of *Arabidopsis thaliana* (L.) Hehyn. *Planta*. 182: 501–505.
- Jinrong , P., Pierre, C., Donald, E.R., Kathryn E.K., Rachel J.C., George P.M., and Nicholas P.H. (1997) The *Arabidopsis GAI* gene defines a signaling pathway that negatively regulates gibberellin responses. *Genes Development*. 11(23): 3194–3205.
- Innis, M.A., Gelfand, D.H., Sninsky, J.J., and White, T.J. (Eds). (1990) PCR protocols: A Guide to Methods and Applications. Academic Press, San Diego. CA.

- Patel, N. J. (1990) GUS reporter system as an alternative to detect Cry2Ab gene in BT cotton. In *Research Guide* 130–147.
- Vollenhofer, S., & Burg, K. (1999). Genetically modified organisms in food screening and specific detection by polymerase chain reaction. *J. of Agric. Food. Chem.* 47: 5038-5043
- Battraw, M.J. and Hall, T.C. (1990) Histochemical analysis of CaMV 35S promoter-beta-glucuronidase gene expression in transgenic rice plants. *Plant Molecular Biology*. 15(4): 527–538.
- Wang, X. Xiao. H., Zhao, X., Li, C., Ren, J., Wang, F., Pang, L. and Croatia, C.M. (2012). Isolation of high-quality DNA from a desert plant *Reaumuria songorica*. *Genetic Diversity in Plants*. 1-12.
- Vitha, S., Benes, K., Gartland, K.M.A. and Elliott, M.C. (1993) An improved indigogenic procedure for in situ localization of β -glucuronidase in roots of transgenic plants. 4th International Symposium on Structure and Function of Roots, Stará Lesná, Slovakia.

Social status of the flower vendors in two selected religious places in Kataragama and Rathnapura

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ABSTRACT

Sri Lankans use flowers mainly for religious purposes and flower markets are available closer to temples with fresh flowers. These huge demanded niche markets are un-noticed and less-studied. This study was conducted to review present status, major constraints faced by the vendors and potentials for younger generations' involvement related to the flower-vending sector in and around "Kiri-vehera temple in Katharagama" and "Maha Saman Dewalaya in Rathnapura" during May - June 2015. In addition, attempts were made to explore the relationship between socio-economic factors and constraints in sector faced by the selected flower vendors. The population of the research was the vendors (both registered and non-registered) at two selected sites. The targeted sample size was the whole group of established "registered formal" flower vendors from the selected areas, which contained 58 respondents in which, 30 respondents were from Katharagama Kiri-Vehera Temple and 28 respondents were from Rathnapura Maha Saman Dewalaya. Data were collected using structured questionnaires, checklists, key informants surveys and interviewing. Data were analyzed through descriptive statistics, correlation analysis and multiple linear regression using SPSS 16 and Minitab 17 statistical packages. Study revealed that majority of the vendors is females (84.5%). Vendors have a mean monthly income level of Rs. 24,966 (~ Rs. 25,000). Most of the respondents (87.9%) are satisfied as a flower vendor. A considerable proportion (46.3%) of younger generation is presently involved in the sector. The number of constraints vendors face is considerably high. Out of the two selected areas, vendors attached to Rathnapura Maha Saman Dewalaya face relatively greater constraints levels. When consider the overall, lack of support from government and other related authorities, laws and regulations, policies were identified as most affecting constraints for the vendors.

Keywords: *Flower Vendors, Religious Areas, flower market, Younger Generations, female participation*

INTRODUCTION

Production and selling of local fresh flowers has become a key business at many religious places in Sri Lanka for many years. Mostly the women are engaged in this activity as a self-employed venture. These niche markets for local fresh flowers have gone unnoticed and less studied (Niranjana and Gunasena, 2006). Evaluation of present status and problems associated with this small business is very much important. There is a massive demand for fresh flowers during festival season in many religious places (Sriwarnasingha *et al.*, 2013), however, due to numerous constraints faced by the

vendors, they are unable to earn profits from their business. Therefore, further studies on current status and constraints in the sector will be helpful for socio-economic development and rural development as well as to upgrade these less-studied niche markets.

Therefore, the present study was conducted to explore the relationship between socio-economic status and constraints faced by the flower vendors who carry out their business activities in the areas of “Kiri-vehera temple in Katharagama” and “Maha Saman Dewalaya in Ratnapura”. More specifically, the study was aimed to find out the present social and economic status of the flower vendors in Kiri vehera temple and Maha Saman Devalaya premises, to identify the potentials for involvement of younger generation in this particular business with special focus on barriers for them to sustainably involve in it and, to find out the constraints faced by the flower vendors in two selected areas.

MATERIALS AND METHODS

A survey was conducted using a structured questionnaire among flower vendors who were attached to two selected religious places – Kiri-vehera temple in Katharagama (KVTK) and Maha Saman Dewalaya in Ratnapura (MSDR) in Sri Lanka. Both “formal/registered vendors” and “informal/non-registered vendors” were selected as the sample. The targeted sample size was the whole group of established registered flower vendors containing 58 respondents. Thirty respondents were from KVTK and twenty-eight respondents were from MSDR. Primary data collection was carried out using questionnaire based personal interviews and checklists, over May to June months in 2015 which was an off season of the flower markets in the selected areas. Respondents were directly asked about identified twenty major constraints they face during vending activities, using a five-point Likert scale from very unimportant to very important. In addition, each respondent was asked to rank five most affecting factors out ten most affecting for younger generations’ potential involvement in the business.

Constraint levels were measured by constructing “constraint indexes” (Jayasinghe-Mudalige and Henson, 2006). Data were analyzed using descriptive statistics, correlation analysis and multiple linear regression using SPSS 16 and Minitab 17 statistical packages. Multiple linear regression (1), Constraint index (2) and potential for younger generations’ involvement (3) were measured by using following indices.

$$Y_i = \beta_0 + \beta_1 X_1 + \beta_2 X_2 \dots + \beta_n X_n + \epsilon_i \quad (1)$$

$$\text{Constraint Index} = \frac{\text{Score obtained by a respondent}}{\text{Possible Maximum Score}} \quad (2)$$

$$\text{Younger generations' constraint index} = \frac{\text{Score obtained by a constraint statement}}{\text{Possible Maximum Score}} \times 100 \quad (3)$$

To explore the relationship between socio-economic status and constraints faced by the flower vendors, multiple linear regression analysis was conducted between constraint index value (dependent variable) obtained and eleven identified socio-economic characteristics (independent variables) which were selected in reference to the previous literature and pilot test. The selected

independent variables were; age of the respondent, marital status, education level, household size, gender of the respondent, experience in industry, working hours per day, distance from residence to working place, stall availability, other income (not having any other income source) and vending area.

RESULTS AND DISCUSSION

According to the study, most of the flower vendors are females (84.5%). Majority of the total respondents are included in to “more than 50 years” group (34.5%). Majority of the flower vendors have a greater experience in the venture as they have involved in the business for more than 20 years. It represents by 43.1%. According to the study, most of the flower vendors involve in the venture within 12-16 hours category. It represents by 55.2% of the total sample. Majority of vendors cited that they have a stall for vending activities (62.1%). Majority of the flower vendors stated that flower vending is the only mode of income of the family. It represents 70.7% of the total respondents. When consider about the marital status, out of the total flower vendors, majority are married and it was 79.3%. According to the study, majority (51.7%) of the total flower vendors have obtained education up to “Grade 7 – Ordinary Level (O/L)”. The second highest goes for both “Grade 1-6” and “Passed Ordinary Level” categories with 17.2% equally. Next highest category is “Not Attended to School” with 10.3% and the “Passed Advanced Level” category was 3.5%.

As mentioned by 70.7% of total flower vendors, the distance from residence to working place is within 1 km. Remaining 29.3% of total flower vendors stated that the distance from residence to working place is more than 1Km. As mentioned by 79.3%, they engage in self-supplement of flowers. In addition, 96.6% of the total flower vendors mentioned that they do purchases from outsiders. Moreover, as mentioned by 44.8% of the vendors, they sell flowers to flower shops within the area and take special customer orders such as for wedding ceremonies, hotels, other road travelers etc. According to the survey, majority (79.3%) of the flower vendors face wastage of flowers.

When consider the sources of getting initial capital, 98.3% of the respondents mentioned that they have used their own capital, 55.2% mentioned that initial capital was given by the relatives and 53.4% have obtained the initial capital from private banks. In addition, 53.4% have obtained initial capital from government banks. 41.4% mentioned that they have received initial capital from friends. As mentioned by the respondents, 24.1% of the respondents have not used banks as the source of initial capital.

As mentioned by 8.6% of the total flower vendors, they hire external assistants for the vending activities. Furthermore, most of the respondents (79.3%) have stated that the family members are involved in the venture to give the support. According to the study, it can be identified that majority of the vendors are getting the support from females.

According to the survey, majority of the flower vendors save money monthly. As a percentage, it represents 87.9% of the total sample. Out of the flower vendors who save money, 13.7% of the respondents have not used banks for saving money as they keep money with themselves. As they mentioned, lack of knowledge for banking activities was the main reason to be identified.

According to the study it can be identified out of the respondents who have children, 50% of the flower vendors have mentioned that their next generation prefers to involve in venture as well 50% have mentioned that their next generation does not prefer to involve in venture. However, out of the flower vendors who have children, 46.3% mentioned that their next generation is already involved in the venture and 53.7% mentioned that their next generation is not involved in the venture. According to the survey it can be identified that out of the flower vendors who have children, majority have stated that their children do not involve in the venture at present. In addition, it can be further identified that, although 50% of the next generation prefers to involve in this venture, only 46.3% is currently involving. Moreover, even though 50% of the next generation does not prefer to involve in this venture, 53.7% is not currently involving in the venture. But according to the survey, majority out of the flower vendors who have children (68.5%) have mentioned that they do not prefer if their next generation involve in the venture.

According to the survey study, majority of the total responded flower vendors have mentioned that they are satisfied with the venture they involve. As a percentage, it represents 87.9% of the total sample. Only 12.1% of the total respondents stated that they are not satisfied with the venture they involve. According to the study, out of the total flower vendors, majority have not already planned to expand the industry. As a percentage, it represents 84.5% of the total sample. Only 15.5% of the total respondents have already decided to expand the industry.

Flower vendors in Katharagama mentioned that peak demanding months are July-August (Perahera season), and school vacation months (January, April, August, December). Flower vendors attached to Ratnapura Maha Saman Dewalaya stated that peak demanding months are August - September (Perahera season), Shripada season (February – May) and school vacation months (January, April, August, December). Responded Flower vendors further mentioned that *poya* days and weekends are the peak demanding days.

The results of the regression analysis revealed that level of constrains (constraint index values) faced by the flower vendors who do their business activities in the areas of KVTK and MSDR are significantly affected by the six socio-economic factors based on obtained P-value. They are; household size, gender of the respondent, working hours per day, stall availability, availability of other income source and vending area. In addition, the R-squared value for the full model's output was 89.1%. The results of the regression analysis revealed that constrains faced by the flower vendors who do their business activities in the areas of KVTK and MSDR are not significantly affected by the socio-economic factors; Age of the respondent, Marital Status, Educational level, Age of the business / Experience in industry and Distance from residence to working place.

According to the Figures 1-3, it could be identified that, majority of the flower vendors attached to MSDR face higher amount of constraints when compared to the vendors at KVTK premises.

Major constraints identified under the category of extremely severe:

In MSDR, non-availability of proper selling locations/areas, competition and laws and regulations, policies (registration, payments etc.) were identified as extremely severe barriers. In KVTK, uncertainty of market demand & its' fluctuation, lack of support by government and other related

authorities and consequent price fluctuations and price instability were identified as extremely severe barriers.

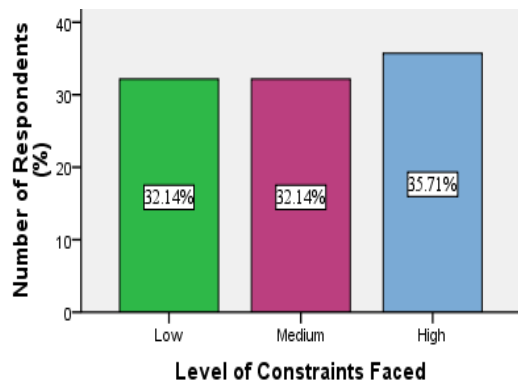


Figure 1: categorized constraint index values (MSDR)

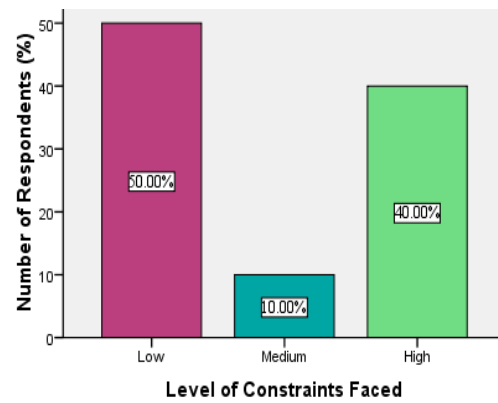


Figure 2: categorized constraint index values (KVTk)

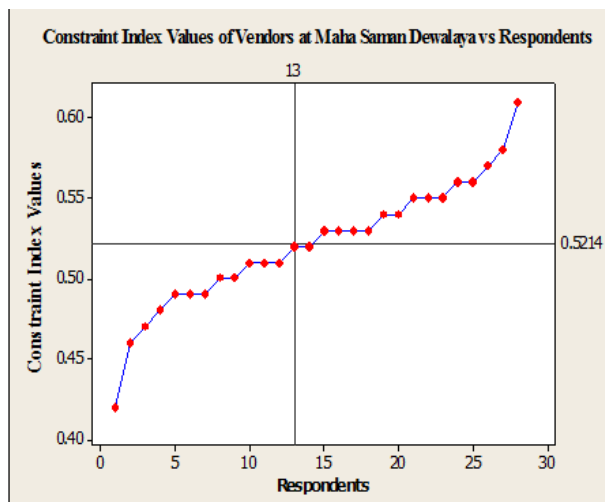


Figure 3: constraint index values vs. Flower vendors at MSDR

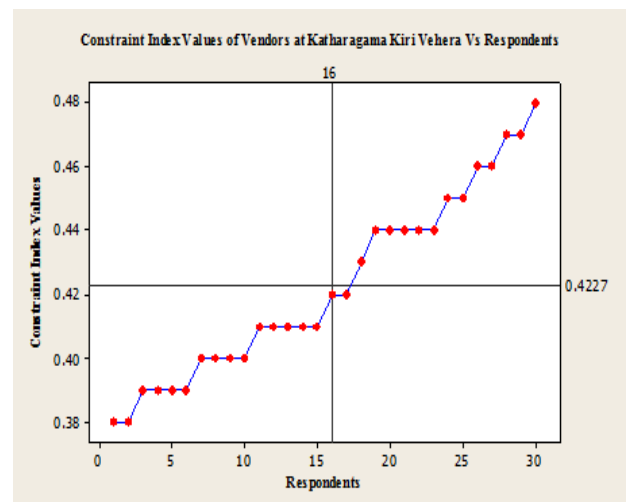


Figure 4: constraint index values vs. Flower vendors at KVTk

Major constraints identified under the category of severe:

In MSDR, lack/fluctuation of market demand, lack of adequate market facilities, lack of awareness on programs providing capital for expansion, lack of financial and banking resources/capital, poor storage facilities, lack of support by government and other related authorities and multiple responsibilities were identified as severe barriers. In KVTk, high cost - purchasing flowers, relatively low prices, seasonal variation of flower supply and shortage of flowers, competition, lack of information on market trends/ requirements, multiple responsibilities and laws and regulations, policies were identified as severe constraints.

Major factors that hinder the potential for involvement of younger generation:

In MSDR, less social acceptance and recognition was identified as extremely severe barriers for potential younger generations' involvement in the venture. In KVTk, lack of ability in obtaining financial and banking resources/ capital, lack of support by government and other related

authorities were identified as extremely severe barriers for potential younger generations' involvement in the sector. In addition, higher competition, non-availability of selling location and laws, regulations and policies were identified as extremely severe barriers that were common at both areas for potential for involvement of younger generation in the venture.

CONCLUSION

Study reveals that majority of the vendors are females (84.5%) and the vendors have an approximate mean monthly income of Rs. 25,000. Most of the respondents (87.9%) are satisfied as a flower vendor and there is a moderate level of present involvement of younger generation in the sector (46.3%), however, the level of constraints they face is considerably high. When consider the overall studied area, lack of market demand and its' fluctuation, non-availability of proper selling location/area, multiple responsibilities of vendors, lack of support by government and other related authorities, laws and regulations, policies were identified as the most affecting constraints. High level of competition, less social acceptance and less recognition, lack of ability in obtaining financial and banking resources/ capital, non-availability of proper selling location/ area, laws, regulations and policies and lack of support by government and other related authorities were identified as the factors that hinder the potential of involvement of the younger generation in the business.

According to the findings of this study, constraints faced by the flower vendors within the studied area are significantly affected by the socio-economic factors such as household size, gender of the respondent, working hours per day, stall availability, availability of other income source and vending area.

There is a greater possibility in expanding these less studied niche markets as there is a high demand and the majority of the vendors are satisfied with their involvement in the sector. In addition, there is a huge potential on attracting unemployed females further for this sector which will be very important for the economic development of the country. Introduction of new rules, regulations and policies can be identified vital for the growth of this religious-based floricultural sector.

REFERENCES

- Jayasinghe-Mudalige, U. K. and Henson, S., (2006). Economic incentives for firms to implement enhanced food safety controls: case of the Canadian red meat and poultry-processing sector. *Review of Agricultural Economics*, 28(4), 494-514.
- Niranjan, S. K. D. F and Gunasena, H. P.M., (2006). Floriculture Sector Development Programme: Small and Medium Scale Entrepreneurs in Sri Lanka. Sri Lanka Council for Agricultural Research Policy.
- Sriwarnasingha, A.N., Beneragama, C.K. and Nalaka, G.D.A., (2013). 1-Methylcyclopropene (1mcp) on the Vase-Life and Floral Opening of Cut Nil Manel (*Nymphaea nouchali*) Flowers. Degree: Faculty of Agriculture Rajarata University of Sri Lanka.

Effects of BAP (Benzylaminopurine) and supplementary calcium nitrate on young leaf abscission of rooted stem cuttings of *Codiaeum variegatum* varieties 'mammy' and 'iceton'

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ABSTRACT

Codiaeum variegatum L. varieties "Mammy" and "Iceton" are popular ornamental foliage plants exported from Sri Lanka. These varieties are exported as rooted stem-top cuttings. However, two to three young leaf buds and/or leaves are abscised prematurely from cuttings during rooting, which reduce the quality and marketability of the cuttings. Therefore two different chemicals, namely 3 mM of calcium nitrate, 0.2 mM of benzylaminopurine (BAP) were tested for reducing leaf abscission, of *C. variegatum* varieties, "Mammy" and "Iceton", keeping chemical-free control (water) in a complete randomized design (CRD) with three replicates during March-July, 2015 in a commercial production unit at Rambukkana (WM₃) Sri Lanka. The highest number of leaves and the largest leaf area was abscised in cuttings in the control ($p < 0.05$). Calcium nitrate and BAP treated cuttings had the lowest leaf abscission and the smallest leaf area of shed leaves. Young leaves of cuttings treated with $\text{Ca}(\text{NO}_3)_2$ were highly concentrated in calcium compared to other treatments. Number of leaves initiated was higher under calcium nitrate treated plants compared to the control. The number of roots and their fresh weights were higher in cuttings in the control compared to other treatments. However, shoots length, shoot fresh and dry weights, and dry weight of roots were similar in all treatments. In conclusion, foliar application of 3mM calcium nitrate improved the bud retention decreasing the leaf shed of cuttings of *C. variegatum* varieties "Mammy" and "Iceton".

Keywords: Calcium nitrate, *Codiaeum variegatum*, benzylaminopurine, leaf abscission

INTRODUCTION

Sri Lankan floriculture industry started in early 1970s and presently floricultural crops hold about 500 ha of land in Sri Lanka and out of this, 472 ha are under ornamental foliage species, indicating the economic importance of foliage species in Sri Lanka. *Codiaeum variegatum* L. (Croton) varieties "Mammy" and "Iceton" (family: Euphorbiaceae) are popular ornamental foliage plants exported by Sri Lanka. The popularity of these varieties is attributed to their attractive foliage colour and shape, and adaptation both to indoor and outdoor conditions.

Mike Flora (Pvt.) Ltd. exports large quantities of rooted stem cuttings of *Codiaeum variegatum* L. var. Mammy and Iceton. The rooted stem cuttings of Mammy and Iceton are produced by rooting a piece of a stem-top cutting, which are exported. However, two to three young leaf buds and/or leaves are prematurely shed from cuttings before they are exported, which reduce the end quality of the rooted cuttings, which are rejected from the export market. The aim of the study was to overcome

the shedding of buds and young leaves of stem cuttings of *C. variegatum* Mammy and Iceton during rooting.

MATERIALS AND METHODS

The experiment was conducted during March to July, 2015, at Mike Flora (Pvt.) Ltd., at Rumbukkana (WM₃), Sri Lanka. The mean ambient temperature and relative humidity during daytime were 32.2 ± 0.6 °C and $73.6 \pm 0.5\%$, respectively during March to July, 2015.

Top shoot cuttings of *C. variegatum* varieties of 'Mammy' and 'Iceton' of 25 cm long, were used for the experiment. Proximal end of the cuttings were dipped in rooting hormone for few seconds to induced rooting. Then cuttings were planted in net pots, which were submerged in water in a shallow cup (hydrocup) and kept in propagators for 6 weeks during which treatments were applied. The complete randomized design (CRD) was implemented where three groups of cuttings from each variety were treated either with 3 mM of $\text{Ca}(\text{NO}_3)_2$ (aq) as a foliar application or 0.2 mM of BAP(aq) or water (control). Each treatment had four replicates.

Temperature (using thermometer) and relative humidity (using electronic RH meter) inside and outside of the propagator house were measured daily. The numbers of leaves initiated were counted. The shoot lengths were measured weekly. The numbers of leaves shed were counted and their leaf area was estimated using a grid chart. Rooted stems were harvested at the end of the experimental duration and the numbers of roots, fresh and dry weight of organs (shoots, roots) were determined. Dry biomass measured by oven drying the fresh plant samples at 85 °C for 24 hours. The calcium concentration of the young leaves was measured at the end of the experiment using flame photometer. Data were analyzed by ANOVA and Dunnet test ($P < 0.05$) by using statistical software, Minitab 16.

RESULTS AND DISCUSSION

For both varieties, the number of leaves shed per plant significantly lower in cuttings treated with calcium nitrate and followed by BAP compared to the control (Fig. 1A). Calcium maintains the cellular integrity by synthesizing more Ca-pectate in leaf tissues (Poovaiah and Leopold, 1973; Chou and Kao, 1992). In plants, exogenous application of cytokinins such as BAP, may inhibit the degradation of chlorophyll and photosynthetic enzymes, and respiratory kinase, thus increasing leaf longevity and retention (Richmond and Lang, 1957). Further, deficit of endogenous cytokinins lead to leaf senescence and abscission (Ferrante *et al.*, 2009).

Stems in the control, shed the highest leaf area while it was the lowest for calcium nitrate and BAP treated stems, indicating lesser damage of leaf area with commodity treatments (Fig. 1B). The highest number of leaves was initiated in calcium nitrate treatment in both varieties (Fig. 2) because calcium is responsible for new leaf formation and terminal bud development (Simon, 1977). The effect of BAP on number of leaves initiated was not significantly different from the control. However, interactions between variety and treatments occurred in weeks, four to six indicating positive effects of BAP on var. Mammy.

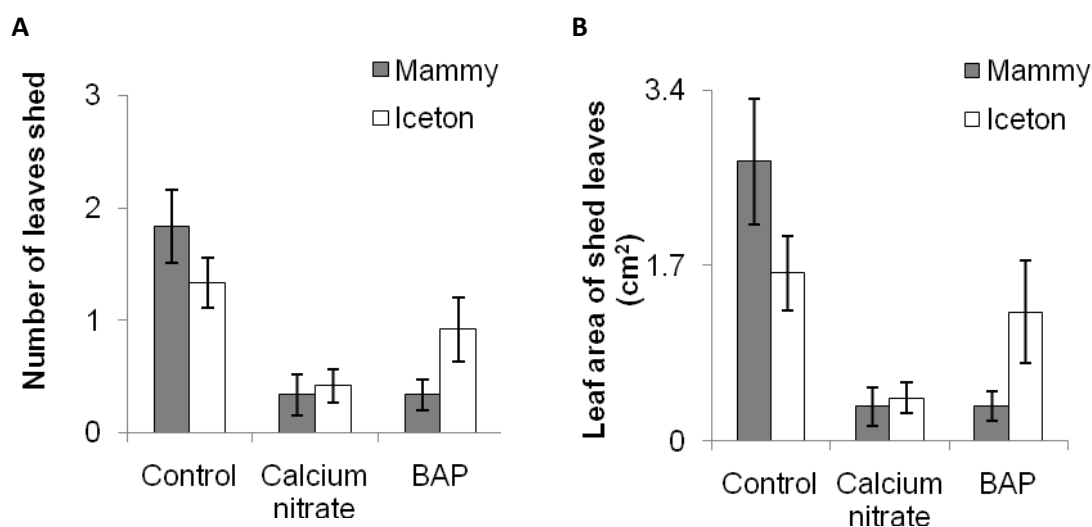


Figure 1: Effect of calcium nitrate and BAP (benzylaminopurine) on (A) mean number of leaves shed and (B) leaf area of shed leaves of cuttings of *C. variegatum* varieties, 'Mammy' and 'Iceton'

Leaves in stems treated with calcium nitrate were highly concentrated in calcium compared to other treatments (Fig. 3A) because the uptake efficiency of Ca^{2+} was 10 to 12 times higher through the leaves than the roots (Tucker, 1999). The calcium concentration in leaves of control and BAP were significantly low. The abscised young leaves from control showed calcium deficiency symptoms which included inhibition of bud growth and death of shoot tips.

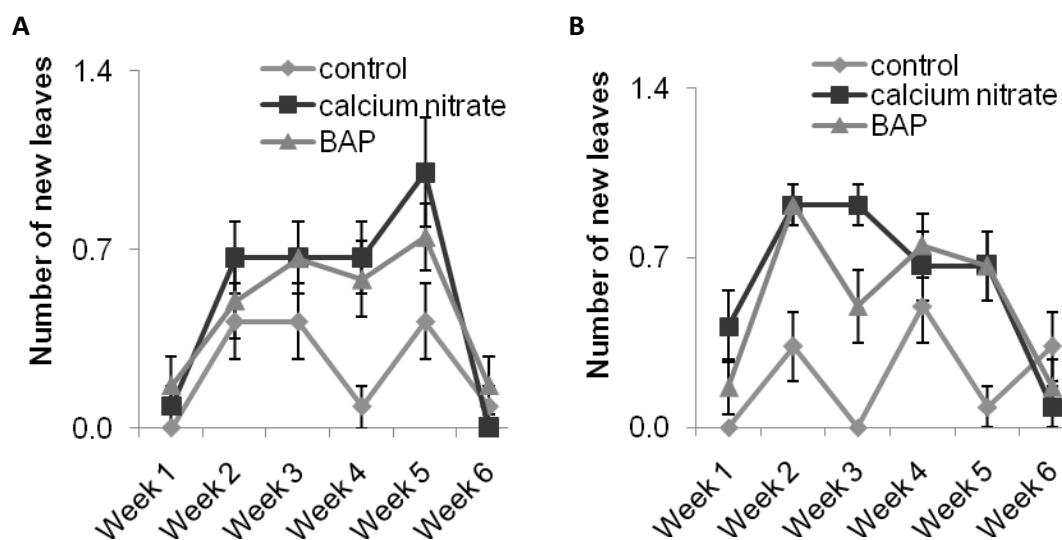


Figure 2: Effect of calcium nitrate and BAP (benzylaminopurine) on (A) mean number of new leaves initiated in cuttings of *Codiaeum variegatum* varieties (A) 'Iceton' and (B) 'Mammy'

In var. 'Iceton', the number of roots was not different between treatments. However, in var. 'Mammy' the control had the highest number of roots and fresh weight of roots compared to the treatments. Fresh weight of shoots, dry weights of roots, shoots and shoots lengths were similar in all treatments.

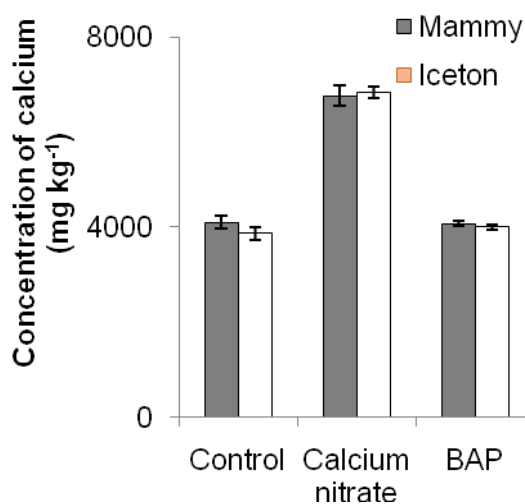


Figure 3: Effect of calcium nitrate and BAP (benzylaminopurine) on calcium concentration in young leaves cuttings of *Codiaeum variegatum* varieties 'Mammy' and 'Iceton'

CONCLUSION

Foliar application of 3 mM calcium nitrate effectively reduced immature leaf shed of rooted stem cuttings of *C. variegatum* varieties, 'Mammy' and 'Iceton'. Further, foliar application of 3 mM calcium nitrate improved the commercial characteristics of cuttings such as number of roots, number of leaves and quality of buds of *C. variegatum* varieties.

REFERENCES

- Chou, C.M. and Kao C.H. (1992). Methyl jasmonate, calcium, and leaf senescence in rice. *Plant Physiology*, 99: 1693–1694.
- Ferrante, A., Mensuali-Sodi A. and Serra, G. (2009). Effect of thidiazuron and gibberellic acid on leaf yellowing of cut stock flowers, *Central European Journal of Biology*, 4: 461-468.
- Pooaiah, B.W and Leopold A.C. (1973). Inhibition of abscission by calcium. *Plant Physiology*: 848–851.
- Richmond, A.E and Lang A. (1957). Effect of kinetin on protein content and survival of detached *Xanthium* leaves. *Science*.125:650–651.
- Simon, E.W (1977). The Symptoms of Calcium Deficiency in Plants, *New Phytologist*, 80: 1-15.
- Tucker, M.R. (1999). Essential Plant Nutrients: North Carolina soils and role in plant nutrition. Retrieved on May 13, 2015 from <http://www.readonlinebooks.net/essential-plant-nutrients-nc-state-university-pdf>. [Accessed on March, 2014]

Growth stage based fertilizer application of petunia to adjust electrical conductivity and pH of the growing medium

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ABSTRACT

In floriculture industry, growers apply numerous fertilizers to improve the yield and quality of foliage and flowers. This indiscriminate use of fertilizers is one of the main reasons behind high solute concentration (measured in terms of electrical conductivity) in the potting medium, resulting in retarded plant growth and low quality of flower yield. In this study, three new fertigation options were compared with the present fertigation program to maintain the electrical conductivity (EC) and pH of the growing medium at a favorable range and thus to attain the required growth rate and a high quality seed yield of greenhouse grown *Petunia hybrida*. Three new fertigation options (Trt 3 – Trt 5) were composed of standard dosage of Alberts® fertilizer (Trt 3), Alberts, Calcium nitrate and Super K® (Trt 4) and UTAH® with TSP as the basal dressing followed by new mix Ca(NO₃)₂, KCl, MgSO₄, (NH₄)₂SO₄ and ZnSO₄ (Trt 5). Treatments 1 and 2 were fertigated similar to commercial sector of pot *Petunia* cultivation where Trt 2 was without foliar amendments of fertilizer. Treatment 5 (new mix) was able to maintain the EC of the growing medium at a moderate range but early flowering was comparatively low. In spite of higher levels of EC maintained in the growing medium (1.5 µS cm⁻¹), vigorous plant growth could be observed in the control (Trt 1). As an evidence of plant nutrient uptake, shoot N contents were found to be high when higher N doses were provided through the growing medium while shoot P content responded very high to high phosphorous content in the growing medium under different fertigation options. In contrast, plant K and Ca contents did not show significant responses to high K and Ca fertigation (higher levels in the potting medium), respectively. Results also indicated that the seed quality, measured in terms of 1000 seed weight, germination percentage and seed vigor, was not affected by changing fertigation programs. Excessive vegetative growth associated with high phosphorous content in the medium gave a low flower yield. Plant growth rate and seed quality of *Petunia* did not respond to varying combinations and dosages of fertilizers practiced in the fertigation program significantly while plants could perform well within an EC range of 0.5 – 1.5 µS cm⁻¹ and the pH range of 5.5-6.5, depending on the growth rate.

Keywords: *Petunia hybrida*, Greenhouse, Fertilizers, EC, pH, hydroponics

INTRODUCTION

Petunia hybrida comes from South America and is a result of the hybridization between two different species of petunia flowers i.e. *Petunia axillaris* (a white large typed and night scented petunia) and *Petunia integrifolia* a mainly violet colored petunia (Lindergren, 2000). *Petunia hybrida* is a very popular flowering plant which is grown commercially as a pot plant. *Petunia* is seed

propagated. *Petunia* hybrid seed production is now practicing in many countries of the world. Crop duration of this plant is 5-6 months. Pollination of the flowers can be started 2 months after sowing. Mature fruits can be obtained nearly one month after pollination. Therefore to take seeds, it requires nearly 4 months. To have good quality seeds, proper care of plant during this period is essential. When *Petunia* is grown for seeds, fluctuation of Electrical Conductivity (EC) especially at the reproductive stage causes to form low quality seeds. These seeds find a low demand in the seed market due to relatively short shelf-life and low germination percentage. In floriculture industry, numerous fertilizer formulae are used in various dosages to improve the quality and yield of the flower, foliage or seed harvest (James and Iersel, 2001, Kang and Iersel, 2009). This unlimited use of fertilizers may be one of the main reasons for high EC in the potting medium (Weerakkody *et al.*, 2012). Based on this hypothesis, this research was carried out to test the influence of different EC and pH levels maintained by, different sources and dosages of basically hydroponics fertilizers on the seed quality of *Petunia hybrida*.

MATERIALS AND METHODS

The research was conducted under greenhouse conditions at Quality Seed Company (Pvt) Ltd., Boralanda (agro-climatic zone, UCIZ), during July to October in 2015. The experiment was laid out as a Complete Randomized Design (CRD) with five treatments (Trt) and four replicates where the plot size was 10 plants. Seed propagated *Petunia* variety, FPE 201 A plants were grown in 2.5 L plastic pots filled with coir dust (coco peat) based medium and with a daily fertigation volume of 250–300. The mean day and night temperatures were 23-28 °C and 14-17 °C, respectively, while the day and night RH of the growing environment was in the range of 53-76% during the experimental period.

Fertigation treatments were formulated as follows. Commercial practice of daily application of standard dosage of the several commercial fertilizer combinations (Trt 1) and the same without foliar spray (Trt 2) were compared with Trts 3, 4, and 5 which were composed of different types and dosages of hydroponic fertilizers. In Trt 3, the commercial fertilizer “Alberts” was applied in alternative days. In Trt 4, Alberts fertilizer, $\text{Ca}(\text{NO}_3)_2$ and the commercial fertilizer “Super K” were applied in alternative days. In Trt 5, triple super phosphate (5 g) (TSP) and 3 g of commercial micro nutrient mixture “Utah” were added as the basal dressing while a new mixture composed of ($\text{Ca}(\text{NO}_3)_2$, KCl, MgSO_4 , $(\text{NH}_4)_2\text{SO}_4$ and ZnSO_4) were applied in three-day intervals. The dosage of each fertilizer in all treatments were maintained at the rates of 0.2 g plant⁻¹ during first 3 weeks after planting (WAP), 0.4 g plant⁻¹ during 3-6 WAP and 0.6 g plant⁻¹ during 6th week onwards.

Crop cultivation was done under the standard practices of pot *Petunia* production except seed production. In case of seed production, emasculation was done 21 days after planting by removing stamens of the flower and hand pollination was done with the pollens collected from a male plant in the following day. Male plants were fertigated according to the standard procedure of the commercial practice (as described under Trt 1). Ripen fruits were harvested 01 month after pollination and were dried in a drier (18-20 °C Temperature, 85% RH). After pre-drying, hand cleaning of seeds was practiced and seeds were separated from the fruits.

Vegetative growth, reproductive growth and seed quality were assessed in terms of leaf number, intermodal length, branch number, number of flowers, seed germination percentage, seed vigor,

1000 seed weight, dry weight/ wet weight ratio. Data were statistically analyzed through PROC ANOVA, using software, Statistical Analysis System (SAS). Mean separation was done using Least Significant Difference (LSD) procedure.

RESULTS AND DISCUSSION

Vegetative growth and EC control

The vegetative growth of *Petunia hybrida* was similar among different treatments. There was no significant difference ($P \leq 0.05$) among different fertilizer programs on any of the vegetative growth parameters (number of leaves, internodal length, number of branches). The treatment which carried new mix of fertilizers (Trt 5) was able to keep its EC at the acceptable range (Fig. 1). In the control treatment (Trt 1), EC reached up to $1.5 \mu\text{S cm}^{-1}$ but plants appeared to be healthier. When comparing with the control, Trt 2 with foliar amendment of fertilizers has shown an additional effect on plant growth. Meanwhile, medium pH (at 1:1.5 dilution) remained at 5.58 - 6.64, favourable in all fertigation treatments.

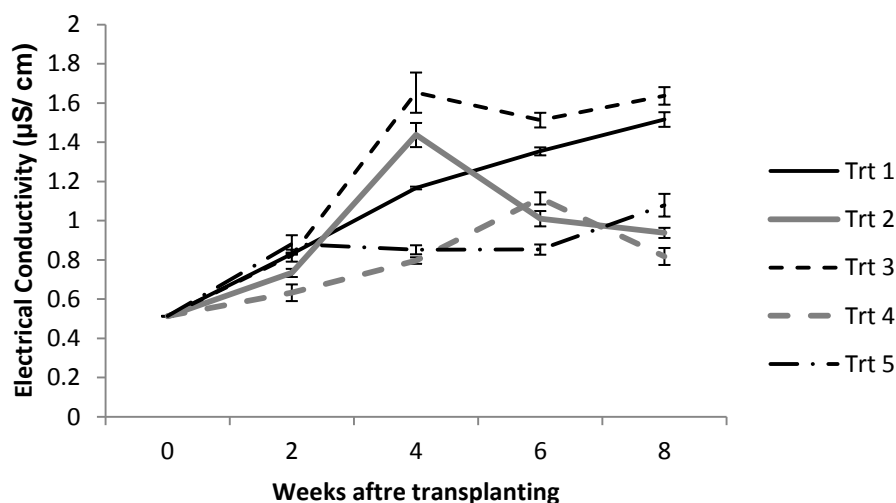


Figure 1: Variation of EC of the growing medium under variable fertigation programs of *Petunia*

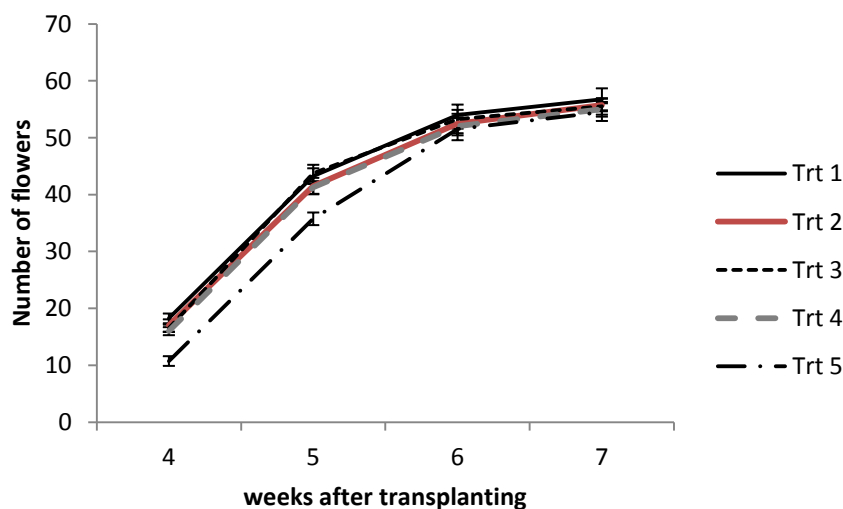


Figure 2: Early flower yield of *Petunia* under different fertigation programs

Flowering and seed quality

In all five treatments, flower initiation was observed 2 days after stopping the disbudding practice. Number of flowers per plant was significantly different ($P \leq 0.05$) in first two weeks of flowering. Significantly low number of flowers was observed in the treatment which carried new mix of fertilizers (Trt 5). However, significant difference of flower yield was not observed in later stage of flowering (Fig. 2).

When compare the control treatment (Trt 1) with the treatment which excluded foliar application of fertilizers (Trt 2), significant difference was not observed in vegetative growth or reproductive growth but no deficiency or toxicity symptoms were observed in the control either. The quality of the seeds of *Petunia hybrida* was not significantly different among treatments. Thousand seed weight was 0.082 g while the germination percentage was 88-90%.

Plant nutrient uptake and availability of plant nutrients in the medium

When considering the nutrient contents of plant and medium, the control (trt 1) showed a higher plant N content with the increasing N contents in the potting medium while Trt 5 showed a much higher P content with the high phosphorous content in the potting medium (Table 1). Plant K and Ca content did not change significantly in response to high K and Ca contents in the potting medium, respectively. Plant K content at 21 days after planting (DAP) was 4473 ± 127 ppm while it was 7500 ± 178 ppm at 52 DAP. Medium contained 1475 ± 118 and 4480 ± 253 ppm K at 21 and 52 DAP, respectively. Meanwhile plant contained 4638 ± 112 and 5850 ± 174 ppm Ca at 21 and 52 DAP, respectively.

Table 1: Plant and medium nitrogen and phosphorous contents at 21 and 52 days after planting

	21 DAP		52 DAP		21 DAP		52 DAP	
	N-Plant	N-Medium	N-Plant	N-Medium	P-Plant	P-Medium	P-Plant	P-Medium
Trt 1	36.0 ± 1.0^a	4.5 ± 0.2^a	51.4 ± 2.6^a	5.8 ± 0.2^a	1116 ± 75^b	45 ± 6.8^b	3268 ± 206^b	182.2 ± 5.1^b
Trt 2	29.1 ± 1.0^c	4.1 ± 0.1^b	41.6 ± 1.4^b	5.5 ± 0.4^{ab}	906 ± 183^b	34.5 ± 3.3^b	3415 ± 137^b	184.5 ± 10.9^b
Trt 3	31.6 ± 1.1^b	4.3 ± 0.1^b	38.0 ± 1.1^c	4.7 ± 0.1^b	1211 ± 25^b	46.6 ± 2.4^b	3576 ± 382^b	191.8 ± 6.3^b
Trt 4	31.4 ± 0.1^b	4.3 ± 0.1^b	43.2 ± 2.3^b	4.8 ± 0.1^b	878 ± 105^b	34.1 ± 6.0^b	3060 ± 706^b	140.5 ± 11.0^c
Trt 5	30.2 ± 1.2^{bc}	3.6 ± 0.1^c	41.7 ± 1.1^b	4.7 ± 0.2^b	1294 ± 110^a	61.0 ± 4.9^a	4936 ± 343^a	343.7 ± 26.8^a

Note: For treatment descriptions, please see Materials and Methods. DAP: Days after planting. Within each column, the same letters indicate no significant difference between treatments.

CONCLUSION

Petunia hybrida can tolerate moderately high EC of $1.5 \mu\text{S cm}^{-1}$ in the potting medium. High phosphorous content in the medium reduced the number of flowers of the plant. Foliar application of fertilizers didn't have an effect on plant growth. Plant nitrogen and phosphorus uptake were proportional to their availability in the medium while potassium and calcium uptake were rather stable. Plant growth rate and seed quality of *Petunia* did not respond to varying combinations and dosages of fertilizers in the fertigation program significantly within the EC range of $0.5 - 1.5 \mu\text{S cm}^{-1}$.

REFERENCES

- James E.C., Iersel M.W.V. (2001). Fertilizer Concentration Affects Growth and Flowering of Sub-irrigated Petunias and Begonias. Hort Science 36(1): 40–44.
- Kang J.G., and Iersel M.W.V. (2009). Managing Fertilization of Bedding Plants: A Comparison of Constant Fertilizer Concentrations versus Constant Leachate Electrical Conductivity. Hort Science 44(1):151–156.
- Lindgren, D.T. (2000). Petunia Hybrida, Nebraska Extension. [on line] Available at <http://www.fyi.uwex.edu> [Accessed 12 June 2015].
- Weerakkody, W.A.P., Kumara K.P.S.S., Samarakoon, S.J.M.V.L. and Chandrasiri, R.A.S. (2013). Media and water management for improved vegetative growth of gerbera (*Gerbera jamesonii*). Acta Hort. 1004: 129-134.

Abstracts

Preservation of *Tabernaemontana divaricata* (wathusudda) for floral arrangements

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Flowers are given new life using dyes to enrich the original color or to completely give blooms a new shadow. Different methods of dyeing help to color coordinate an arrangement or meet the specific needs of customers. Preservationists who like keeping blooms in scrapbooks or in display cases often dye flowers before drying. The added colorant keeps the flower from fading too much during the drying process. In this study we observed how flower dyeing and drying affect the floral appearance and shelf life for different occasions. Food coloring (mL), Turmeric (g), and cochineal (g) mixed with water in different ratios; 1:0.5, 1:1 and 0.5:1 respectively were used as flower dyeing agents. Their coloring intensities were ranked using a scale 0, 1, 2, 3, and 4 according to the flower color absorption viz color is not changed, color change is very less, color change is less, color change is moderate and color change is higher. *Tabernaemontana divaricata* (Apocynaceae) white color flowers were used in the experiment. A microwave 80 power range for two minutes was used for flower drying with silica. The mean values of given marks were recorded in all tested specimens. Experiment was arranged according to three factors factorial Completely Randomized Design (CRD) and comprised of four treatment sets, including control and each treatment having three replicates. Statistical analysis was performed using ANOVA followed by the Duncan's Multiple Range Test using SAS software (version 9.1.3). No color changes were observed in Cochineal and Turmeric treatments. Higher color intensity was observed in food coloring 1 ml: water 0.5 ml kept in more than 12 hours.

Keywords: *Tabernaemontana divaricata*, cochineal, foliage, dyeing, drying

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Progress of the “Suwahas Mal” flower societies in the Gampaha District, registered in the Department of National Botanic Gardens from 2005 to 2015

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Department of National Botanic Gardens implemented ‘Suwahas mal’ programme in 2005 to enhance the social economic states of small and medium level flower growers in Sri Lanka. The main objectives of this programme were to generate long term income and investment of small and medium scale flower growers, to organized flower growers at correct manner, to build up new technical knowledge of cultivators, to distribute planting materials, shade nets, polythene and other infrastructure facilities, to enhance local markets for flowers and to earn foreign exchange. Basically this project was established within five provinces (Western, Southern, Central, Northwest and Sabaragamuwa) covering ten Districts. Gampaha district has the most favorable climatic condition and marketing facilities for floriculture sector and therefore ten societies were established within 10 agrarian services centers in the Gampaha district in 2005. At present, thirty societies are organized in Gampaha district. This study was conducted to evaluate the improvement of floriculture in the Gampaha district. Results showed that the increase of societies were from ten to thirty, increase of members in societies was from 430 to 1700 within ten years. Fifty-six Acres of land area under flower cultivation in 2005, has been increased up to 94.1 Acres with coverage of net house 8.4 Acres which distributed under Suwahas programme. Of all, 73.6% growers use 0-25 perches, 13.7% growers use 25-50 perches, 5.6% growers use 50-75 perches, 4.5% growers use 75-100 perches, 2.6% growers use more than 100 perches only for cultivation of flowers in Gampaha district. Of the total respondents, 17.9% were males and 81.1% were females, contributed to cultivation. Considering age limit of growers, 18-30 years were 7.8%, 30-40 years were 21%, 40-50 years were 29.4%, 50-60 years were 26.4% and more than 60 years were 15.4%. Analyzing different cultivars, 41.7% growers cultivate Anthurium, 33.2% growers cultivate Foliage plants, 12.9% of growers cultivate Orchids, 9.6% growers cultivate ornaments and 2.6% growers cultivate palms and others in Gampaha district.

Keywords: *Suwahas mal societies, Gampaha district, floriculture*

Developing a disease control method for *Zamioculcas zamiifolia* using beneficial microbial communities

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This study was conducted to investigate the disease causing microbes of *Zamioculcas zamiifolia* (Zz) and then to isolate and identify bacterial/fungal species from different plant parts, and to develop a disease control method using beneficial microbial communities. To develop a disease control method, endophytic and epiphytic microbes were first isolated from different plant parts and tested for pathogenesis. Then, they were screened for organic acid production (i.e. pH) and growth in combine carbon medium (CCM), which indicated presumptive diazotrophs. Nitrogenase activity of the selected bacteria from above was evaluated by acetylene reduction assay. Antagonistic activity of the selected fungal and bacterial isolates above was investigated using the dual culture method. Monocultures selected from above screening methods were used for developing microbial communities or biofilms, which were subsequently used as biofilmed biofertilizers (BFBFs). Then, another experiment was conducted in the greenhouse to examine the effect of BFBFs on Zz plant. Microbial isolates of effective BFBFs and pathogens were identified using their morphologies and biochemical tests. The BFBFs showed their ability to control leaf blight disease of Zz, and also to improve quality of the plant, thus illustrating their potential to act as biocontrolling agents too. Pathogens causing leaf blight disease of Zz were two *Pseudomonas* spp. and a *Bacillus* sp.

Keywords: biofilmed biofertilizers, *Zamioculcas zamiifolia*, biocontrolling agents

Effect of different levels of nitrogen on plant biomass and flower diameter of jasmine (*Jasminum sambac* L.) cv. 'Local' in the Batticaloa District

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An experiment was carried out to evaluate the effects of different nitrogen levels on plant biomass and flower diameter of Jasmine (*Jasminum sambac* L.) in Batticaloa district. The experiment was arranged in a completely randomized design with three replications. Graded levels of nitrogen were defined as treatments viz. 0 (T1), 50 g (T2), 100 g (T3), 200 g (T4), 300 g (T5), and 400 g (T6) of nitrogen/plant/year. Phosphorous and potassium levels were kept constant and applied as basal dressing. Urea was used as nitrogen source and split application was practiced at monthly interval. Agronomic practices were followed uniformly for all treatments. Plant biomass and flower diameter were measured at monthly intervals. Analysis of Variance was performed to determine significant difference among treatments ($p < 0.05$). Results revealed that plant biomass and flower diameter were significantly higher in T4. Plants grown at this nitrogen level would have received optimum amount of nitrogen. Therefore biomass and flower diameter of Jasmine was higher at this treatment. Through polynomial regression analysis, it was found that, optimum nitrogen level for Jasmine was 243.75 g/plant/year. From this experiment, it could be concluded that nitrogen level of 243.75 g/plant/year applied in split at monthly interval is optimum for growing Jasmine in Batticaloa district of Sri Lanka.

Keywords: Flower diameter, Nitrogen level, Plant biomass, Polynomial regression analysis.

Morpho-phenotypic plastic responses in *Gerbera jamesonii* in response to light heterogeneity

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Gerbera jamesonii is a popular cut flower, pot plant and a decorative plant in the international floriculture trade. Though recommended light levels for cultivation of *Gerbera* are well established, Sri Lankan growers cultivate *Gerbera* under varying light intensities. This study attempted to test whether the leaf and flower morphological traits are plastic in response to light heterogeneity and to identify the interrelationships among leaf and flower parameters in *Gerbera*. Leaf and flower parameters were measured in twenty different commercial farmer fields in Nuwara Eliya and Bandarawela Districts. Number of leaves, leaf length, leaf width, petiole length, leaf area and leaf color grade were taken as the leaf parameters, whereas number of flowers, flower diameter, length of ray florets, center diameter, angle between flower and pedicel and color grade of flower were measured as the flower parameters. The light level inside and outside of the net house was measured at each location in bright sunny days. Correlation analysis was performed to see the interrelations among variables. Interestingly, the shade percentages in *Gerbera* cultivations sampled in the study varied from 27 to 70%. The results revealed that the shading has a significant effect on some of the leaf and flower parameters of *Gerbera*. Leaf length, leaf width and leaf area showed moderate relationships with shade heterogeneity ($p < 0.05$, r^2 0.13-0.67) while fitting to a quadratic model and, petiole length did not show a correlation with shade heterogeneity ($p > 0.05$). Flower diameter and pedicel length showed moderate relationships with shade ($p < 0.05$, r^2 0.3-0.6) fitting to a quadratic model. Leaf length and width appeared to be plastic in response to change in light levels ($p < 0.05$, r^2 0.22-0.64). Leaf parameters showed interrelationships while fitting to a quadratic model. As well-known, leaf area is increased as an adaptive response under limiting light levels. Flower diameter, center diameter, pedicel length and length of ray florets were significantly interrelated to each other ($p < 0.05$, r^2 0.4). The flower parameters in *Gerbera* were interrelated to each other as a plastic response, to maintain quality flower characters under varying environment conditions. The optimum shade level was found to be ~50% to maintain high quality cut flower production for *Gerbera* in both districts. However, *Gerbera* can be recognized as moderately plastic in morphological characters in response to shade heterogeneity.

Keywords: *Gerbera jamesonii*, light heterogeneity, phenotypic plasticity, shade tolerance

1-Methylcyclopropene (1-MCP) on the postproduction quality attributes of rooted cuttings of *Pleomele reflexa*, *Codiaeum variegatum*, and cut greens of *Scindapsus aureus*

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Rooted cuttings of *Pleomele reflexa*, and *Codiaeum variegatum*, and cut greens of *Scindapsus aureus* are regularly exported from Sri Lanka. To earn high profits, these types of commodities should be transported without quality deterioration to the markets overseas. During overseas transportation, plants are subjected to accelerated ethylene, causing postproduction losses such as leaf yellowing, leaf wilting and chlorophyll degradation. This has become a major obstacle to earn maximum profits from the international market. 1-MCP, an inhibitor of ethylene reception, prevents or delays leaf abscission and chlorophyll degradation in a wide range of plant species. However, in Sri Lanka, 1-MCP has not adequately been tested on ornamental foliage. Thus, this study was conducted to identify the effective dose of 1-MCP on postharvest quality attributes of ready-to-ship rooted cuttings of *Pleomele reflexa* 'Song of India', *Codiaeum variegatum* var. *pictum* (Lodd.) Muell. 'Aucubaefolia' and cut greens of *Scindapsus aureus*. Different concentrations of 1-MCP and exposure times (0, 0.5, 1, 1.5 ppm for 12 h or 24 h), (0, 1, 3 ppm for 24 h) and (0, 1, 3, 6 ppm for 20 h) were used for *P. reflexa*, *C. variegatum* and *S. aureus* respectively. After treating with 1-MCP, the plant materials were packed and kept in dark to mimic the export transport conditions. The experimental periods for these three species were 25, 22 and 12 days respectively. Chlorophyll content was not significantly different ($p>0.05$) among treatments and exposure times in *P. reflexa*. The present study shows that there was no significant effect of 1-MCP for 12 h on leaf retention in both *P. reflexa* and *C. variegatum*. However, same concentrations showed a significant ($p<0.05$) effect on increasing leaf retention with 24 h treatment. Rooted *C. variegatum* plants had shown significantly higher ($p<0.05$) leaf retention percentages compared to control, 22 days after treatment. There were significantly lower mean scores ($p<0.05$) for leaf yellowing compared to control in both type of storage (with a water base by wet cotton plug and without a water base) of *S. aureus* cut greens. Of all treatments, 3 ppm 1-MCP showed the least yellowing in cut greens with a water base and 1 ppm without a water base. There was no any significant difference in mean scores for wilting between control and treatments when the cut greens are with a water base. Cut greens of *S. aureus* without water base showed significantly lower ($p<0.05$) mean scores for wilting compared to control. It was proved that there is a positive effect of 1-MCP treatment in preserving quality attributes of rooted cuttings and cut greens in exports. Thus, 1-MCP can be suggested as an economically and practically viable postharvest remedial measure to the Sri Lankan exporters.

Keywords: 1-Methylcyclopropene, rooted cuttings, cut greens, postproduction.

Glycerin preservation and microwave drying to enhance the keeping quality and exactness of differently textured flowers

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Flowers are Perishable in nature thus very difficult to store for long period of time. Therefore, it is important to introduce flower preservation methods and modify currently available techniques favorable for flower drying. There are many problems related to flower drying because drying change natural color and texture, the shape and exactness, in particular. In most cases, the final color can be a light shade of dead brown. It is a major concern when flowers are used for decorations. Other problems include, brittle texture and shape change of dried flowers, making these difficult to handle. Modified glycerin preservation technique can be used as a solution to overcome these problems. In the present experiment, three differently textured flowers were evaluated by using glycerin with microwave drying to minimize the color and textural changes. The used flowers were, *Anthurium andrianum*, *Bougainvillea glabra* and *Gerbera hybrida*. Treated Glycerin concentrations were 1, 3 and 5% with red color food dye. Flowers were treated with combined solutions for three days and finally microwave-dried for three minutes. The results were analyzed using SAS. According to the results, glycerin with dye treated flowers showed a significant difference ($p < 0.05$) in their shape, exactness and color. With all three types of flowers, when treated with glycerin, petal ends were rolled and petals became more pliable and became smooth compared to water treated flowers. In high glycerin concentrations (5%), petal rolling was higher than low glycerin concentrations (1%). When microwave drying was done, flower shrinkage and color fading was observed. However, dye treated flowers were not faded remarkably. Glycerin helps to increase the color absorption rate thus, dye absorption is needed to practice after dissolving in glycerin solution. When flowers are microwave-dried after glycerin treatment, the color fading and browning can be reduced due to high heat, and the reduction of effective surface area of a particular flower can also be reduced. However, due to glycerin, flower brittleness is increased after microwave drying. Only glycerin treatment without microwave drying minimizes the brittleness and flowers become soft and pliable. Flowers with hidden blemishes and damages on the petals cannot be preserved by the Glycerin+dye treatment, because it causes scars on flower petals. From the tested flower types, Gerbera outperformed the other two in all the aspects investigated.

Keywords: *Anthurium*, *Gerbera*, *Bougainvillea*, *Glycerin*, *Microwave drying*.

Potential of *Murraya koenigii* (karapincha) in indoor decorations

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Indoor plants contribute to improve indoor environmental quality through providing aesthetic value, reducing urban air pollution and producing some useful volatile compounds. *Murraya koenigii* (Karapincha) is a highly valued plant for its medicinal value and characteristic aroma. The plant is a rich source of volatile compounds which brings health benefits to human. Potential of *M. koenigii* as indoor plant was studied in three aspects as vegetative propagation ability, early flowering ability and shade tolerant ability. Stem cuttings in three different maturity stages (hard wood, Semi hard wood and soft wood) were tested to select best propagation materials for pot plant production and flowering. Shade tolerant ability of acclimatized and non-acclimatized potted *M. koenigii* was studied by placing them under five different shade levels 10±5 lux, 50±10 lux, 500±50 lux, 1500 ± 100lux and 5,000-10,000 lux for two weeks and leaf shedding percentages were calculated. Non-acclimatized *M. koenigii* plants kept below 500lux light intensity started leaf shedding at 3rd day and increased gradually. Average leaf shedding per day during first week were 9.2%, 4.9% and 3.1% at the light levels 10±5 lux, 50±10 lux and 500±50 lux respectively. Plants kept under 1,500 ± 100lux and open environment (5,000-10,000 lux) did not show significant leaf shedding. Plants acclimatized at 1,500-2,000lux did not show significant leaf shedding at indoors with 500 lux for 2weeks. At the end of 3rd week of establishment 75% & 68% from semi hard wood & soft wood cuttings showed root initiation while 18% hardwood cuttings had initiated rooting. 96% of rooted semi hard wood turned to flowering pot plants at 3 months of age when transfer to full sun light from shade (1500 ± 100lux). It can be concluded that *M. koenigii* has potential as indoor plant in aesthetic as well as human health aspects.

Keywords: *Murraya koenigii*, Shade tolerant, leaf shedding, acclimatization

Selection of foliage plants for indoor decorations based on the rate of CO₂ emission/absorption and stomatal density

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Use of foliage plants for indoor decoration is a pursuing tradition in several cultures. With contemporary living patterns, frequent replacement of indoor plants has become impracticable. Therefore, this experiment was conducted to identify indoor plants with low CO₂ emission or high CO₂ absorption ability in nights to keep in indoors continuously. Five common indoor plants (*Cryptanthus sp.*, *Dieffenbachia seguine*, *Dracaena sanderiana*, *Sansevieria trifasciata* and *Zamioculcas zamiifolia*) were placed in 1000 L concealed barrels separately for 12 hours in dark. The CO₂ level in each barrel was measured before and after the experiment and balance was calculated. The stomatal count of both upper and lower leaf surfaces was taken in each plant sample and opening width of stomata were measured in dark and light periods of the day to determine the relationship between CO₂ emission / absorption efficiency, stomatal density of tested ornamental species. From the selected species *D. seguine*, *D. sanderiana* and *Z. zamiifolia*, showed positive CO₂ balance in the barrel and CO₂ increments were 0.16, 0.39 and 0.18 ppm cm⁻² of leaf area respectively. Both *Cryptanthus sp.* and *S. trifasciata* showed negative CO₂ balances as around - 0.20 ppm cm⁻² of leaf area. *S. trifasciata* possesses stomata in both upper and lower leaf surfaces while stomatal number in upper surface of all other 4 species was negligible. Average stomatal numbers were 5.56x10⁴, 5.03x10⁴, 9.05x10⁴, 5.25x10⁴ and 3.51x10⁴ cm⁻² in *Cryptanthus sp.*, *D. seguine*, *D. sanderiana*, *S. trifasciata* and *Z. zamiifolia*, respectively. Stomata in *Cryptanthus sp.* and *S. trifasciata* were closed during day time and open in the night. Present study concluded that potted *Cryptanthus sp.* and *S. trifasciata* (plants with CAM photosynthesis pathway) used for indoor decoration absorb CO₂ during the night, hence safe to keep in indoors.

Keywords: foliage plants, CO₂ emission / absorption, *Cryptanthus sp.*, *Dieffenbachia seguine*, *Dracaena sanderiana*, *Sansevieria trifasciata* and *Zamioculcas zamiifolia*

Leaf reddening and browning problem in *Nepenthes*

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Nepenthes, popularly known as tropical pitcher plant or monkey cup, is a genus of carnivorous plant in the monotypic family Nepenthaceae. *Nepenthes*, a native of Southeast Asia and Australia, forms pitchers (cups) that hang from trees. The genus *Nepenthes* is mostly found within the Malay Archipelago with the greatest bio diversity found on Borneo, Sumatra, and the Philippines, especially in the Borneo montane rain forests. Most species are restricted to very small ranges, including some only found on individual mountains. These limited distributions and the inaccessibility of the regions often means some species go decades without being rediscovered in the wild. Most *Nepenthes* species grow in environments that provide high humidity and precipitation and moderate to high light levels. Because of the nature of the habitats which *Nepenthes* species occupy, they are often graded as either lowland or highland species. Sun scald is a common problem with *Nepenthes*, but usually not fatal unless extreme. Damping off, sooty mould and root rot are the recorded diseases in *Nepenthes* so far in the world. Most literature has recorded red rash on the leaves but the causal agent has not identified yet. Very recently in Sri Lanka *Nepenthes* grower, Borneo Exotics noticed a reddening and browning of leaves which leads to reduce market quality and effect the growth of the plant. Initial symptom was red or brown spots on the leaf then it spreads throughout the leaf. Causal agent of this disorder is not known. Therefore this study was conducted to identify the causal agents of this problem and to develop a suitable management strategy. Disease samples were cultured on potato dextrose agar (PDA) and nutrient agar (NA) medium containing Petri plates and incubated 5 days at 28-30 °C. Microscopic observations with morphological features and colony characters were carried out to identify the isolated fungus spp. In vitro plate test was conducted to find the efficient chemical to control the isolated fungus. Treatments T1- Captan, T2- Cu oxichloride, T3 – Mancozeb, T4- Captan + Cu oxichloride + Mancozeb, T5 – control. Diameter of the fungus colony was measured in weekly intervals. According to the morphological features and colony characters *Colletotrichum* spp was identified. Captan + Cu oxichloride + Mancozeb mixture showed better reduction of mycelium growth under in vitro condition.

Keywords: *Colletotrichum*, in vitro, leaf reddening, *Nepenthus*, symptoms

Demand and supply of floriculture products in the Colombo District

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The general objective of this study was to analyze the demand and supply of floriculture products in the Colombo District. Floriculture is the study of growing and marketing flowers and foliage plants. It is a high income generating agri-business and can open up great opportunities to growers in Sri Lanka encompassing both domestic and international trade. Consumers buy floricultural products because of traditions, culture and lifestyle. Buyers require products with constant quality, price and added value. Therefore a study was conducted to find the present status of supply of cut flowers and the demand of cut flowers for the local market. This survey research was carried out from June to November in 2015 in the Colombo district. Twenty-Five sales outlets were visited and personal interviews were conducted by using interview schedule. Collected data were analyzed statistically. The study showed that demand for flowers is seasonal. Furthermore, demand for flowers had two components: a steady component and a seasonal component. The Steady demand for flowers comes from the use of flowers for religious purposes, decoration of homes and for making garlands and wreaths. The bulk of seasonal demand comes from festivals and marriages and is generally for specific flowers. Results of the survey showed that the supply was insufficient to meet the demand.

Keywords: *Floriculture, Demand, Supply*

Status of women in floriculture industry: Western province

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Floriculture supplies and exports facilitate 5000 direct employments and over 15,000 indirect employments in Sri Lanka. Hence not surprisingly, able present itself as a prominent candidate in path towards being the countries' major foreign export venture, concentered by the noticeable upsurge in this industry. Floriculture industry has impacted rural farmers, centralizing focus on female florists. Sri Lanka offers only a slight glimpse of research on social aspects of women in floriculture: however, Srikrishnah, 2011, identified a pattern whilst studying Batticaloa district, majority of growers were female operating on part-time basis with reliance on family members for support. Researchers attempt to focus on a solitary fragment of women entrepreneurs and they're lifestyle. Sample focuses on Colombo district, 108 participants from Colombo, 110 from Kalutara, and 102 from Gampaha: overall 320 women operating in the floriculture industry. Core research method comprises of quantitative questionnaire surveys and data analysis conducted utilizing parametric testing approaches, in assistance with computer aided analysis tool (SPSS). Findings provided an in-depth view into hindered segment of the floriculture industry, where most participants' were in fact married women past the age of retirement (over 50 years old) or concentrative of 'unemployed human resource'. Over half a century of experience and networking is indicative to their survival in the floriculture industry. Satisfaction and emotional joy experienced in an instance a flower blooms has offered these women a direction, a motivation to dream, and socially bond: humane implications uncovered by researchers demonstrate a unique perspective into the lives of women in floriculture. Researchers note a significant relationship between floriculture pockets and their geographical location: majority in urban suburbs. The bridging destination amidst the urban and sub-urban locations hence offer both access to land and urban markets. Therefore, this research provides an insight for policy implementations to upgrade the lifestyles of women growers in Sri Lanka.

Keywords: *Women, Floriculture, Sri Lanka*

Present status of flower growers of the 'Suwahas Mal' programme in Kurunegala District

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The "Suwahas Mal" Programme was launched by the then Division of the Botanical Gardens, and presently Department of National Botanic Gardens about ten years ago. Since then it has spread island-wide. The objective of carrying out the survey was to assess the present situation of the flower growers coming under the programme in the Kurunegala district. For this purpose, the entire population of the growers of ten flower grower's societies was individually interviewed, using a questionnaire. Data were analyzed using Microsoft Excel. The survey results showed that the majority (85 %) of the growers were female, with males comprising only 15 %. Most of the growers were between the age group of 40-50 years. It was also found that 50 % of the growers earned less than Rs. 10,000 (net income) per month, while 14 % earn more than Rs. 30,000/- per month. Anthurium is the major flower crop cultivated by these growers while orchids ranked next. 65% of the growers carried out all cultivation related the work on their own without engaging additional external human resources. Important problems faced by growers as perceived by them were marketing issues, low infrastructure facilities, pest and disease incidence, high cost of mother plants and other planting materials as well as scarcity of quality planting material and new varieties.

Keywords: Kurunegala district, "Suwahas Mal" programme, flower growers

Evaluation of different commercial available fertilizers for flowering of *Ixora coccinea* in cooler climates

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Ixora coccinea is a tropical landscape plants belong to family Rubiaceae. There is a good potential for the expansion of cultivation of *Ixora* as a landscape plant in Sri Lanka. *Ixora coccinea* flower production in cooler areas of Sri Lanka is very poor. The reasons for this insufficient production is, mainly due to lack of warm and sunny conditions. *Ixora* is grown well in warm climate and it's difficult to grow in cooler climates. Therefore *Ixora* cultivation is done in few areas of Sri Lanka. Therefore this study was planned to find out the most suitable commercially available fertilizers and their concentrations for enhancing the flower production of *Ixora* in cool areas. Two experiments were conducted using two *Ixora coccinea* varieties (red color and orange color) and two levels of eight commercial fertilizers (15:7:20, 14:14:21, 14:10:18, 8:15:30, 13:2:44, 6:30:30, 10:20:30, 10:30:44) and control as without applying fertilizers. Eight week old rooted, 8 cm height plants were used as planting materials and top soil 4: Compost 2: Sand 1: Cow dung 1: Brick¼: Charcoal ¼ were used as growing medium for both experiments. The effectiveness of those treatments was determined by measuring the time taken to first flowering, number of spikes per plant, number of flowers per spike, life span of the spikes and data were recorded at day to day observations. The treatments were arranged as Two Factor Factorial Completely Randomize Design with three replicates. In experiment one, the number of flower per spike and the number of spikes per plant of *Ixora coccinea* were significantly ($p < 0.05$) increased with 8: 15: 30 granular fertilizer low level (1.5g /plant) and the time taken for first flowering of *Ixora coccinea* were significantly increased with 13: 2: 44 foliar fertilizer low level (3g/ l). The life span of spike was not increased significantly at any fertilizer treatment. In experiment two, the number of flowers per spike and time taken for first flowering of *Ixora coccinea* were significantly increased with 8: 15: 30 granular fertilizer low level (1.5g /plant) and the number of spike per plant were significantly increased with 14: 14: 21 granular fertilizer low level (1.5g/ plant) and the life span of spike were significantly increased with 10: 20: 40 foliar fertilizer low level (0.85 ml/ l). The poor flowering performance of *Ixora coccinea* was show 6: 30: 30 foliar fertilizer low level (0.65g/ l) and 8: 15: 30 granular fertilizer low level (1.5g /plant) was better fertilizer for flowering of *Ixora coccinea*

Keywords: *Ixora coccinea*, flowering, fertilizer, cooler climate

Variation of flower morphology of endemic plant *Osbeckia octandra*: a case study in Kamburupitiya, Matara

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Osbeckia octandra (*Heen Bovitiya* in Sinhala) is an endemic perennial shrub belonging to Family Melastomataceae. *Osbeckia octandra* produces an attractive purple flower which could be a candidate in future floriculture industry in Sri Lanka. This study was carried out to determine the variation in flower morphology from selected locations around Kamburupitiya in southern Sri Lanka. Ten quantitative characters (of petal number, width and length, flower length and width, stamen number, sepal number, peduncle length, OD value of pigment extract from a unit petal length and vascular pH of pigment extract) were recorded from 24 accessions from 10 random locations. The color of pigment extract in different pH conditions was also recorded. Petal number varied from 5 to 6 among 24 accessions while number of stamens varied from 4-5. Number of sepals varied from 5 to 6. Petal extract in 0.1M NaOH turned blue indicating that the pigment of *Osbeckia octandra* is anthocyanin. Light microscopic view of the petal vertical cross sections indicated pigments were mainly restricted to the outer most hypodermal layers. The vascular pH was around 3.8. The OD values at 525 nm wave length of the anthocyanin extract was 0.024 per 1 g of fresh petal weight. With the increase of pH in pigment extract turned the purple color to light brown and then to green in basic pH conditions. Our study would be useful as an initial step in potential morphological and genetic variation of *Osbeckia octandra* for variety of flowers.

Keywords: Flower, morphological variation, *Osbeckia octandra*

Recent pest infestation reported in local orchid cultivations

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An insect pest of recent origin has been reported from many parts of the island causing much damage to Orchids cultivations specifically at the blooming stage. Initial reports of this pest infestation in Sri Lanka were in 2012. Symptoms reported were bud blast, brownish spots that appear chewed, holes in buds and floral tissue as well as premature bud drop. It was frequently reported in Dendrobiums while fewer records on Phalaenopsis and Cattleya have also been obtained. The insect pest was identified as *Contarinia maculipennis* the Orchid Blossom Midge. A fungal infestation was also reported subsequent to damage by the insect pest, the fungus was identified as *Fusarium proliferatum* (Dissanayake et. al. 2014). This insect pest of recent origin has been reported from Gampaha, Nainamadama, Wennappuwa, Sooriyawawa, Mirijiwela and Peradeniya in the Western, North Western, Southern and Central provinces. It has been reportedly difficult to control while causing much damage economically as well, since flowers are affected at the bud stage and cannot be sold or used for decorative purposes. Control with integration of chemical, cultural and physical methods such as spraying of insecticides coupled with use of light traps/ greased yellow polythene traps as well as removing and safely destroying inflorescences is recommended.

Keywords: *Contarinia maculipennis*, Orchid, Orchid Blossom Midge, chemical method, cultural method, physical method

A simplified method for rapid multiplication of lily plantlets *in-vitro*

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Lily, one of the important cut flowers in floriculture industry, is propagated through bulbs and bulblets. However, major problem in commercial cultivation is the unavailability of disease free planting materials in large scale. Systematic build up of the pathogens due to vegetative propagation adversely deteriorates the crop performance generation after generation (Manish 2014). Therefore, one of the possible alternatives is the use of tissue culture techniques in mass production of disease free planting materials. The bio-reactor system has been advanced as such alternative to produce transplants in mass scale. However use of such systems is limited due to its complexity and expensiveness. Therefore, this experiment aims to study the possibility of using a simplified bio-reactor system to multiply lily plants *in-vitro*. The experiment was conducted at the tissue culture laboratory in RARDC, Bandarawela. Lilly bulb Scales were used as explants, and were sterilized using 20% Clorox for 10 min, 0.2% Mercuric Chloride for 3 min and 70% ethanol for 1 min consecutively. Then scales were established in solidified ½ MS medium supplemented with 0.5 mg/L BAP, 2mg/L NAA and 3% sucrose. After 8 weeks, bulblets were transferred to solidified MS medium supplemented with 0.5 mg/L BAP, 2mg/L NAA and 6% sucrose. A simplified bio-reactor (continuous emersion with aeration) was used to multiply the shoots. In solid medium average of 4.3 shoots/bulb was obtained while 6.2 shoots/bulb was obtained with the simple bio-reactor. Plantlet height in the liquid system was significantly higher (9.35 cm) when compared to solidified medium (4.45 cm). In solidified medium, plants have to be sub cultured after two months, whereas in liquid aeration system liquid medium can be poured to the culture vessel. Therefore this system can be used effectively to multiply Lilly plants continuously.

Keywords: *Lily, in vitro, simplified bio-reactor*

Response of NPK on flower yield of *Symphytotricum lanceolatum* in commercial cultivations

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Symphytotricum lanceolatum (Super Daisy) is a species of flowering plant in the Aster family, which has a considerable value as fillers in floral arrangements. It is commercially cultivated in up country areas, where the climatic condition is highly suitable for quality flower production. However, studies on nutrient management in commercial cultivation are limited and fertilizer recommendations are not available. This study was conducted to find out the response to NPK on flower production. Three nitrogen levels (0, 100, 200 kg/ha), three P₂O₅ levels (0, 50, 100 kg/ha), and three K₂O levels (0, 75, 150 kg/ha) were applied in twenty seven treatments using experimental design of 3³ confounded block design. Irrespective of the nutrient, number of primary stems, and inflorescence height increased with the increment of NPK level. When NPK levels increased from 0 level to level 2, number of primary stems increased from 2.2 stems/plant to 2.8 stems/plant, and stem length increased from 30.6 cm to 37.4 cm. Response of super daisy to application of N is also significant. Higher number of primary stems (3.8 stems/plant) and highest inflorescence length (46.7 cm) was obtained with the application of 100, 100, 0 kg/ha NPK levels. However, number of secondary branches per inflorescence was higher in NPK levels of 0, 50, 75 kg/ha.

Keywords: *super daisy, NPK fertilizer, nutrient management, fertilizer response*

Response of selected cultivars and fungicides on chrysanthemum white rust, caused by *Puccinia horiana* (henn)

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White rust disease is one of the most serious diseases in Chrysanthemum (*Chrysanthemum morifolium*) cultivations. Susceptibility of chrysanthemum cultivars to white rust, caused by *Puccinia horiana* (Henn) is varied with cultivar and use of fungicides is one effective way to control this disease. This study focused on rating selected chrysanthemum cultivars based on resistance to white rust and identification of effective fungicides to control it. Selected 35 chrysanthemum cultivars for cultivar screening and BW-ch-Ac-045 plants for fungicide screening were maintained in a polytunnel. *Puccinia horiana* was artificially inoculated and kept under dark moist condition for 24 hrs. Fungicides spraying to BW-ch-Ac-045 plants were started with the onset of disease and continued until flowering. Chlorothalonil 500 g/L SC, Tebuconazole 250 g/L EW, Hexaconazole 50 g/L EC, Propiconazole 250 g/L EC, Pyraclostrobin 250 g/L EC, Pyraclostrobin 5% + Metiram 55% WP and their combinations were evaluated. Flower height, number of flowers per bunch, plant height, number of leaves and disease severity were recorded. Results categorized 12 cultivars for immune, 19 cultivars for very resistant, 1 cultivar for resistant, 2 cultivars for moderately resistant and 1 cultivar for susceptible. Pyraclostrobin 250 g/L and Pyraclostrobin 5% + Metiram 55% WP followed by Chlorothalonil 500 g/L SC significantly reduced the white rust compared to others. Tebuconazole showed some phyto-toxic effect on plant growth and flower yield.

Keywords : *Puccinia horiana, cultivars, resistance, control*

Effect of different calcium chloride concentrations in Murashige and Skoog basal medium for germination of *Exacum ritigalensis* (Binara/Ginihiriya) in *in-vitro* culture

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Exacum ritigalensis (Binara/Ginihiriya) is an endemic plant in which seed germination is difficult in a saline medium. Present study was carried out to evaluate the suitable calcium chloride concentration in Murashige and Skoog basal Medium for germination of *Exacum ritigalensis* under *in-vitro* conditions. Pods of *Binara* were collected from Pannala in Kurunegala district, Sri Lanka and washed under running tap water with liquid soap. Thereafter pods were sterilized with 70% ethanol for 3 minutes followed by 20% Clorox (Sodium hypochloride) for 20 minutes. Sterilized pods were then rinsed three times with sterilized distilled water and dried on sterile filter papers before removing the coat under aseptic conditions. Seeds were introduced to glass tubes (10 seeds per tube) containing 3 mL of hormone-free Murashige and Skoog (MS) basal medium with different concentrations of Calcium Chloride (CaCl_2 : 0, 25, 50, 75 and 100%). Calcium Chloride concentration was calculated according to ratio of 4.4 gL^{-1} stock solution and the medium was balanced using distilled water. The seeds were cultured under light in an air-conditioned room for 14 days. The lengths of the aseptically raised plant seedlings and number of seeds germinated were recorded after four weeks period. Experiment was repeated three times. Statistical analysis was performed using SAS software (version 9.1.3). Highest germination percentage (90%) as well as the highest seedling height (0.48 cm) was observed in the medium with 50% CaCl_2 indicating that 50% CaCl_2 in the MS medium as the best concentration for seed germination of *Exacum ritigalensis*.

Keywords: *calcium chloride, Binara, Murashige and Skoog basal Medium, Seed germination*

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**Effect of natural potting mixtures on the germination of *Exacum ritigalensis*
(Binara/Ginihiriya)**

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Binara (*Exacum ritigalensis*) is one of the endemic plants which grows in the dry intermediate zone in Sri Lanka. There are conservation concerns over this species because they are not widespread and may be confined to only one or two protected areas. Planting Binara in natural habitats is difficult because they are only adapted to special environmental conditions. The present study was conducted to find the appropriate potting mixture for seed germination and growth of *E. ritigalensis* plant. Experiment was conducted using seven different potting mixtures; river sand only (control), and 1:1 combination of river sand with dried cow dung, poultry litter, goat manure, Teak (*Tectona grandis*) bark compost, Albizia (*Albizia lebbek*) bark compost and leaf degraded compost. Pots were wetted and twenty seeds were introduced to each pot. They were covered in transparent polythene and kept under shade for four weeks. Numbers of germinated seeds were recorded. Completely Randomized Design (CRD) with five replicates was used for study. Statistical analysis was performed with ANOVA procedure followed by Duncan's multiple range test using SAS software (version 9.1.3). Seed germination percentages were significantly different among treatments. The seed germination was observed on the media river sand (95%), river sand with compost (75%) and river sand with Albizia (10%). River sand with cow dung, poultry litter, goat manure and Teak bark compost did not result in any seed germination. Study revealed that river sand is the best potting mixture for seed germination of *E. ritigalensis* among treatments tested in the present study.

Keywords: *Exacum ritigalensis*, potting mixture, seed germination, *Tectona grandis*, *Albizia lebbek*

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