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Short Communication
Propagation of *Dillenia suffruticosa* (Griff.) Martelli Stem Cuttings Using Plant Hormones: A Promising Approach to Supply Plantlets to Revegetate Degraded Tropical Heath Forests in Brunei Darussalam

[1]S. Matali, [1]N. Abidin, [1]W.H. Tuah, [2]A.M.Q. Pg Yusof, [1]H.H. Mohd Din, [1]H. Taha, [1]R.S. Sukri and [1]F. Metali

[1]Environmental and Life Sciences Programme, Faculty of Science, Universiti Brunei Darussalam, Jalan Tungku Link, BE1410, Brunei Darussalam
[2]Department of Biotechnology, UCSI University, No. 1, Jalan Menara Gading, UCSI Heights, Cheras, 5600 Kuala Lumpur, Malaysia

Abstract

Background and Objective: *Dillenia suffruticosa* (Griff.) Martelli is abundantly thriving in various forest types in Brunei Darussalam, including the tropical Bornean heath forests. This study aimed to investigate the plant hormones that can be used to propagate stem cuttings of *D. suffruticosa*, which is a promising approach to supply plantlets to revegetate degraded tropical heath forests. **Methodology:** Stem cuttings of *D. suffruticosa* were treated using three single auxin hormone: 0.1% Indole Acetic Acid (IAA), 0.1% Indole Butyric Acid (IBA) and 0.2% Naphthalene Acetic Acid (NAA) and 5 commercially available rooting hormones: Yates, Clonex and Seradix 1, 2 and 3 and planted in a medium containing a mixture of equal parts black soil and perlite. The survival and rooting percentages, number and lengths of roots produced and number and surface areas of new leaves were assessed at 4-weekly intervals until week 16. **Results:** Results indicated that the cuttings treated with single auxin treatments (mainly 0.2% NAA and 0.1% IAA) had higher survival and rooting percentages, longer roots, larger leaf surface areas and produced more leaves than those cuttings treated with commercial rooting hormones. **Conclusion:** It is concluded that compared to commercial rooting hormones, 0.2% NAA and 0.1% IAA were the most effective auxin hormones in promoting survival and growth of *Dillenia suffruticosa* cuttings.

Key words: Bornean heath forests, IAA, IBA, NAA, rehabilitation, rooting hormones, vegetative propagation, auxins

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Corresponding Author: F. Metali, Environmental and Life Sciences Programme, Faculty of Science, Universiti Brunei Darussalam, Jalan Tungku Link, BE1410, Brunei Darussalam Tel: +6732463001

Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Heath and Peat Swamp Forests of Brunei Darussalam, which cover 3,500 and 90,000 ha of the country's total land area, respectively are unique in terms of their biodiversity and ecosystem features[1]. In the Belait District of Brunei Darussalam, two types of tropical heath forests exist: Kerangas (on higher and drier areas) and kerapah (on permanently waterlogged and lower areas)[2,3]. These forests are intermixed with the Peat Swamp Forests at Lumut and Badas and some of them occur along the Brunei Liquefied Natural Gas Company (BLNG) water pipelines that carry water from the Belait River to the BLNG plant in Lumut (pers. obs). In 2013, a 20 m wide fire corridor was created along the BLNG Pipeline Road as a preventive measure to reduce fire threats to the water pipelines (BLNG, pers. com.). However, the removal of vegetation cover has conversely increased the risk of fires to the dry and exposed peat within the fire corridor. As such, there is currently an urgent need to revegetate this fire corridor especially with increasing frequencies of dry spells and droughts that have increased fire incidences.

Cleared and degraded forests created from anthropogenic disturbances take a long time to recover naturally, thus supplementing their regeneration through rehabilitation and reforestation activities using tree planting methods are necessary[4]. Wildings and stem cuttings of native plant species have been widely used to revegetate degraded lands and forests[5,6]. A rapidly growing number of rooted stem cuttings of plants has been vegetatively propagated and used in many reforestation programmes[7,8].

An investigation on the vegetative propagation of *Dillenia suffruticosa* (Griff.) Martelli, an abundant shrub that is native to Brunei Darussalam[9] was conducted. *Dillenia suffruticosa* from the family Dilleniaceae was chosen because it is a potential candidate for rehabilitation of degraded lands due to its ability to grow in various habitats including roadsides, secondary forests, open sites in undisturbed forest, peat swamps, heath forests, white sands and also well drained and eroded soils[10]. *Dillenia suffruticosa* can be used to stabilise slopes in preventing landslides[11]. Since obtaining *D. suffruticosa* seedlings via seed propagation is difficult and almost impossible[12], using auxins: Naphthalene Acetic Acid (NAA) and Indole Acetic Acid (IAA) in stem cuttings seem to be the best option to produce plantlets[9].

This study investigated the different hormones (either using single auxin or commercially available plant rooting hormones) that could increase the survival and rooting percentages as well as root and leaf production of *D. suffruticosa* stem cuttings. Therefore, the main aim of the study was to determine which hormone could be used for the mass propagation of *D. suffruticosa* and thus, help to rapidly supply plantlets for the revegetation of degraded heath forests in Brunei Darussalam.

MATERIALS AND METHODS

Sampling of stem cuttings: A total of 150 softwood stem cuttings of *Dillenia suffruticosa* (Griff.) Martelli were collected from the heath forests within the Universiti Brunei Darussalam campus (4°58'41.5"N, 114°53'39.8"E) and along the Muara-Tutong highway (4°58'59.81"N, 114°54'20.34"E). A maximum of two leafy softwood stem cuttings (8-12 cm) were sampled per plant to reduce any damage to the wild stock plants.

Preparation of stem cuttings: Similar to Abidin and Metali[9], all organs on the stem cuttings were removed to ensure they concentrate their energy in forming root and shoot instead of producing reproductive organs. A replicate of 10 cuttings were treated with single auxin hormones, 0.2% Naphthalene Acetic Acid (NAA), 0.1% Indole Acetic Acid (IAA) and 0.1% Indole Butyric Acid (IBA) and 20 cuttings with commercial rooting hormone powders, Seradix 1, 2, 3 and Yates and rooting hormone gel, Clonex, using a quick-dip method[9,13]. The auxins and their respective concentrations were selected and prepared by following Abidin and Metali[9], whereas the commercial hormones were purchased from the local markets. The active constituent(s) of Seradix (unknown concentrations) and Clonex (0.3%) was IBA and Yates were 0.005% IAA and 0.002% NAA. Distilled water was used as a control treatment.

All stem cuttings were planted into square plastic pots containing autoclaved growth media of equal parts black soil and perlite. The pots were placed on the benches in a plant house with misted conditions and the benches with pots were covered with a PVC plastic cover to maintain high relative humidity. The temperatures, relative humidity and light intensities recorded in the plant house were between 22-29°C, 80-90% and 300 μmoL m^{-2} sec^{-1} (Apogee Quantum MQ-200 PAR Meter, Apogee Instruments, USA), respectively. After 6 weeks, the cuttings were re-potted into large pots with only black soils. The pots were subsequently transferred to a plant shade with temperatures between 22-34°C, relative humidity of 60-75% and light intensities of 450 μmol m^{-2} sec^{-1}. Fertilizers (NPK, 15:15:15) were added into the potted soil every 4 weeks and watered twice a day.

Assessment of cuttings and statistical analysis: The cuttings were assessed every 4 weeks until the 16th week for their percentages of survival and rooting, number and lengths of new roots produced and number and surface areas of new leaves. All statistical analyses were conducted using R version 3.3.0[14]. Differences in the number of roots, root lengths, number of leaves and leaf surface areas between the different hormones were determined using one-way ANOVA and Tukey's HSD tests. Assumptions of normality and equal variances were checked during one-way ANOVA and were not violated.

RESULTS

Stem cuttings treated with single auxin treatments (i.e., 0.2% NAA, 0.1% IAA and 0.1% IBA) had 100% survival recorded throughout the 1st 12 weeks (Table 1). However, by week 16, only stem cuttings treated with 0.2% NAA remained at 100% survival. Stem cuttings treated with auxins produced roots faster with 100% rooting percentage achieved by week 4 when treated with 0.2% NAA and 100% rooting achieved by week 8 when treated with 0.1% IAA (Table 1). Cuttings treated with 0.1% IBA produced roots more slowly compared to the other hormone treatments, reaching only 90% rooting by week 16. In contrast, percentage survival for all stem cuttings treated with commercial rooting hormones fell considerably to 40% and below by week 4, except for cuttings treated with Yates (Table 1). Percentage survival for all commercial rooting hormones reached 0% survival by week 12, except Yates-treated stems. None of the cuttings treated with commercial hormones (except Yates) had successfully rooted (Table 1).

Cuttings treated with 0.2% NAA recorded significantly higher production of roots (Fig. 1a) and significantly longer root lengths (9.34±1.42 cm, Fig. 1b) than the other plant hormones. In contrast, cuttings treated with 0.1% IAA had significantly higher number of new leaves produced (10 new leaves, Fig. 1c) and significantly larger leaf surface area (166±48.6 cm^2, Fig. 1d). Cuttings treated with distilled water (control) and all commercial rooting hormones (except for Yates) did not successfully produce any roots and leaf, thus no data for these treatments were recorded by week 16.

DISCUSSION

Stem cuttings survived and rooted better when they were treated with single auxin treatments than the control and commercial rooting hormone treatments. The present findings on survival and rooting percentages are consistent with a previous study by Abidin and Metali[9], in which both studies reported that cuttings of *D. suffruticosa* treated with NAA and IAA produced more and longer roots, more new leaves and greater leaf areas, respectively than any other treatments. Proper root formation is important for survival and growth of cuttings[9] and well-formed root systems allow plants to grow and compete for resources, like nutrients and water[15]. In this study, NAA (0.2%) and IAA (0.1%) showed significant effects on root and leaf productions, thus contributed to higher survival and rooting percentages of the cuttings. Similarly, Kebede *et al.*[16] also reported very high survival (98-100%) of stem cuttings of two native tree species treated with NAA at concentrations of 0.2 and 0.4%. In addition, Usman and Akinyele[17] recorded the highest number of roots and root length production of cuttings of a tree species treated with NAA. Hossain and Urbi[18] reported that, other than NAA, IAA was also known to be very effective in promoting growth and root length production in stem cuttings of an herb.

Table 1: Survival percentages and rooting percentages of *Dillenia suffruticosa* stem cuttings treated with different hormones (0.2% Naphthalene Acetic Acid (NAA), 0.1% Indole Acetic Acid (IAA), 0.1% Indole Butyric Acid (IBA), Yates, Clonex, Seradix 1, 2 and 3) and distilled water as the control treatment at an interval of 4 weeks until week 16

	Survival percentage (%)					Rooting percentage (%)				
	Week					Week				
Hormones	1	4	8	12	16	1	4	8	12	16
0.2% NAA	100	100	100	100	100	0	100	100	100	100
0.1% IAA	100	100	100	100	90	0	60	100	100	90
0.1% IBA	100	100	100	100	90	0	20	30	60	90
Yates	100	100	100	75	35	0	20	80	75	35
Clonex	100	40	10	0	0	0	0	0	0	0
Seradix 1	100	40	15	0	0	0	0	15	0	0
Seradix 2	100	10	5	0	0	0	0	5	0	0
Seradix 3	100	10	0	0	0	0	0	0	0	0
Control	100	0	0	0	0	0	0	0	0	0

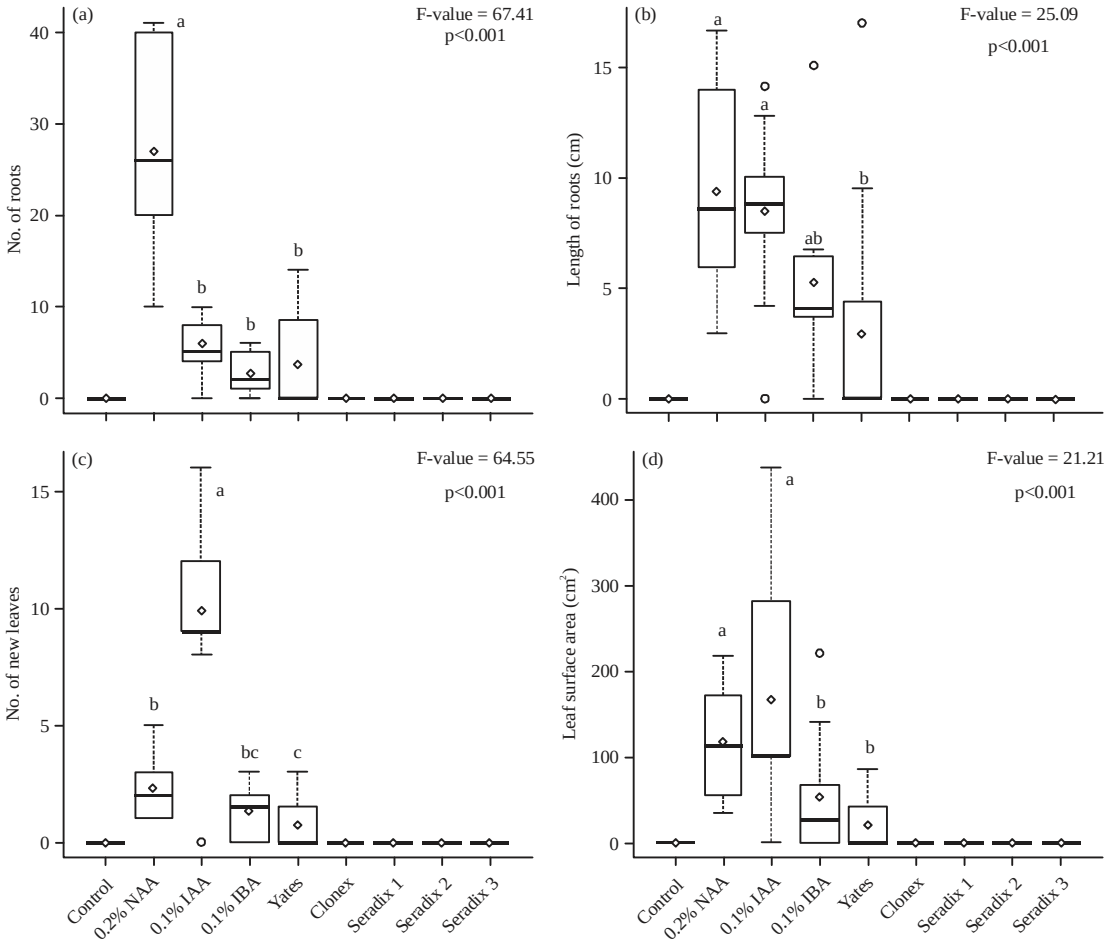

Fig. 1(a-d): Variation in the (a) Number of roots produced, (b) Length of roots, (c) Number of new leaves produced and (d) Surface area of leaves of *Dillenia suffruticosa* stem cuttings treated with plant hormones (0.2% NAA, 0.1% IAA, 0.1% IBA, Yates, Clonex and Seradix 1, 2 and 3) and distilled water as the control treatment. Mean value is denoted by the diamond-shaped symbol. Different letters indicate significant difference between treatments (p<0.001). *Dillenia suffruticosa* stem cuttings treated with Clonex, Seradix and Control had zero mean values for all 4 parameters. Only data at week 16 were presented

Higher percentage survival of cuttings treated with Yates compared to the other commercial rooting hormones used in this study could be due to the presence of both NAA and IAA as the active ingredients in Yates. However, due to its very low concentrations (0.005% IAA and 0.002% NAA), stem cuttings treated with Yates recorded lower rooting and survival percentages in week 16 and the effect was not clear-cut as those of single NAA and IAA hormone treatment. Kebede *et al.*[16] also reported that at low concentrations of NAA and IBA hormones, root productions were also low. Various studies have reported that different concentrations of hormones differentially affected survival, rooting percentage, leaf and roots production of cuttings[17-20]. Copes and Mandel[21] noted that a combination of NAA and IBA did not significantly increase root and leaf productions compared to the significant effects achieved with one hormone treatment, NAA or IBA, which was consistent with the present findings.

In this study, stem cuttings treated with Clonex rooting hormone gel which contained 0.3% IBA had negative results on survival, root and leaf production compared to 0.1% synthetic IBA treatment. This finding was similar to studies of stem cuttings propagation of three tropical species, whereby cuttings treated with commercial rooting hormone Clonex also showed lower rooting percentage compared to cuttings treated with synthetic IBA[22] and this could possibly be due to high IBA concentrations in Clonex.

The use of single auxins (especially 0.2% NAA) will result in a high survival rate of stem cuttings. The stem cutting

technique with plant hormone will produce plantlets that will be used in the revegetation of tropical degraded heath forests in Brunei Darussalam. Raising plantlets from vegetative propagation using plant hormones then consecutively using them to revegetate forests are not new[7,8]. There is a possibility that the revegetation programme in Lumut using *D. suffruticosa* will also be extended to the Peat Swamp forests as this species is well known to be grown in many forest habitats[10].

It is concluded that *Dillenia suffruticosa* stem cuttings grow better when treated with single auxin treatment than using commercial root-promoting hormones. Synthetic auxins, in particular 0.2% NAA and 0.1% IAA are more suitable for use in the mass propagation of *D. suffruticosa* stem cuttings. *D. suffruticosa* plantlets can be rapidly mass produced using stem cutting techniques with a single auxin treatment (mainly NAA and IAA), which can then be transplanted to the degraded heath forests in Lumut. The suitability of this species in the revegetation and rehabilitation programme will be further assessed via field monitoring of planted stem cuttings.

SIGNIFICANCE STATEMENTS

- The effects of auxins on growth of *Dillenia suffruticosa* cuttings were investigated for 16 weeks
- This study compared the effects of single auxin and commercial root-promoting hormone treatments on survival and rooting percentages and leaf and root production of cuttings
- A higher survival and rooting percentages and significantly longer root and greater number of roots were reported in NAA-treated cuttings than the other hormone treatments and water
- Based on the number and surface area of new leaves, IAA performed significantly better than other hormones
- It is concluded that compared to the commercial root-promoting hormones, 0.2% NAA and 0.1% IAA were the most effective auxins in propagating *Dillenia suffruticosa* cuttings
- The raised plantlets will be used in revegetation of degraded heath forests in Brunei Darussalam

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REFERENCES

1. Forestry Department, 2011. Sustainable forestry in Brunei Darussalam. Ministry of Industry and Primary Resources, Brunei Darussalam.
2. Wong, K.M. and A.S. Kamariah, 1999. Forests and trees of Brunei Darussalam. Universiti Brunei Darussalam, Forestry Department and Brunei Shell Petroleum Co. Sdn Bhd, Brunei Darussalam.
3. Din, H., F. Metali and R.S. Sukri, 2015. Tree diversity and community composition of the Tutong white sands, Brunei Darussalam: A rare tropical heath forest ecosystem. Int. J. Ecol. 10.1155/2015/807876.
4. Harrison, R.D. and T. Swinfield, 2015. Restoration of logged humid tropical forests: An experimental programme at Harapan Rainforest, Indonesia. Trop. Conserv. Sci., 8: 4-16.
5. Sakai, C., A. Subiakto, I. Heriansyah and H.S. Nuroniah, 2001. Rehabilitation of Degraded Forest with *Shorea Leprosula* and *S. Selanica* Cuttings. CIFOR, Bogor, Indonesia, ISBN: 9789798764707, Pages: 226.
6. Brundu, G. and D.M. Richardson, 2016. Planted forests and invasive alien trees in Europe: A Code for managing existing and future plantings to mitigate the risk of negative impacts from invasions. NeoBiota, 30: 5-47.
7. Gage, E.A. and D.J. Cooper, 2004. Controls on willow cutting survival in a montane riparian area. J. Range Manage., 57: 597-600.
8. Ritchie, G.A., 1991. The commercial use of conifer rooted cuttings in forestry: A world overview. New For., 5: 247-275.
9. Abidin, N. and F. Metali, 2015. Effects of different types and concentrations of auxins on juvenile stem cuttings for propagation of potential medicinal *Dillenia suffruticosa* (Griff. Ex Hook. F. and Thomson) Martelli shrub. Res. J. Bot., 10: 73-87.
10. Ashton, P.S., A.S. Kamariah and I.M. Said, 2003. A Field Guide to the Forest Trees of Brunei Darussalam and the Northwest Borneo Hotspot. University of Brunei Darussalam, Brunei Darussalam, ISBN: 9789991710921, Pages: 281.
11. Prasad, A., S. Kazemian, B. Kalantari, B.B.K. Huat and S. Mafian, 2012. Stability of tropical residual soil slope reinforced by live pole: Experimental and numerical investigations. Arabian J. Sci. Eng., 37: 601-618.
12. Manjul, N.M.J. and F. Metali, 2016. Germination and growth of selected tropical pioneers in Brunei Darussalam: Effects of temperature and seed size. Res. J. Seed Sci., 9: 48-53.

13. Awang Kamis, N.D.Z., H. Taha and F. Metali, 2016. Effects of commercial plant hormones on the survival, rooting and growth of stem cuttings of an herbal tea plant, *Aidia racemosa*. Res. J. Med. Plants, 10: 414-419.
14. R Development Core Team, 2013. R: A language and environment for statistical computing. The R Foundation for Statistical Computing, Austria, Vienna.
15. Yan, Y.H., J.L. Li, X.Q. Zhang, W.Y. Yang and Y. Wan *et al*., 2014. Effect of naphthalene acetic acid on adventitious root development and associated physiological changes in stem cutting of *Hemarthria compressa*. PloS One, Vol. 9. 10.1371/journal.pone.0090700
16. Kebede, M., H. Hulten and G. Balcha, 2013. Vegetative propagation of juvenile leafy stem cuttings of *Prunus africana* (Hook.f.) Kalkm and *Syzygium guineense* (Willd.) DC. Int. J. Bot., 9: 30-36.
17. Usman, I.A. and A.O. Akinyele, 2015. Effects of growth media and hormones on the sprouting and rooting ability of *Massularia acuminata* (G. Don) Bullock ex Hoyl. J. Res. For. Wildlife Environ., 7: 137-146.
18. Hossain, M.S. and Z. Urbi, 2016. Effect of naphthalene acetic acid on the adventitious rooting in shoot cuttings of *Andrographis paniculata* (Burm. f.) Wall. ex Nees: An important therapeutical herb. Int. J. Agron., Vol. 2016. 10.1155/2016/1617543
19. Aminah, H., J.M. Dick, R.R.B. Leakey, J. Grace and R.I. Smith, 1995. Effect of Indole Butyric Acid (IBA) on stem cuttings of *Shorea leprosula*. For. Ecol. Manage., 72: 199-206.
20. Memon, N., N. Ali, M.A. Balochi and Q. Chartar, 2013. Influence of Naphthalene Acetic Acid (NAA) on sprouting and rooting potential of stem cuttings of Bougainvillea. Sci. Int., 25: 299-304.
21. Copes, D.L. and N. Mandel, 2000. Effects of IBA and NAA treatments on rooting Douglas-fir stem cuttings. New For., 20: 249-257.
22. Cullina, W., 2002. Native Trees, Shrubs and Vines: A Guide to Using, Growing and Propagating North American Woody Plants. Houghton Mifflin Harcourt, Boston, ISBN: 9780618098583, Pages: 354.