# VEGETATIVE PROPAGATION OF *ARUNDINA GRAMINIFOLIA*: INFLUENCES OF NODE NUMBERS, PHYSIOLOGICAL AGE AND POSITION OF CUTTING FOR BEST SHOOT-BUD PROLIFERATION

Sakinah Idris
*School of Biosciences and Biotechnology, Faculty of Science and Technology,*
*Universiti Kebangsaan Malaysia, 43600 Bangi, Selangor, Malaysia.*
*Email: kinnsbi2@gmail.com*

Ab. Kahar Sandrang
*Horticulture Research Centre, Malaysia Agriculture Research and Development Institute,*
*43400 Serdang, Selangor Malaysia.*
*Email: skahar@mardi.gov.my*

Che Radziah Che Mohd. Zain
*School of Biosciences and Biotechnology, Faculty of Science and Technology,*
*Universiti Kebangsaan Malaysia, 43600 Bangi, Selangor, Malaysia.*
*Email: cradziah@ukm.edu.my*

**ABSTRACT**

*A study to determine the best vegetative propagation was carried out for Arundina graminifolia. Commonly, A. graminifolia was propagated through techniques such as seed, a division of root mass or aerial plantlet (keikis). The rate propagation by these techniques is very limited and slow to produce more planting material. Therefore, this research aims to establish an efficient technique to propagate of A. graminifolia in a short time. Studies investigated on the selection of cutting on node numbers (single node, two nodes and three nodes), types of physiological age (young, moderate and old explant) and type of position of stem cuttings (upper, middle and lower part) were carried out. All these variables were shown to be important factors affecting for shooting, confirming the hypothesis that successful shooting can be achieved if these primary variables are optimized. Influence of node number on average of shoot growth (shoot width, shoot height, shoot gap and number of leaves) were significantly increase ($p<0.05$) as the more node was used. Three nodes cutting gave highest shoot growth (2.76mm, 20.45mm, 6.00mm and 1.75), followed by two nodes cutting (2.75mm, 14.39mm, 4.90mm and 1.5) and single node cutting (2.40mm, 8.55mm, 3.00mm and 1.0). A greater average of shoot growth were generally highest and lowest in young physiological age and old physiological age, respectively. While, the best cutting position on shoot height is on upper (15.69mm), middle (15.12mm), lower (11.95mm) cutting from young physiological age and similar on upper (15.67mm) cutting from the moderate physiological age. It was concluded that using three nodes cuttings should be taken from young and moderate physiological age with middle and upper position of stem is the best cutting for shoot proliferation of A. graminifolia in the short time.*

Key words: Arundina graminifolia, cutting position, number of node, physiological age, shoot-bud proliferation

## INTRODUCTION

*Arundina graminifolia* is commonly known as bamboo orchid. Bamboo orchid is a terrestrial perennial orchid, easy to maintain, heat-resistant, resistant to disease and flowering throughout the year and it is suitable for use in outdoor landscapes. It is widely distributed in Southeast Asia, from the Himalayas to western Indonesia (Hong et al., 1983). Currently, Malaysian Agriculture Research and Development Institute (MARDI) had produced a new hybrid of *A. graminifolia* which has good characteristic and at the same time has the potential to be commercialised (Wan Rozita, W.E. and Rozlaily, Z. 2015). The new hybrid of *A. graminifolia* has a modest height with a more attractive, unique flower colour and shape (Wan Rozita, W.E. and Rozlaily, Z. 2015). Thus *A. graminifolia* has received high demand from local authority like Kuala Lumpur City Hall (DBKL) and housing developers like SP Setia Company as a landscape plant. Therefore, a study on mass propagation of the new hybrid of *A. graminifolia* should be carried out.

Commonly, *A.graminifolia* is propagated through seed, division of the root mass or aerial plantlet and *in vitro* culture technique. Propagation through seeds produced the not true-to-type plant. Propagation of *Arundina* sp. has been reported through seed culture (Bhadra et al., 2005; Chen et al., 2006), while propagation through division of the root mass or aerial plantlet are very limited and requires a longer time. Meanwhile, *In vitro* culture technique have been done by many researcher (Das et al., 2013; Martin, K.P. 2007; Chen et al., 2006; Bhadra et al., 2005) but take a long growth period and require an effective protocol. According to Das et al (2013), only 50% explants showed positive response and took 45-47 days for bud proliferation. Martin, K.P. (2007) has also established micro propagation of Arundina through PLBs using node explant and 89% conversion of PLBs to shoot at two months. However, no researches have been tried through selection of stem cutting via conventional propagation on *A. graminifolia*.

Selection of stem cuttings on *A. graminifolia* can come in many form likes numbers of node (single node, two nodes and three nodes), types of physiological age (young, moderate and old explant) and type of position of stem cuttings (upper, middle and lower part). All these variables is important factors affecting for shooting of *A. graminifolia.* Therefore, this present study aimed

to evaluate the shoot-bud proliferation of *A. graminifolia* by selected explant which is node numbers, physiological age and position of the cutting using open permanent immerse system (OPIS).

## MATERIALS AND METHODS

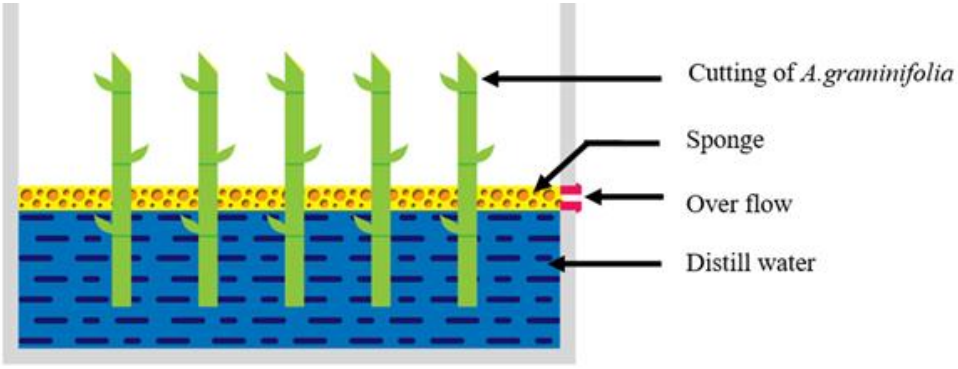
### Planting materials

*Arundina graminifolia* was obtained from Malaysia Agriculture Research and Development Institute (MARDI), Serdang. Single node, two nodes and three nodes cutting were used as explants in first experiment and three nodes cutting about 10-12cm long were used as explants in second experiment with different physiological age wich is young (before flowering) , moderate (flowering) and old (after flowering) as explants with difference type of position of cutting (upper, middle and lower part). Cutting positions were assigned starting at node 4 from the apex of the shoots as upper, followed by middle and lower part positions sequentially down the shoots for all physiological age explants. All leaves on the cuttings were removed, soaked with twenty-20 solution to disinfect the germs in the explants and washed in running tap water for 20 min before planting. These cutting were then inserting in a propagator system namely open permanent immers system (OPIS) in the nursery with 90% level of shading at MARDI, Serdang, Selangor, Malaysia.

### Preparation of open permanent immers system (OPIS)

The OPIS system which made from the polystyrene and the diagram of this system showed such as bellow.

**Figure 1. Open permanent immerse system (OPIS)**




### Statistical analysis

The design of the experiment was performed in Randomized Complete Block Design (RCBD) with 4 replications and each replicate consist of thirty cuttings. The experiments were repeated twice. Data on shoot width, shoot height, shoot gap and number of leaves were taken every week after planting. Data was measured using digital caliper (Mitutoyo 0.01mm). Data was analyzed by analysis of variance (ANOVA) using SAS software version 9.3 and tested for significance using Least Significant Difference (LSD) at $P \leq 0.05$.

## RESULT AND DISCUSSION

*Exsperimen 1 - Influence of node number*

Generally cuttings of all treatments able to produce shoot-bud proliferation after 4 weeks insertion respectively. Average of the shoot growth such as shoot width, shoot height, shoot gap and number of leaves were significantly increase ($p<0.05$) as the more node was used shown in table 1. Three nodes cutting gave highest shoot growth (2.76mm, 20.45mm, 6.00mm and 1.75), followed by two nodes cutting (2.75mm, 14.39mm, 4.90mm and 1.5) and single node cutting (2.40mm, 8.55mm, 3.00mm and 1.0). All treatment is without leaf, so they are only depend on availability of carbohydrates and nutrients store within the stem tissues to induce shoots. This finding clearly showed that more node number have more capacity of carbohydrates and nutrients in stem cutting, thus can stimulates the growth of shoot-buds easily. The finding is similar with the study in the shooting process in *Larix hybrid* (Pellicer et al., 2000) and in *Khaya ivorensis* (Tchoundjeu and Leakey, 2000) which is the cutting length or more node number has a major influence on shooting ability.

**Table 1: Shoot growth with difference node numbers of *A. graminifolia* at 4 weeks**

| Node numbers | Single node | 2 nodes | 3 nodes |
|---|---|---|---|
| Shoot width | 2.40 $^{b}$ | 2.75 $^{a}$ | 2.76 $^{a}$ |
| Shoot height | 8.55 $^{c}$ | 14.39 $^{b}$ | 20.45 $^{a}$ |
| Shoot gap | 3.00 $^{c}$ | 4.90 $^{b}$ | 6.00 $^{a}$ |
| Number of leave | 1.0 $^{b}$ | 1.5 $^{ab}$ | 1.75 $^{a}$ |

Means with the same letters within the row do not differ significantly according to LSD p < 0.05

**Figure 1: Shoot growth with difference node numbers of *A. graminifolia* at four weeks.**


**(A) Single node cutting, (B) Two nodes cutting and (C) Three nodes cutting.**

*Exsperimen 2 - Influence of physiological age* x *position of cutting*

All results on influence of type of physiological age x position of cutting on shoots-bud growth were shown a significant difference ($p<0.05$) as summarized in Table 2. For shoot width, cutting from young physiological age with middle part (2.72mm) and from moderate physiological age with upper part (2.73mm) of plant produced greater shoot width compare to old physiological age but slightly same with young physiological age with upper part (2.56mm), moderate physiological age with middle part (2.48mm) and old physiological age with middle part (2.48mm).

Base on the results of shoot height, cutting from young physiological age with upper, middle part (15.69mm, 15.12mm) and from moderate physiological age with upper part (15.67mm) of plant produced greater shoot height compare to old physiological age but slightly same with young physiological age with lower part (11.95mm) and old physiological age with middle part (10.53mm).

Meanwhile the maximum shoot gap is highest in young physiological age with upper and middle part (5.62mm and 5.73mm) compare to other. For number of leave showed cutting from young physiological age with middle part (3.75) and from moderate physiological age with upper part (3.75) produced greater number of leave compare to others.

A greater average of shoot growth were generally highest and lowest in young physiological age and old physiological age, respectively. While, the best cutting position is on upper, middle, lower cutting from young physiological age and similar on upper cutting from the moderate physiological age. It is same result from Leakey (1983), there are considerable differences in shooting ability between the physiological age and cutting positions, with those from upper shoots being best. Stem cuttings taken from juvenile donor plants are generally considered easy to propagate by cuttings due to their young ontogenetic, physiological and chronological age and possibly to its low production of secondary metabolites (Husen and Pal 2006).

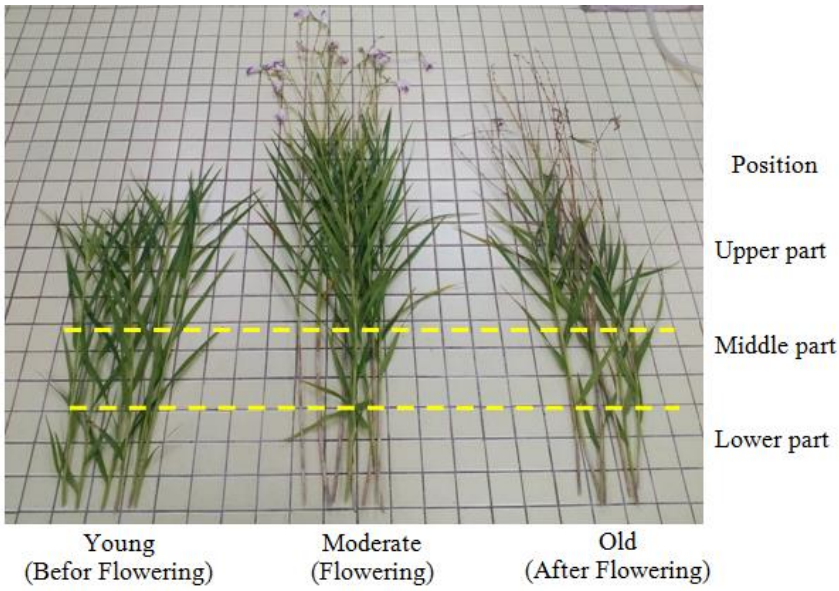
All cutting used in this experiment showed a good finding to improve shoot-bud proliferation within 30 days compare to Das et al. (2013) and Martin (2007) were propagated *A. graminifolia* through *in vitro*. According to Das, they used single node explant to the Murashige & Skoog media (MS media) and took 45-47 days to bud break. Meanwhile Martin took 60 days to conversion of protocorm-like bodies (PLBs) to protocorm and took some more time to growth as a plant.

**Table 2: Shoot growth with difference physiological age and cutting position on eight week**

| Explant | Position | Shoot width | Shoot height | Shoot gap | Number of leave |
|---|---|---|---|---|---|
| Young | Upper | 2.56ab | 15.69a | 5.62ab | 2.75abc |
| (Befour flowering) | Middle | 2.72a | 15.12a | 5.73ab | 3.75a |
| | Lower | 2.39cb | 11.95ab | 3.77bcd | 2.5abc |
| Moderate | Upper | 2.73a | 15.67a | 4.43bc | 3.75a |
| (Flowering) | Middle | 2.48abc | 7.17cd | 4.22bc | 1.75abc |
| | Lower | 2.18cd | 4.96de | 0.37e | 1.0c |
| Old | Upper | 2.03d | 6.70cde | 2.71cd | 1.5bc |
| (After flowering) | Middle | 2.48abc | 10.53bc | 4.42bc | 2.0abc |
| | Lower | 1.38e | 2.51e | 1.84de | 1.0c |
| LSD | | 0.32 | 4.59 | 2.15 | 2.03 |

Means with the same letters within the column do not differ significantly according to LSD p < 0.05

**Figure 2: Explants from difference physiological age (young, moderate and old) and position of the cutting (upper, middle and lower part)**




## CONCLUSION

In conclusion, all these variables were shown to be important factors affecting for shooting, confirming the hypothesis that successful shooting can be achieved if these primary variables are optimized. It was concluded that using three nodes cuttings should be taken from young and moderate physiological age with middle and upper position of stem is the best cutting for shoot proliferation of *A. graminifolia* in the short time.

## REFERENCES

Bhadra, S.K. and Bhowmik, T.K. (2005). Axenic germination of seeds and rhizome-based micropropagation of an orchid *A. graminifolia* (D. Don.) Hochr. *Bangladesh Journal of Botany*, 34, 59-64

Chen, Z.l., Zeng, S.J., Wen, T.l., Duan, J. (2006). Asepsis sowing and *in vitro* propagation of *Arundina graminifolia* D. Don. Hochr. *Plant Physiology Communications*, 42, 66.

Das, S., Choudhury, M.D. and Mazumder, P.B. (2013). *In vitro* propagation of *A. graminifolia* D. Don. Hochir- A bamboo orchid. *Asian Journal of Pharmaceutical and Clinical Research*, 6, 156-158

Hong, D.Y., Lian, Y.S. and Shen, L.D. Orchidaceae. (1983). In Flora of China; Chinese Science Press: Beijing, China Volume 73, p. 320

Hartmann, H.T & Kester, D.E. (1983). Plant propagation: principles and practices. 4th ed. Englewood, New Jersey, USA; Prentice Hall. 727 pp. ISBN 978–0–13–0–13

Husen A, P.M. (2006). Variation in shoot anatomy and rooting behaviour of stem cutting in relation to age of donor plants in teak (Tectona grandis Linn. F). New For 31:57–73

Leakey, R.R.B. (1983). Stockplant factor affecting root initiation in cuttings of *Triplochiton scleroxylon* K. Schum., an indigenous hardwood of West Africa. *Journal of Horticutureal Science*, 58, 277-290.

Martin, K.P. (2007). *In vitro* propagation of *A.graminifolia* through PLBs. *Propagation of Ornamental Plants*, 7(2), 97-100

Pellicer, V, Guehl, J-M, Daudet, F-A, Cazet, M, Riviere, LM and Maillard, P (2000). Carbon and nitrogen mobilization in Larix x eurolepis leafy stem cuttings assessed by dual 13C and 15N labelling: Relationships with rooting. *Tree Physiology* 20, 807-814.

Tchoundjeu, Z and Leakey, (2000). Vegetative propagation of Khaya ivorensis (*African mahogany*): effects of stockplant flushing cycle, auxin and leaf area on carbohydrate and nutrient dynamics of cuttings. *Journal of Tropical Forest Science* 12, 77-91.

Sakinah, I., Che Radziah, C.M.Z., Ab. Kahar, S. and Wan Rozita, W.E. (2018). Preliminary study of propagation system to induce shoot-bud proliferation of *Arundina graminifolia.* Poster paper at *28th Malaysian Society of Plant Physiology Conference*. pp. 71.

Wan Rozita, W. E. and Rozlaily, Z. (2015). Assesment of genetic variation in *Arundina raminifolia* D. Don. Hochir. using AFLP markers. *2nd International Congference on Crop Improvement*, hlm. 75.