



MODULATING EFFECT OF INDIVIDUAL AND CLUSTERED TREATMENTS OF MIRACLE FRUIT PLANT (*Synsepalum dulcificum*) STEM CUTTINGS WITH QUICK DIP ROOTING HORMONE

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Abstract

Hormone application has been more of individual application. To ascertain the best application technique, *Synsepalum dulcificum* cuttings were used for this study. The experiment was a 2 x 5 factorial treatment laid out in a completely randomized design. Two quick-dip methods (cluster and individual) as one factor, and Indole-3-butyric acid (IBA 200 mg/l and 250 mg/l), and Naphthalene acetic acid (NAA 200 mg/l and 250mg/l) as the other factor. Distilled water was used as control, and cuttings assessed two months after planting. Data collected were statistically analysed using two factor ANOVA procedure. Treatment means were separated using Least Significant Difference procedure at 5% probability level. Results show that IBA 200mg/l had a better effect on number of roots per cutting (2.38), and roots length (4.4), followed by NAA 200mg/l (number of roots per cutting as 2.75, while roots length was 1.42cm). Although NAA 200mg/l cuttings had a higher number of roots, but IBA 200mg/l treated cuttings will perform better considering their root lengths. Also, clustered treated cuttings recorded appreciable increase in number of roots, new leaves and roots length than individual treated cuttings. Cluster treatment is thus, recommended for the much needed vegetative propagation of *S. dulcificum* and probably for other species.

Keywords: Stem cuttings, quick deep solution, cluster application, and individual application

Introduction

The growth and development of a plant are influenced by genetic and external environmental factors, and chemical hormones inside the plant (Baca and Elmerich, 2003). Hormones are organic substances, produced endogenously by plants and capable of regulating plant growth at very low concentration. They alter root architecture and promote plant development, thus playing a significant role in increasing the root surface area, and number of root tips in many plants (Bhattacharyya and Jha, 2012). Enhancement of adventitious root formation through vegetative propagation methods such as stem cutting using synthetic hormone have been a common practice, and have been known to produce good results in the nursery. However, hormones are to be handled with care. Over-application of some hormone formulations can cause damage(s) to the cuttings; auxin in excessive concentrations may result in inhibition of bud development, yellowing of leaves, leaf abscission, blackening of stems and even death of cuttings. It has also been reported that misapplication to leaves may result in curling, or other distortion of plant growth.

Formulations dissolved in alcohol are more prone to cause burn, or dehydrated plant tissue (Cervený and Gibson, 2015). Different types of hormone formulation such as powdered forms, quick-dip solutions, water soluble formulations and post-planting sprays are found in the market. They are prepared and applied to cuttings differently. Five major groups of hormone are recognized by botanists: auxin, gibberellins, ethylene, cytokinins and abscisic acid (Baca and Elmerich, 2003).

However, in most of the natural vegetatively propagated species, adventitious root formation can occur without any need for pretreatment of cuttings with hormones, while some other crop species will require growth regulators for effective rooting (Robert, 2020). Kumari *et al.* (2010) recorded increase in rooting and sprouting of *Jatropha curcas* cuttings treated with IBA than untreated cuttings, which were also confirmed by Kochhar *et al.* (2008), but were contrary to the findings of Adekola and Akpan (2012) and Aminu-Islam *et al.* (2010) who reported the untreated cuttings of *J. curcas* to be better than hormone treated ones. The disparity in

the findings of these studies could possibly be as a result of the hormone application technique/method. Consequently, it became necessary to investigate the best method of applying quick-dip hormone solutions on cuttings (that is dipping of several cuttings at once, or dipping of cuttings individually). Two node stem cuttings of *Synsepalum dulcificum* (Daniel) also known as Miracle fruit plant, a slow-growing medicinal shrub that belongs to the family Sapotaceae was used in this study. Propagation of miracle fruit are believed to be restricted due to difficulties in rooting through cuttings, and the seeds are recalcitrant (Chen *et al.*, 2012). This study was aimed at determining the best method of applying quick deep rooting hormone on Miracle fruit plant cuttings

Materials and Methods

Study location

This study was conducted in the tree improvement nursery of Forestry Research Institute of Nigeria (FRIN), Ibadan, Oyo State. FRIN is located on latitude 7° 26' N and longitude 3° 54' E of the equator. The climate of the area is tropical, and dominated by rainfall pattern between 1300mm-1500mm. The average temperature is about 36°C, and relative humidity ranges between 80-85% (FRIN, 2019).

Experimental design

This study was a two-factor experiment laid out in completely randomized design (CRD), with four (4) hormone treatments: Indole-3-butyric acid (IBA 200 mg/l and 250 mg/l), and Naphthalene acetic acid (NAA 200 mg/l and NAA 250mg/l) as one factor, and two quick-dip methods (cluster and individual), as the other factor. The treatments were replicated four (4) times. Distilled water was used as control. The individual root dipping was carried out by dipping only one two-node stem cuttings of *S. dulcificum* at a time in the hormone solution, while the cluster method was carried out by dipping two-node stem cuttings of *S. dulcificum* cluster into a hormone solution (at the same time), followed by dipping another set in a fresh treatment of the same concentration, using quick dip method, according to Erturk *et al.* (2011). Four two-node stem cuttings made up a cluster. The hormone treated cuttings were then set in a germination tray filled with sterilized river sand with four cuttings placed in a tray, and this was replicated four times and placed under humid propagator according to Sumbele (2012) in a completely randomized design to develop new roots and shoots

Data collection

The following growth parameters were assessed after two months by counting: number of survived cuttings, number of rooted cuttings, number of callused cuttings, number of roots on cutting, and Length of roots using meter ruler.

Statistical analysis

Data collected were statistically analysed using two factor ANOVA procedure. Treatment means were separated using Fisher Least Significant Difference

(FLSD) procedure at 5% probability level.

Results and Discussion

Effect of the quick dip rooting hormone on individual and cluster treated cuttings of *S. dulcificum* (Table 1) shows significant difference on number of roots per cutting with NAA 200mg/l under cluster dipping with the highest number (4.50), and control (0.50). Significant difference was also detected in the number of new leaves, with NAA 250mg/l under cluster dipping with the most significant effect (4.50). The longest root length (7.65cm) was recorded under cluster dipping with IBA200mg/l. Effect of the two application method of the quick dip rooting hormone was further clarified on Fig 1, showing their effect on number of root per cutting, and number of new leaves and root length. This revealed that cluster treated cuttings enhanced the performance of the three growth parameters better than the individual dipping of cuttings. Effect of hormone at different concentrations on the cuttings of *S. dulcificum* (Fig 2) clearly shows that stem cuttings propagation were better enhanced, when treated with rooting hormone, than when propagated without hormone. This was also in agreement with the finding of Kumari *et al.* (2010), who recorded increase in rooting and sprouting of *Jatropha curcas* cuttings treated with IBA than untreated cuttings. Among the hormone treatments used, IBA 200mg/l has a better effect on the stem cuttings (number of root per cuttings was 2.38, while the root length was 4.4cm), followed by NAA 200mg/l (number root per cutting was 2.75, while its roots length was 1.42cm), while the control had the least (0.75) number of root per cutting, and 0.4cm for root length. Although, NAA 200mg/l had a higher number of root per cutting, but with longer root length recorded for IBA 200mg/l treated cuttings. The cuttings will perform better than other cuttings, because soil nutrient will easily be absorbed by the roots, and made available to the cuttings. This will also be of an advantage to the cuttings, when finally transplanted. Better performance of hormone treated cuttings in this study is a clear indication that exogenous hormone level has a role to play in the successful adventitious root formation, and development of cuttings. This has been found to exert the same effect on hormone treated stem cuttings of other species (Kumari *et al.*, 2010 and Kochhar *et al.*, 2005). The two different hormone application techniques (individual and cluster dipping) used in this study had no significant effect on the remaining growth parameters recorded at $p < 0.05$, but considering their mean value, cutting survival rate was better influenced by IBA 200mg/l (3.0), NAA 200mg/l (3.0) and NAA 250mg/l (3.0), than the control (1.00) when cuttings were treated in cluster. Number of root per cuttings were also better influenced when cuttings were treated in cluster with rooting hormone, NAA 200mg/l (4.50), followed by IBA 200mg/l (2.75), and control (0.50).

Considering the outcome of this study, cluster treatment of stem cuttings with quick dip rooting hormone will enhance stem cuttings better than individual treatment of cuttings. The appreciable result recorded against

cluster treatment could be as a result of the cuttings being exposed to the same hormone concentration at the same time. With individual treatments, hormone concentration decreases as the dipping progresses for cuttings soaked in water to avoid dehydration, exposing the cuttings to different concentrations. Thus, there will be variation in the modulating effect of the hormone on the cuttings, resulting to unappreciable overall growth of cuttings/plant. Also, for cuttings not soaked in water, period of hormone application for the whole experiment is shorter with cluster treatment than individual treatment of cuttings, thus causing evaporation of hormone solution, which will result to variation in the concentration of hormone applied on cuttings. According to Cerveny and Gibson, (2015), quick dip solution tends to increase in auxin concentration as the solution evaporates. IBA application has been confirmed to be the best rooting hormone on *S. dulcificum* for cell expansion and proliferation in cortex, which subsequently break fiber layers, and allowed the roots primordial to emerge (Xing *et al.*, 2012). This study clearly showed that stem cuttings response to quick dip rooting hormone is application technique (cluster or individual treatment) related. Table 1 shows that cuttings dipped individually in quick dip rooting hormone solution had a better growth performance, when compared with the control, but appreciable increase in growth parameter were recorded for cluster

treated cuttings. Cluster treatment of cuttings may probably have the same effect on other species stem cuttings for the much needed vegetative propagation of plants through stem cuttings for the production of early fruiting planting materials. Secondly, with data generated from this study, cluster application of IBA 200mg/l on stem cuttings of *S. dulcificum* is recommended to be the best for its propagation. Further study is thus recommended to be carried out on the method of application (cluster or individual treatment) with quick dip rooting hormone on stem cuttings for mass propagation and improvement of the overall performance of cuttings of other plants.

Conclusion

Enhancement of plant growth and yield using synthetic hormones (IBA and NAA) has been a common practice. They must not just be prepared appropriately, but also applied rightly in order to exert appreciable effect on plants. Cluster treatment is thus, recommended for the much needed vegetative propagation of *S. dulcificum*, and probably for other species.

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Table 1: Effect of quick deep hormone on individual and cluster treated cuttings of *S. dulcificum* cuttings 2 months after planting.

Quick deep method	Hormone Treatment	No of roots that survived	No of rooted cuttings	No of callused roots	No of roots per cutting	No of new Leaves	Root length (cm)
Cluster dip	CONTROL	1.00	1.00c	0.00	0.50de	1.00b	0.00c
	IBA 200	3.00	3.00a	0.00	2.75b	2.00b	7.65a
	IBA 250	2.50	2.00ab	0.50	1.00cde	1.50b	1.30bc
	NAA 200	2.00	1.50abc	0.50	4.50a	1.50b	2.30b
	NAA 250	3.00	2.50ab	0.50	1.50cd	4.50a	2.65b
Individual dip	CONTROL	1.00	1.00abc	0.00	1.00cde	2.00b	0.80bc
	IBA 200	2.50	2.00abc	0.50	2.00bc	1.00b	1.15bc
	IBA 250	1.00	1.00abc	0.00	1.50cd	0.00b	0.00c
	NAA 200	2.00	0.50bc	1.50	1.00cde	0.50b	0.55bc
	NAA 250	1.00	1.00abc	0.00	1.00cde	1.00b	1.10bc
LSD @ (p ≤ 0.05) quick dip method (QD)		1.136	0.891	0.546	0.459**	0.891*	0.906**
Hormone treatment (HT)		1.796	1.409	0.863	0.726**	1.409	1.432**
QD X HT		2.540	1.993*	1.220	1.027**	1.993	2.025**

Mean values with same alphabets are not significantly different from each other at p < 0.05

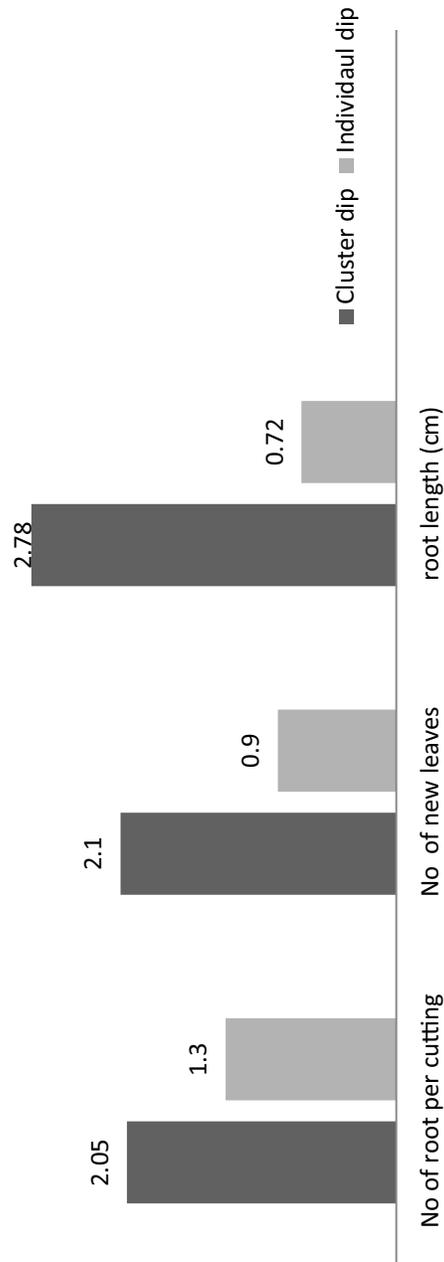


Fig. 1: Mean effect of cluster and individual quick dip method on number of root per cutting, number of new leaves and root length

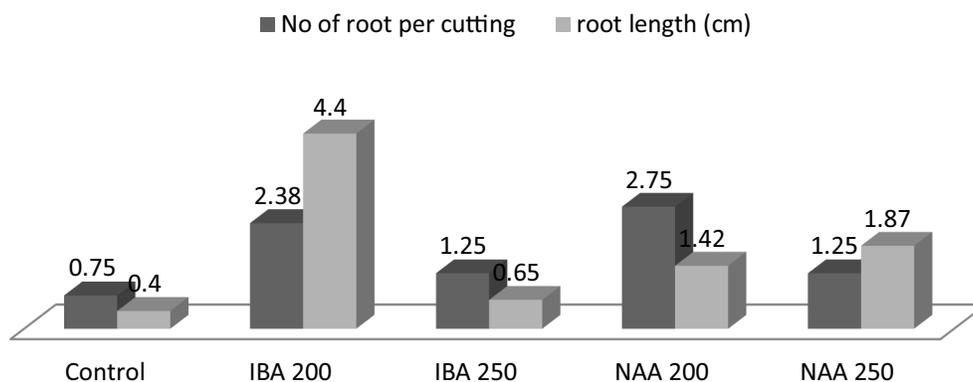


Fig. 2: Mean effect of Hormone treatments on number of root per cutting and root length

References

- Adekola, O. F. and Akpan, I. G. (2012). Effects of Growth Hormones on Sprouting and Rooting of *Jatropha curcas* l. Stem cuttings. *Journal of Applied Science, Environment and Management*, 16 (1): 153 – 156.
- Aminul-Islam, A. K. M., Zahira, Y., Nurina A. and Mohammed, O. (2010). Propagation Potentials of Genotypes and different Physiological Ages of Stem Cuttings in *Jatropha curcas* L. *Journal of Agricultural Science*, 2(4): 75-82.
- Bhattacharyya, P. N. and Jha, D.K. (2012). Plant Growth-Promoting Rhizobacteria (PGPR): Emergence in Agriculture. *World Journal of Microbiology Biotechnology*, 28: 1327–1350.
- Baca, B. E. and Elmerich, C. (2003). Microbial Production of Plant Hormones. In Elmerich, C and Newton, W.E. (eds.). *Associative and Endophytic Nitrogen-fixing Bacteria and Cyanobacterial Associations*, Kluwer Academic Publishers. Printed in the Netherlands.
- Cervený, C. and Gibson, J. (2015). Grower 101: Rooting Hormones. PROMIX <http://www.gpnmag.com/site/default/files/growe.pdf>. Retrieved on 20/01/2015.
- Chen, X. W., Abdullah, T. L., Abdullah, N. A. P. and Hassan, S. A. (2012). Rooting response of miracle fruit (*Synsepalum dulcificum*) softwood cuttings as affected by indole butyric acid. *American Journal of Agricultural and Biological Sciences*, 7(4): 442-446.
- Erturk, Y., Cakmakci, R., Duyar, O. and Turan, M. (2011). The Effect of Plant Growth Promoting Rhizobacteria on Vegetative Growth and Leaf Nutrient Contents of Hazelnut Seedlings (Turkish hazelnut cv, Tombul & Sivri). *International Journal of Soil Science* 6(3): 188-198.
- FRIN (2019). Forestry Research Institute of Nigeria. Meteorological Report 2018-19. Nigeria. Unpublished report
- Kochhar, S., Singh, S. R. and Kochhar, V. K. (2008). Effect of Auxins and Associated Biochemical Changes during Clonal Propagation of the Biofuel Plant-*Jatropha curcas*. *Biomass and Bioenergy*, 32: 1136-1143.
- Kumari, M., Patade, V. Y., Arif., M. and Ahmed, Z. (2010). Effect of IBA on Seed Germination, Sprouting and Rooting in Cuttings for Mass Propagation of *Jatropha curcas* L Strain DARL-2. *Research Journal of Agriculture and Biological Science*, 6(6): 691-696.
- Robert, P. (2020). Rooting hormone: Use them well. Garden fundamentals. <http://www.gardenfundamentals.com/use-rooting-hormone-correctly>. Retrieved 16/06/2020
- Sumbele, S. A. (2012). Effects of Auxins and Leaf Size on Rooting of *Treculia africana* (Decne) Stem Cuttings, Science. *Journal of Environmental Engineering Research*, Volume 2012, Article ID sjeer210, 5 Pages, 2012. doi: 10.7237/sjeer/210.
- Xing, W. C., Thohirah, L. A., Nur Ashikin, P. A. and Siti, A. H. (2012). Rooting Response of Miracle Fruit (*Synsepalum dulcificum*) Softwood Cuttings as affected by Indole Butyric Acid. *American Journal of Agriculture and Biological Sciences*, 7(4): 442-446