Performance of Coffee (*Coffea Conephora*) Shoot Cuttings in Response to Levels of Naphthalene Acetic Acid under Clonal Chamber Condition

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Abstract- Mass propagating robusta coffee through shoot cuttings using naphthalene acetic acid (NAA) is an innovative practice to hasten and mass produce planting materials of coffee for commercial production. A study was conducted to determine the performance of *Coffea conephora* in response to different levels of NAA using shoot cuttings under a rooting chamber condition with five treatments and four replications in a Completely Randomized Design (CRD). The different treatments were: T_0 (Control, 0 ppm NAA), T_1 (50 ppm NAA), T_2 (100 ppm NAA), T_3 (150 ppm NAA) and T_4 (200 ppm NAA). Robusta shoot cuttings dipped in 100 ppm NAA (T_2) and grown under clonal chamber performed best with the most number of leaves (2.38), most number of shoots (1.65), highest percentage of rooting success (78.56%), least percent mortality (3.5%) and the highest ROI (231.12%). The most number of roots was also obtained in T_1 (50 ppm NAA) with 1.13, while the tallest shoot (2.22 cm) and the longest root (15.31 cm) were produced in T_3 (150 ppm NAA) with the control treatment (T_0) performing poorly. Thus, dipping coffee shoot cuttings between 100 to 150 ppm NAA in the clonal chamber could enhance excellent root and shoot growth of the cuttings as well as the ROI. However, cuttings dipped above 150 ppm NAA had declining shoot and root growth performance. Hence, recommended for mass propagation of Robusta coffee for a short period of time.

Index Terms- *Clonal chamber, Coffea conephora, hormone, naphthalene acetic acid, rooting success*

I. Introduction

Coffee belongs to the botanical family Rubiaceae, a self-pollinating plant and has some 500 genera and over 6,000 species. It has a shallow root system and grows as a robust tree or shrub to about 10 meters tall. It flowers irregularly, taking about 10–11 months for cherries to ripen, producing oval-shaped beans. It originated from the upland forest of Ethiopia. It grows indigenously in Western and Central Africa from Liberia to Tanzania and South Africa to Angola and now grown as commercial crop in most of the tropical countries [13].

Coffee is an extremely powerful commodity, reigning as the world's most heavily traded product, behind petroleum [1]. However, commercial coffee production relies mainly on two species, *Coffea arabica* and *Coffea conephora* which accounts for more than 70% of the world's coffee production having superior quality and flavor relative to C. *conephora*. Nevertheless, *C. conephora* is the main ingredient of the coffee, which is increasingly consumed throughout the world and the popularity of coffee products worldwide is almost astonishing. It has a universal appeal to people of different income levels, ethnicities, and religions [6]. Aside from its beverage uses, coffee has been reported to be of clinical importance. It was found that caffeine, which reduces the swelling of blood vessels, can reduce both the intensity and frequency of headaches. It is also found that green coffee bean extract may work by reducing the absorption of fat and glucose in the gut. It may also reduce insulin levels, which would improve metabolic function [8].

However, coffee farming itself is a laborious activity which aside from many problems affecting the coffee production, propagation of poor quality seedlings is a grieve concern for majority of coffee farmers. Traditionally, coffee farmers used seed-derived plantlets rather than vegetative high yielding ones. Some farmers propagate seedlings using hard wood or stem, chopped and planted it directly into the soil without inducing it with any rooting hormone which takes much longer to produce than seed derived seedlings [2]. Such seedlings are not superior, vulnerable to diseases, gives poor yield, etc., which greatly affects the production [12].

Even today, coffee farmers are still having difficulty in producing quality coffee seedlings that can give good yield. Coffee seedlings produced through vegetative propagation method offers greater advantages to both coffee farmers and the coffee industry [4]. Such seedlings have high resistance to diseases, grows at a uniform rate making management more predictable as well as producing a more uniform and higher quality coffee beans which can have an important effect on both harvesting and processing and also economically [10]. Plants cannot germinate nor develop into a viable plant without the guidance of hormones [5]. Naphthalene Acetic Acid (NAA) is a plant hormone that plays fundamental role in vegetative propagation, shoot and root development, the cell cycle, chloroplast formation, bud formation and many more [3]. However, limited informations are available on nodal cuttings of C. *conephora* particularly on the rooting performance in response to different levels of NAA.

This study was conducted to determine the performance of *Coffea conephora* in response to different levels of NAA using shoot cuttings under a rooting chamber condition. Specifically, this study attempted to determine the root and shoot formations of coffee shoot cuttings in response to different levels of

naphthalene acetic acid (NAA) application, identify the best level of naphthalene acetic acid (NAA) that will give the highest root and shoot performance of coffee shoot cuttings and determine the cost and return analysis of all the treatments

II. Materials and Methods

This study was conducted at the Nursery House of the City Agriculture Office, Malaybalay City, Bukidnon, Philippines from June 2015 to October 2015. The following materials were used for the study: Small basins, Scissor, Fine sand, Sawdust, Soil, Coffee seedlings, Naphthalene Acetic Acid (NAA), Cellophane, Rubber band, 30 centimeter ruler, ball pen, calculator and digital camera. The *Coffea conephora* cultivar was used in the study. The cultivars where obtained from the Northern Mindanao Integrated Agricultural Research Center (NOMIARC), Dalwagan, Malaybalay City.

The study was laid-out in a Completely Randomized Design (CRD) with five treatments and replicated four times with ten cuttings per replication. The different treatments are as follows: T_0 – 0 ppm NAA, T_1 –50 ppm NAA, T_2 – 100 ppm NAA and T_3 – 500 ppm NAA, T_4 – 1000 ppm NAA.

In a portion of the nursery house/green house, seedbed was made using wooden planks as sides for seedbeds which were about 12.5 cm high, 75 cm wide and 1 m length. Before filling the seedbed with sand and soil, 20 pieces of small basins with diameter of 18.5 cm and holes punched at the bottom to drain access water out were placed in a manner shown in the diagram below (Appendix Figure 1). In between the basins in the seedbed, 50% of sand and 50% of sawdust where placed level with the height of the sides of the seedbed and the basins. The basins were then filled with pure fine sand as the rooting media.

A cutting consisted of one pair of leaves and the internodal below it. Where growth has been good, it was possible to obtain 2 or 3 cuttings from one plant, each with a pair of leaves and green pencil-thick flexible semi-hard wood. The cuttings were trimmed at an angle of 45 degrees above the leaf axil to facilitate moisture drainage and prevent terminal infection, and at the same time, angle 4 to 6 cm below the leaf axil (this ensures maximum callus formation for root development).

Two-third of each leaf was trimmed off; the cut stems were dipped briefly in respective NAA solutions for 5 minutes. The cuttings were then placed in the rooting medium, ensuring firm contact with the medium, and pushed into the medium to the point where the leaf petioles (stem) just rest on top of the medium. A clear plastic/polyethylene tunnel covering was made as shown in Figure 3 to cover the entire seed bed containing coffee cuttings. The tunnel had the same width of the seedbed with 75 cm high, well secured all round to make moisture tightly sealed. Watering was done when condensation on the inside of the polythene started to disappear. The study was terminated after 60 days from the day of planting the cuttings into the rooting media and the results were gathered to compile.

Among the data being gathered were percentage rooting success, percent mortality, length of roots, number of shoots, heights of shoots, number of leaves and cost and return analysis.

The analysis of variance (ANOVA) using Completely Randomized Design (CRD) was used to determine the level of significance. The Duncan's Multiple Range Test (DMRT) was used to test significant differences among treatment means.

III. Results and Discussion

Average Number of Leaves. The average number of leaves of coffee cuttings in response to different levels of naphthalene acetic acid (NAA) under clonal chamber condition is presented in Table 1. Statistical analysis revealed that different levels of NAA significantly affected the production of leaves of coffee stem cuttings.

Treatment 2 with 100 ppm NAA showed the most number of leaves with 2.38. It is significantly different from the rest of the treatments. However, no significant differences were likewise observed among Treatments 0, 4, 3 and 1.

The NAA is an auxin, hence, it helps in the proliferation of rooting formation of cuttings. While rooting is done, shooting is formed, thus the production of leaves is likewise maximized. There should be an equal ratio between shoots and roots [7]. The production of leaves is attributed to the fast shoot formation brought about by NAA at 100 ppm, thus obtaining the most number of leaves. Below and above 100 ppm NAA would have lesser production of leaves.

Table 1. Average number of leaves of coffee (*Coffea conephora*) cuttings in response to different levels of naphthalene acetic acid under clonal chamber condition

TREATMENTS	AVERAGE NUMBER OF LEAVES
T_0 (0 ppm NAA, control)	1.82^b
T_1 (50 ppm NAA)	1.18^b
T_2 (100 ppm NAA)	2.38^a
T_3 (150 ppm NAA)	1.61^b
T_4 (200 ppm NAA)	1.78^b
F-test	*
CV (%)	24.60

Means of same column followed by a common letter are not significantly different at 5% using DMRT.

Average Number and Length of Roots. Statistical analysis revealed no significant differences were observed on the number of roots regardless of the levels of applications of NAA to coffee stem cuttings as shown in Table 2. However, the lengths of the roots were greatly affected by the levels of NAA applied to the cuttings (also in Table 2).

Under a clonal chamber condition, the heat build-up inside causes the formation of adventitious roots in the basal ends [7]. Thus, it helped enhance the formation of roots of cuttings even if they were not dipped with NAA solutions. It showed, however that at 50 ppm (T_1), cuttings produced more than one root as compared to the rest of the treatments with less than one root per cutting.

However on the length of roots, it showed that T_3 dipped in 150 ppm NAA obtained the longest with 15.31 cm and is significantly different from the rest of the treatments. The control

treatment had the shortest with only 6.84 cm. This supports to the findings of Middleton [11] that low hormone concentrations (5 – 150 ppm) favored the rooting percentage more than high concentrations (1000 – 10,000 ppm) in most cuttings. Correspondingly, NAA as an aqueous solution or an emulsion to cuttings resulted in a greater number of roots formation than the untreated cuttings [9].

Table 2. Average number and length of roots of coffee (*Coffea conephora*) cuttings in response to different levels of naphthalene acetic acid under clonal chamber condition

TREATMENTS	AVERAGE NUMBER OF ROOTS/CUTTING	AVERAGE LENGTH OF ROOTS (cm)
T_0 (0 ppm NAA, control)	0.52	6.84^{b}
T_1 (50 ppm NAA)	1.13	7.94^{b}
T_2 (100 ppm NAA)	0.58	9.76^{b}
T_3 (150 ppm NAA)	0.54	15.31^{a}
T_4 (200 ppm NAA)	0.70	8.13^{b}
F-test	n.s.	**
CV (%)	21.07	24.61

Means of same column followed by common letters are not significantly different at 5% using DMRT.

Average Number and Height of Shoots. Table 3 reveals the number and heights of coffee cuttings. Statistical analysis showed that the number and heights of shoots of the cuttings were highly affected by the application of the different levels of NAA.

Cuttings in T_2 dipped in 100 ppm NAA obtained the most number of shoots and is statistically different from the rest of the treatments. Treatments 0 and 1 had the least number of shoots. On the other hand, cuttings in T_3 (150 ppm NAA) were the tallest with 2.22 cm, while the shortest were those in the control treatment with 0.95 cm long.

Shoot proliferation is the function of auxin as exhibited by NAA. As the NAA induces root formation on stem cuttings, the shoots are likewise induced. It was emphasized that rooting and shoot performances in cuttings were high, indicating that this plant growth regulator is effective in promoting roots as well as shoots in cuttings [2].

Table 3. Average number and heights of shoots of coffee (*Coffea conephora*) cuttings in response to different levels of naphthalene acetic acid under clonal chamber condition

TREATMENTS	AVERAGE NUMBER OF SHOOTS	AVERAGE HEIGHT OF SHOOTS (cm)
T_0 (0 ppm NAA, control)	0.78^{b}	0.95^{c}
T_1 (50 ppm NAA)	0.78^{b}	1.66^{b}
T_2 (100 ppm NAA)	1.65^{a}	1.41^{bc}
T_3 (150 ppm NAA)	0.89^{b}	2.22^{a}
T_4 (200 ppm NAA)	0.79^{b}	1.27^{bc}
F-test	**	**
CV (%)	23.98	25.47

Means of same column followed by common letters are not significantly different at 5% using DMRT.

Percent Rooting Success. The percent rooting success of coffee cuttings was highly affected by the levels of application of NAA (Table 4). Statistical analysis showed that T2 (100 ppm NAA) obtained the highest percentage of rooting success with 78.56% which is also not significantly different from those in Treatments 3 and 1 with 60.75% and 59.25%, respectively. The control treatment had the least percent of rooting success with 32.50%.

Low hormone concentrations (5 – 150 ppm) favored the rooting percentage more than high concentrations (1000 – 10,000 ppm). NAA at 5 to 150ppm induces early flowering and root formation in most of the plant cuttings (Andres, 2000). Thus, the higher the levels of concentration of NAA applied to the cuttings, the lesser is the percent of rooting success.

Table 4. Percentage (%) rooting success of coffee (*Coffea conephora*) cuttings in response to different levels of naphthalene acetic acid under clonal chamber condition

TREATMENTS	PERCENT ROOTING SUCCESS (%)
T_0 (0 ppm NAA, control)	32.50^{c}
T_1 (50 ppm NAA)	59.25^{ab}
T_2 (100 ppm NAA)	78.56^{a}
T_3 (150 ppm NAA)	60.75^{ab}
T_4 (200 ppm NAA)	46.00^{bc}
F-test	**
CV (%)	25.47

Means of same column followed by common letters are not significantly different at 5% using DMRT.

Percent Mortality. The percent mortality of coffee stem cuttings was not significantly affected by the levels of NAA application (Table 5). The conditions inside the clonal chamber favorably ensured rooting success of the cuttings, thus mortality was minimal.

Table 5. Percentage (%) mortality of coffee (*Coffea conephora*) cuttings in response to different levels of naphthalene acetic acid under clonal chamber condition

TREATMENTS	PERCENT (%) MORTALITY
T_0 (0 ppm NAA, control)	5.00
T_1 (50 ppm NAA)	5.00
T_2 (100 ppm NAA)	3.50
T_3 (150 ppm NAA)	5.00
T_4 (200 ppm NAA)	5.00
F-test	n.s.
CV (%)	15.85

Cost and Return Analysis. The cost and return analysis is presented in Table 6. Treatment 2 applied with 50 ppm NAA obtained the highest gross sales of US $53.19. It incurred total expenses of US $16.06 with the highest net income of US $37.13; thus, obtained the highest return on investment (ROI) of 231.12%. The higher the concentration applied with NAA solution, the higher are the expenses incurred beyond 100 ppm NAA application to the cuttings, thus, the lesser the ROI obtained. The lowest ROI was registered in the control treatment (T_0) with no application of any NAA solution.

Table 6. Cost and return analysis (US $) of coffee (*Coffea conephora*) cuttings in response to different levels of naphthalene acetic acid under clonal chamber condition

TREATMENTS	GROSS SALES	EXPENSES	NET INCOME	ROI (%)
T_0 (0ppm NAA, control)	42.55	15.43	26.91	174.40
T_1 (50 ppm NAA)	53.19	15.85	26.70	168.46
T_2 (100 ppm NAA)	53.19	16.06	37.13	231.12
T_3 (150 ppm NAA)	42.55	17.77	24.79	139.50
T_4 (200 ppm NAA)	42.55	19.89	22.66	113.93

IV. Conclusion

The use of naphthalene acetic acid between 100 to 150 ppm could enhance root and shoot growth as well as mass produce *Coffea cenephora* shoot cuttings in shorter period of time

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