

# Effects of cutting position along mother plants on rooting of hybrid coffee varieties

Jeremiah M. Magesa<sup>1\*</sup> • Theodosy J. Msogoya<sup>2</sup> • Cornel L. Rweyemamu<sup>2</sup>

<sup>1</sup>Tanzania Coffee Research Institute, P.O Box 3004, Moshi, Tanzania.

<sup>2</sup>Department of crop Science and Horticulture, P.O Box 3005, Chuo Kikuu Morogoro, Tanzania.

\*Corresponding author. E-mail: jeremagesa@gmail.com; jeremiah.magesa@tacri.org. Tel: +255-757-563165, +255-655-563165.

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**Abstract.** The study was conducted on-station at Tanzania Coffee Research Institute to evaluate the effect of stem cuttings position along the mother plants on rooting of hybrid coffee varieties. Stem cuttings were assessed in a rooting medium of forest soil and sand at a ratio of 2:1 by volume under semi-controlled environment. A split plot experiment in a randomized complete block design with four replications was used. The main factor was five Arabica coffee varieties and the sub-factor consisted of four types of position of stem cuttings, namely base, middle, apex and control (mixture). Four months after planting, stem cuttings were evaluated for root growth characteristics. Data collected were subjected to analysis of variance (ANOVA) using CoStat software and treatment means were separated based on Tukey's test at ( $P \leq 0.05$ ). Results obtained indicate that positions of stem cuttings along the mother plant had a significant effect ( $P = 0.04$ ) on rooting of coffee varieties with stem cuttings taken from basal and middle positions having the highest rooting percentage with varieties N39-1 and KP423-2 having the highest rooting percentage. Further, interaction between varieties and position of stem cuttings significantly ( $P = 0.04$ ) affected the rooting percentage and the number of lateral roots at ( $P = 0.01$ ). The results from this study indicate that clonal multiplication of coffee stems cuttings is affected by varieties and position taken along the mother plant. Further studies are required to determine yield performance of the stem cuttings in relation to their positions along the mother plants.

**Keywords:** Basal, clonal propagation, position, semi-controlled, stem, varieties.

## INTRODUCTION

Coffee can be propagated by seeds or vegetatively using cuttings or grafting. Propagation by seeds leads to genetic variability due to segregation of genes during fertilization while vegetative propagation through stem cuttings guarantees uniformity as it maintains the genetic make-up of the planting materials (Kumar *et al.*, 2006). However, stem cuttings sometimes have low rooting percentage (Etienne *et al.*, 2002) if due instructions are not adhered to. The rooting ability of stem cuttings is the function of species, type of cutting, physiological status of the plant and suitability of the rooting medium and environment (Hartmann *et al.*, 2002).

Normally, coffee mother plants are bent to produce shoots from the base to the apex at each stem nodes. Tanzania coffee research institute (TaCRI) has been multiplying the improved hybrid coffee by vegetative propagation through stem cuttings by bulking orthotropic stem cuttings from the base, middle and apex of the vigorous mother plants of hybrid coffee varieties (TaCRI, 2011). Despite the use of the above practices, the average rooting of stem cuttings has remained low. The low rooting ability of the hybrid is possibly associated with the mixing of juvenile and aged stem cuttings from different positions along the mother plants under controlled

**Table 1.** Characteristics of the five improved hybrid Arabica coffee varieties used\*.

Variety name	Yield (kg/ha) clean coffee	Bean size (AA+A %)	Height (cm)	Stem girth (cm)	Leaf size (mm <sup>2</sup> )
N39-1	2 056	77	171.00	3.63	5 341
N39-2	2 708	77	201.32	3.61	6 100
N39-4	1 961	74	165.0	3.54	4 429
KP423-1	2 225	80	207.74	3.53	5 219
KP423-2	1 851	68	217.46	3.57	7 434

\*According to Teri *et al.* (2011).

conditions.

Stem cuttings collected from the base of mother plants have been reported in other plant species to be more juvenile and produce roots easier than those collected from the apex (Yeboah *et al.*, 2009). However, there are no research results on the effects of position of stem cuttings along the mother plants on the rooting of hybrid coffee. The objective of this study was to determine the effect of position of stem cuttings along the mother plants on rooting of hybrid coffee varieties.

## MATERIALS AND METHODS

### Study sites description and planting materials

The experiment was set at Tanzania Coffee Research Institute (TaCRI), Lyamungu station, Moshi, Tanzania in December 2013 to March 2014. The station is located at latitude 03°14.699' S and longitude 037°14.762'E with altitude of 1268 m a.s.l. The soil is classified as Nitisols volcanic origin with pH range of 4.8 to 5.7 and the climate is classified as tropical with two different seasons: warm-dry (August to February) and rainy (March to June). The average annual rainfall is about 1250 mm, with temperature range of 17 to 28°C and relative humidity range of 40 to 90%.

### Environmental condition of the study area during experimentation

The mean temperature was 21.8°C with the minimum temperature 21.0°C in February and maximum temperature 22.4°C in January; whereas the mean relative humidity was 85.0% with the minimum relative humidity of 85% in February and the maximum relative humidity 85.6% in December.

Uniform orthotropic stem cuttings of selected five improved hybrid coffee varieties commonly grown in Tanzania were used as shown in Table 1. Stem cuttings with 4-8 internodes were harvested early in the morning from ten years vigorous mother stocks at TaCRI nursery. To minimize stress, the stem cuttings were prepared under shade into three nodes with six leaves as

described by Twardowski *et al.* (2012). Leaves were cut half to reduce water loss by transpiration (Rezende *et al.*, 2010; TaCRI, 2011).

### Preparation of rooting medium

Forest soil was collected at a depth of 10 cm from Sawe forest reserve as described by Amri *et al.* (2009) whereas fine sand soil was collected from Weruweru River. The rooting medium was prepared by thorough mixing forest soil and fine sand sieved through 2 mm sieve at a ratio of 2:1 by volume, and moistened to 20 to 35% moisture content. The medium was sterilized by heating at 80°C using soil sterilizing machine for one hour, cooled for 24 hours and then moistened to 20 to 35% moisture content as recommended by Chong *et al.* (2008) and Yeboah *et al.* (2011). Temperature (°C), relative humidity (%) and moisture content (%) of the rooting medium inside the propagation box were recorded throughout the experiment.

Physical and chemical characteristics of the rooting medium was determined at the beginning of the experiment are shown in Table 2. Nitrogen fertilizer (N.P.K-20:10:10) at a rate of 5 g/m<sup>2</sup> was incorporated into the rooting media at planting to improve seedlings quality after rooting of the stem cuttings (TaCRI, 2011).

### Experimental design and treatments

A split plot experiment in a randomized complete block design (RCBD) with four replications was used. The main plot factor consisted of five hybrid coffee varieties (N39-1, N39-2, N39-4, KP423-1 and KP423-2) and the sub plot factor consisted of four types of positions of collecting stem cuttings, namely base, middle, apex and mixture. The conventional treatment used was the mixture of the above three types of stem cuttings applied as the control. Stem cuttings for the control (mixture) were collected in equal number from the base, middle and apex parts of the mother plants.

A propagation box measuring 3.65 × 1 × 0.9 m made of cement blocks was used. The propagation box with three sections consisting of 15 cm thick gravels formed the base

material, followed by 15 cm thick rooting medium. The third section of 60 cm was left empty. The size of experimental unit was 0.8 m<sup>2</sup> and the spacing between plots was 15 cm. This resulted into 320 experimental units in four replicates. Moisture of the rooting medium was maintained at 20% by spraying water using a 15 Litre knapsack sprayer (SOLO 425 Classic, Germany) before planting (TaCRI, 2011). A digital moisture meter Ridde AG (LA3200) was used to determine the moisture content of the rooting medium.

The basal ends of 1 to 3 cm stem cuttings were sterilized by dipping into 5 g/L solution of copper oxychloride (50 WP) for controlling fungal and bacterial diseases, and air-dried at 25°C for five minutes followed by immediate planting into the rooting medium as described by Alikhani *et al.* (2011). Planting was done in the medium using a dibble at 5 × 5 cm and the planting of 2.5 cm were used as recommended by Akwatulira *et al.* (2011) and TaCRI (2011). After planting, the stem cuttings were sprayed with 5 g/l solution of copper oxychloride (50 WP) using a knapsack sprayer. The propagation box was covered with transparent white polyethylene sheets (5 mm thick) supported with semi-circular iron rods to conserve humidity at 60 to 85% (Pandey *et al.*, 2011) under the black shade net absorbing 30% of ultra violet (UV) light.

Irrigation in fine droplets using a knapsack sprayer was applied whenever the moisture content of the rooting medium was about 20% in order to maintain moisture content at 20 to 35%. Spraying of 5 g/L solution of copper oxychloride (50 WP) was used to control fungal and bacterial diseases as described by TaCRI (2011).

### Data collection

Data were taken four months after planting by gently uprooting the stem cuttings from the rooting medium as recommended by Yeboah *et al.* (2009). Rooting of stem cutting was considered if it had at least one visible lateral and fibrous roots of ≥2 cm long without magnification according to Ou Yang *et al.* (2015). Data were collected on number of rooted cuttings (lateral and fibrous roots ≥2 cm long), number of roots (laterals and fibrous ≥2 cm long), rooting percentage and lateral root length as described by Pandey *et al.* (2011).

The number of lateral and fibrous roots (roots with ≥2 cm long) were counted and the longest lateral roots were measured with a graduated ruler from the collars of the stem cuttings to the apex of longest laterals (Ou Yang *et al.*, 2015); number of fibrous roots per stem cutting was determined by counting stem cuttings which rooted by considering laterals and fibrous roots with ≥2 cm root long (Rosier *et al.*, 2004; Ou Yang *et al.*, 2015) and percentage of rooted stem cuttings was determined by the following formula as described by Hae and Funnah (2011): %T = N/n × 100; where: T = % rooted stem cuttings, N = total number of rooted stem cuttings, n =

total number of planted stem cuttings.

### Data analysis

Data collected were subjected to analysis of variance (ANOVA) using CoStat software version 6.311 and declared significant at P ≤ 0.05 using the following statistical model for the split-plot design as described by Kuehl (2000).

$Y_{ijk} = \mu + \alpha_i + P_k + d_{ik} + \beta_j + (\alpha\beta)_{ij} + \varepsilon_{ijk}$ . where  $\mu$  = the general mean;  $\alpha_i$  = the effect of the *i*th level of factor A;  $P_k$  = the effect of the *k*th block;  $d_{ik}$  = the whole-plot random error;  $\beta_j$  = effect of the *j*th level of factor B;  $(\alpha\beta)_{ij}$  = the interaction effect between factors A and B;  $\varepsilon_{ijk}$  = is the sub-plot random error. The differences between the treatment means were separated using Tukey's test method at P ≤ 0.05.

## RESULTS

### Effect of coffee hybrid varieties on rooting

The results of varietal effect on rooting are as presented in Table 3. Coffee hybrid varieties had a highly significant effect (P = 0.00) on rooting with variety KP423-2 having the highest rooting of 58.1% and was significantly different from varieties N39-4 and KP423-1 with rooting of 41.6 and 38.5%, respectively. Variety KP423-1 had the lowest rooting of 38.5% but not significantly different (P ≤ 0.05) from variety N39-4 and N39-2 with rooting of 41.61 and 45.95%, respectively. Further, varieties did not significantly (P ≤ 0.05) affect the number of fibrous roots, lateral root length and number of lateral roots.

### Effect of position of stem cuttings on rooting of hybrid coffee varieties

The position of stem cuttings along the mother plants significantly (P = 0.04) affected the rooting of hybrid coffee varieties Table 4. Stem cuttings from apex had the lowest rooting of 39.59% and was significantly different from stem cuttings collected from the base and middle of the mother plants with rooting percentage of 55.6 and 51.5, respectively. No significant difference (P ≤ 0.05) was found for cutting position for root length, number of lateral roots and number of fibrous roots per cutting.

### Interaction effect between hybrid coffee varieties and positions of stem cuttings on mother plants on rooting of stem cuttings

Results on interaction effect between hybrid coffee varieties and positions of stem cuttings on mother plants

**Table 2.** Physical and chemical characteristics of forest soil + sand rooting media.

Property	Method used	Value	Remarks*
Physical			
Soil bulk density ( g cm <sup>-3</sup> )	Core	0.48	Medium
Moisture (%)	Moisture meter	34.70	Medium
Chemical			
pH	1:2.5 Soil water suspension	4.9	Low
Electrical conductivity (dS m <sup>-1</sup> )	1:2.5 Soil water ratio	0.46	Low
CEC (cmol (+) kg <sup>-1</sup> )	Ammonium acetate method	14.0	Medium
Exchangeable Calcium (cmol (+) kg <sup>-1</sup> )	Atomic Absorption Spectrometry	6.23	Low
Exchangeable Magnesium (cmol (+) kg <sup>-1</sup> )	Atomic Absorption Spectrometry	0.77	Low
Exchangeable Potassium (cmol (+) kg <sup>-1</sup> )	Flame photometry	0.82	Low
Total Nitrogen (%)	Semi micro kjeldahl	0.17	Medium
Available Phosphorus (mg kg <sup>-1</sup> )	Bray and Kurtz 1	12.95	Low
Organic Carbon (%)	Walkley and Black	4.72	High
Exchangeable Aluminium (cmol (+) kg <sup>-1</sup> )	Titrimetric	0.3	Low
Extractable Copper (mg kg <sup>-1</sup> )	Atomic Absorption Spectrometry	4.25	Medium
Extractable Iron (mg kg <sup>-1</sup> )	Atomic Absorption Spectrometry	393.3	High
Extractable Manganese (mg kg <sup>-1</sup> )	Atomic Absorption Spectrometry	102.50	Medium
Extractable Zinc (mg kg <sup>-1</sup> )	Atomic Absorption Spectrometry	5.42	Medium

\*According to Beyl and Trigiano (2008).

**Table 3.** Effects of varieties of stem cuttings on rooting of hybrid coffee varieties.

Varieties	% rooted cuttings	Number of fibrous roots/cuttings	Lateral root length (cm)	Number of lateral roots/cutting
KP423-1 control	38.5 <sup>c</sup>	71.2	12.1	2.6
KP423-2	58.1 <sup>a</sup>	75.9	12.7	2.6
N39-2	45.9 <sup>abc</sup>	58.2	12.3	2.6
N39-1	56.6 <sup>ab</sup>	60.0	11.3	2.3
N39-4	41.6 <sup>bc</sup>	82.9	12.7	2.7
Mean	48.2	69.6	12.2	2.6
CV (%)	29.8	39.2	22.5	21.4
P-values	0.00	0.11	0.65	0.35

Means followed by the same letter in the same column are not significantly different at ( $P \leq 0.05$ ) according to Tukey's Test.

**Table 4.** Effect of position of stem cuttings on rooting of hybrid coffee varieties.

Position of cutting	% rooted cuttings	Number of fibrous roots/cutting	Lateral root length (cm)/cutting	Number of lateral roots/cutting
Control (mixed cuttings)	45.8 <sup>ab</sup>	71.7	12.2	2.5
Cuttings from the base	55.6 <sup>a</sup>	72.6	12.7	2.8
Cuttings from the middle	51.5 <sup>a</sup>	74.5	12.8	2.7
Cuttings from the apex	39.5 <sup>b</sup>	59.9	11.2	2.4
Mean	48.1	69.6	12.2	2.6
CV (%)	27.8	35.1	18.1	19.5
P-values	0.04	0.24	0.11	0.08

Means followed by the same letter in the same column are not significantly different at ( $P \leq 0.05$ ) according to Tukey's Test.

**Table 5.** Interaction effect between hybrid coffee varieties and position of stem cuttings on mother plants on rooting of stem cuttings.

Varieties × Positions of stem cuttings	% rooted cuttings	Number of fibrous roots/cutting	Lateral root length (cm)/cutting	Number of lateral roots/cutting
N39-1 x cutting from the base	51.1 <sup>abc</sup>	59.6	10.8	2.2 <sup>ab</sup>
N39-1 x cutting from the middle	62.4 <sup>abc</sup>	65.8	12.4	2.7 <sup>ab</sup>
N39-1x cutting from the apex	43.0 <sup>abc</sup>	44.4	9.4	2.2 <sup>ab</sup>
N39-1 x control (mixed cuttings)	70.0 <sup>ab</sup>	70.4	12.7	2.2 <sup>ab</sup>
N39-2 x cutting from the base	47.5 <sup>abc</sup>	57.6	12.4	2.5 <sup>ab</sup>
N39-2 x cutting from the middle	55.0 <sup>abc</sup>	70.4	14.6	3.2 <sup>a</sup>
N39-2 x cutting from the apex	46.3 <sup>abc</sup>	58.2	11.6	2.5 <sup>ab</sup>
N39-2 x control (mixed cuttings)	34.9 <sup>bc</sup>	46.7	10.7	2.2 <sup>ab</sup>
N39-4 x cutting from the base	50.3 <sup>abc</sup>	96.8	14.5	3.2 <sup>a</sup>
N39-4 x cutting from the middle	44.0 <sup>abc</sup>	63.7	10.4	2.2 <sup>ab</sup>
N39-4 x cutting from the apex	38.5 <sup>abc</sup>	76.4	12.5	2.5 <sup>ab</sup>
N39-4 x control (mixed cuttings)	33.4 <sup>c</sup>	94.7	13.2	3.0 <sup>ab</sup>
KP423-1 x cutting from the base	56.0 <sup>abc</sup>	57.6	12.2	2.7 <sup>ab</sup>
KP423-1 x cutting from the middle	34.2 <sup>c</sup>	93.5	12.6	2.7 <sup>ab</sup>
KP423-1 x cutting from the apex	31.7 <sup>c</sup>	69.2	11.8	3.0 <sup>ab</sup>
KP423-1 x control (mixed cuttings)	31.9 <sup>c</sup>	64.6	11.9	2.2 <sup>ab</sup>
KP423-2 x cutting from the base	73.2 <sup>a</sup>	91.3	13.8	3.2 <sup>a</sup>
KP423-2 x cutting from the middle	62.0 <sup>abc</sup>	79.1	13.8	2.7 <sup>ab</sup>
KP423-2 x cutting from the apex	38.2 <sup>abc</sup>	51.1	10.8	1.7 <sup>b</sup>
KP423-2 x control (mixed cuttings)	58.7 <sup>abc</sup>	82.0	12.3	2.7 <sup>ab</sup>
Mean	48.1	69.6	12.2	2.6
CV%	29.8	35.1	18.1	21.1
P-values	0.04	0.24	0.12	0.01

Means followed by the same letter in the same column are not significantly different at ( $P \leq 0.05$ ) according to Tukey's Test.

on rooting of stem cuttings are shown in Table 5. Interaction effect on the two factors used had a significant effect ( $P = 0.04$ ) on percentage rooted cuttings and significantly ( $P = 0.01$ ) affected the number of lateral root length. Variety KP423-2 x cutting from the base treatment had the highest rooting of 73.2% which was significantly different from variety KP423-1 x cutting from middle, N39-4 x control, KP423-1 x control, KP423-1 x cutting from the apex treatments with rooting percentage of 34.2, 33.4, 31.9 and 31.7%, respectively. Further, significant differences were found between variety N39-2 x cutting from the middle, N39-4 x cutting from the base, KP423-2 x cutting from the base with number of lateral roots of 3.2 and variety KP423-2 x cutting from the apex treatments with number of lateral root of 1.7.

Moreover, variety N39-1 performed best when cuttings were collected from middle with 62.4% and followed by the control which had 70.0% rooted cuttings. However, variety KP423-2 had the best performance when cuttings were collected from base with rooting percentage of 73.2% and those from the middle with 62.0%. Similarly, varieties KP423-2 and N39-3 had the highest number of lateral roots (3.2) when stem cuttings were collected from the base followed by N39-4 with (3.0) when cuttings were collected from the control (mixed cuttings).

## DISCUSSION

### Effects of hybrid coffee varieties on rooting of stem cuttings

The significant differences on root growth characteristics may have occurred due to variation in varietal characteristics. Such variations in rooting percentage recorded in this study were due to the differences in the genetic, physiological and morphological characteristics of the varieties used as also reported by Hartmann *et al.* (2002) and Amisah *et al.* (2008).

Research by Bartolini *et al.* (1996) using Vitis 140 Ruggeri cultivar concluded that rooting ability was related to water soluble carbohydrate contents of stem cuttings which is a physiogenic characteristic whereas Dick *et al.* (2004) found that there was a genetic variation in rooting potential in a tropical tree of Africa (*Triplochiton scleroxylon* K. Schum) where the rooting ability was mediated through genetic differences in cutting morphology or physiology within species. This also has been reported by Beyl (2008) and Guan *et al.* (2015).

During rooting, the carbohydrates act as source of energy and as constitutive elements for the newly formed cells (Bartolini *et al.*, 1996). It is reported that in addition

to soluble carbohydrates, root promoting substances such as phenolic compounds and auxins are also produced in leaves and transported to the base of the cuttings where they protect auxins especially indole-3-acetic acid (IAA) from being oxidized and hence more auxins become available to induce roots (Hartmann *et al.*, 2002; De Klerk *et al.*, 2011).

Varieties with larger leaf and stem sizes are reported to have more nutrient storage and water soluble carbohydrate contents than others (Akinyele, 2010; Rana and Sood, 2012). The five hybrid coffee varieties used in this study differed in morphological characteristics Table 1. where varieties KP423-2 that had the highest rooting of 58.09% had also the largest leaf size of 7434 cm<sup>2</sup> and variety KP423-1 that had the lowest stem size and relatively high leaf size of 6519 cm<sup>2</sup> recorded the lowest rooting percentage of 38.48. The results obtained in this study are also consistent with previous research conducted by Christopher *et al.* (2002) on Coleou tree (*Solenostemon scutellarioides* L.) and those by Ky-Dembele *et al.* (2011) who worked on African mahogany tree (*Khaya senegalensis* A. Juss) and Gehlot *et al.* (2015) worked on *Azadirachta indica* A. Juss (Neem) and concluded that larger leaf and stem sizes significantly increased rooting of stem cuttings.

#### Effect of position of stem cuttings on rooting of hybrid coffee varieties

The significant differences recorded between basal and middle stem cuttings along mother plants on rooting of stem cuttings in this study could be related to the physiological status of the mother plants from which they were collected (Hartmann *et al.*, 2002; Beyl, 2008). Stem cuttings collected closest to the base of the mother plants were chronologically the oldest but the most juvenile physiologically and thus having the ability to form roots easier than those at the apex as reported by Beyl (2008). The results are consistent with previous studies which show that plant regeneration potential is high during juvenile phase, and declines as the plant ages. Similarly Amri *et al.* (2010) found that stem cuttings collected from juvenile African Blackwood (*Dalbergia melanoxylon* Guill. and Perr.) performed better in all rooting parameters than those collected from mature stock plants.

Further, Wen-wen *et al.* (2015) reported significant rooting percentage of stem cuttings of three *Salix* species (*Salix fragilis* L., *S. matsudana* L. and *S. babylonica* L.) collected at the base compared to the apex and similar results were found by Graves (2002) in stem cuttings of *Rhamnus caroliniana* (Walter) A. Gray commonly known as Carolina Buckthorn, which are in agreement with the findings obtained in this study.

The high rooting percentage is possibly associated with higher levels of root-promoting substances especially auxin and sugars which are higher in juvenile phase than the root-inhibiting substances such as cinnamic acid

derivatives, kinetins, zeatins and borate (Hartmann *et al.*, 2002; Amissah *et al.*, 2007; Islam *et al.*, 2010). The high rooting ability of cuttings during juvenile phase and middle over those at apex is attributed to the effect of changes in plant developmental process that occur with aging. Such effects are known as maturation or ontogenetic aging, which is commonly found in the upper parts of the tree and least advanced near the base/crown (Browne *et al.*, 1997; Beyl, 2008). This means stem cuttings from the apex are too mature and highly lignified to develop roots than the cuttings collected at the juvenile phase (Cheng *et al.*, 2008).

#### CONCLUSION AND RECOMMENDATION

From this study it shows that stem cuttings from the base and middle are the best for clonal propagation because of their highest ability of rooting thus can be used for mass multiplication of KP423-2 and N39-1 hybrid coffee varieties that had the highest rooting percentage. Moreover, interaction effect between varieties and positions of stem cuttings indicates that varieties N39-1, KP423-1 and KP423-2 had the highest rooting percentage and number of lateral roots when cuttings were collected from base and middle. Further studies are required to determine yield performance of the stem cuttings in relation to their positions along the mother plants.

#### CONFLICT OF INTERESTS

The authors have declared they do not have any conflict of interest.

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