Substrate and genotype effects on kola (Cola nitida [Vent.] Schott and Endlicher.) tree cuttings growth in nursery

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The kola tree (Cola nitida) belongs to family Malvaceae grown mainly for its seed. It is highly prized for its socio-cultural and industrial uses. However, there is a significant high variability due to a high proportion of allotropy in the reproduction mode. The influence of substrate and genotype on kola tree cuttings has been studied in order to propagate it vegetatively. Six growing substrates: 100% Black soil, 50% Black soil + 50% Wood sawdust, 100% Wood sawdust, 50% Soil + 50% Black-Coco, 100% Black-Coco, 50% Wood sawdust + 50% Black-Coco were tested on genotypes 305 and D9L7A1 using a split-plot experimental design with genotype as main factor and substrate as subplot treatment replicated three times. The experimental unit constituted 20 cuttings. Six months after transplanting, the average success rate of tunnel cutting from the kola tree to the nursery was 76.2±7.6%. The best success rates were obtained with 100% Wood sawdust, 100% Black soil and 50% Black coco + 50% Wood sawdust substrates with 85±4.5, 80.8±6.6 and 79.7±5.8%, respectively, regardless of the plant material used. The substrates 100% Black soil and 100% Wood sawdust gave the best results on growth parameters such as aerial dry biomass, height growth, roots number and new formed leaves. It also showed that genotype D9L7A1 had a higher vegetative leaves and roots development than genotype 305. These genotypes were well suited for cutting.

Key words: Cola nitida, cuttings, genotype, substrate.

INTRODUCTION

The kola tree (Cola nitida) is a fruit tree whose average height is about 25 m. It is a species that belongs to the genus Cola of the family Malvaceae (Whitlock et al., 2001). Côte d’Ivoire is the world’s leading producer of Kola nuts ahead of Nigeria with an estimated 260,000 tons of fresh kola nuts per year (MINADER, 2018), for a
turnover of around one hundred billion CFA francs (Aloko-N’Guessan, 2000).

Despite this economic importance, kola nut production in Côte d’Ivoire faces several challenges. Domestication and cultivation of kola require the adoption of propagation techniques adapted to the crop species. The objective of this study is to optimize kola (*C. nitida*) vegetative propagation by cutting under tunnel. Indeed, the kola nut has a slow germination process and the plant enter production late (5 to 6 years after planting). To shorten the time to production, the cutting of the kola tree has been initiated (Séry et al., 2019). Unfortunately, in nurseries, the survival rate of the plants is low for the species and the growth of plants from cuttings is very slow. It takes 12 to 18 months to obtain plants suitable for transplanting in the field. In addition, the quality of the roots from these plants is poor (Séry et al., 2019). It is therefore necessary to propose methods to improve the survival rate of cuttings, to accelerate the growth and root development of the kola tree. Previous research on several tropical tree species, including the kola tree, has indicated a wide range of factors influencing the rooting of cuttings in nurseries. These factors include genotype, substrate type, leaf area, cuttings length and rhizogenic substances (Paluku et al., 2018). In this study, substrate type and genotype are examined. Specifically, the test aims to determine the effect of substrate and genotype on the cuttability (survival rate, growth and root development) of the kola tree. On the other hand, to improve the survival (success) rates and rooting of the kola tree’s cuttings.

MATERIALS AND METHODS

Description of the experimental site

The test was set up in April 2019, at the beginning of the rainy season, at the site of the nursery of the Centre National de Recherche Agronomique (CNRA) of Man, in western Côte d’Ivoire (7° 19.130’ N; 8° 19.452’ W); This six-month test ended in October 2019. The rainfall in the Man zone is of a monomodal type. The dry season from April to October. The site received an average annual rainfall in 2018 of 1632 mm. The temperature in 2018 varied from 23 to 27°C.

Plant material

The plant material used consisted of 720 kola tree cuttings. These cuttings were collected at the CNRA research station in Divo on two (02) genotypes with 320 cuttings per genotype. These two genotypes have been identified by the following codes D9L7A1 and 305. These genotypes were selected based on their productivity.

Technical equipment

The technical equipment used for this study consisted of pruning shears for sampling and dressing the cuttings and a decameter for measuring the circumference and height of the cuttings. White plastic bags, sealed with a stapler and stored in ice boxes were used for the conservation of the cuttings during transport. Three types of materials were used for the formulation of the 6 growing media (substrate) in this test. These were compost (black soil), decomposed wood sawdust, industrial substrate based on decomposing coconut fiber "Black-coco". Bags 30 cm high by 15 cm diameter were used for transplanting the cuttings. IVORY × 80% WP fungicide (a.m.: Maneb, Manufacturer: ARYSTA Life science) was used for preventive treatment from the transplanting of cuttings.

Description of the test

Experimental design

The experimental design was a Split-Plot with two (02) factors which are substrate type and genotype. We had the substrate type in small plot and the genotype in large plot. Twenty (20) pots (bags used for the nursery) containing each a cutting of the same genotype used per treatment. The substrate type with six (06) modalities was the main factor. The different modalities were: S1: 100% Black soil; S2: 50% Black soil + 50% Wood sawdust; S3: 100% Wood sawdust; S4: 50% Black soil + 50% Black coco; S5: 100% Black coco; S6: 50% wood sawdust + 50% Black coco. The genotype with two (2) modalities (C1: D9L7A1 and C2: 305) was the secondary factor. A total of 12 treatments were studied in this trial. These treatments were repeated three times. A total of 360 cuttings from the same genotype were used for this test (120 cuttings per genotype for each repetition). This gives a number of 720 cuttings for both genotypes. The cutting method used was tunnel cutting. The reinforcement of the tunnel to house the pots (bags) containing the cuttings were made of 2.4 m long hoops connected by bamboo slats. The tunnel was covered with a transparent plastic sheeting 100 μ thick and 2.6 m wide, plated on the sides and ends with stones and bamboos. The tunnels were placed under a nursery shelter which consisted of a 2 m high palm tree structure that allowed about 50% of the total light to pass through (Figure 1).

Sampling and transplanting of cuttings

Three hundred and twenty (320) cuttings per genotype were collected early in the morning from the semi-lined terminal twigs in a plot at Divo station. A total of 720 cuttings were collected using pruning shears for the 2 genotypes used in this study. The cuttings from each tree were transported in plastic bags containing water to minimize dehydration. The bags, closed with a stapler, were placed in glaciers to maintain humidity. The size of the cuttings was between 10 and 12 cm. The cuttings had 4 leaves cut in half. For the implementation of this test, in each tunnel, the pots were filled with the six growing substrates previously homogenized and disinfected. The cuttings were transplanted by hand to a depth of about 3 cm. The water storage capacities of the six growing media used in the pots were determined.

Assessment of the water storage capacities of the different growing substrates

The water storage capacities of the six growing substrates were determined (Table 1) according to Colombani et al. (1973). A 30 cm high by 15 cm diameter pot culture, perforated, was completely filled with each substrate. Each pot was then saturated with a known amount of water. The excess water was then drained for 2
Figure 1. Tunnel cuttings.

Table 1. Water storage capacity of growing substrates.

| Substrate type                          | Water storage capacity of substrates (ml/L) | Substrate water storage rate (%) |
|-----------------------------------------|---------------------------------------------|----------------------------------|
| 100% Black soil                         | 400                                         | 40                               |
| 100% Black-coco                         | 300                                         | 33                               |
| 100% Wood sawdust                       | 420                                         | 42                               |
| 50% Black soil + 50% Black-coco         | 440                                         | 44                               |
| 50% Black soil + 50% Wood sawdust       | 800                                         | 80                               |
| 50% Black coco + 50% Wood sawdust       | 360                                         | 36                               |

days and the water storage capacity \((W.S.C)\) was measured for each substrate according to the following formula:

\[
W.S.C = \left( \frac{\text{amount of water supplied (ml)} - \text{quantity of water drained after 48 hours (ml)}}{\text{quantity of water supplied (ml)}} \right) \times 100
\]

The water storage rate (%) of the substrates was determined.

**Carrying out the test**

The arrangement of the pots and the phytosanitary treatment were carried out the day before the cuttings and placed in tunnels. For phytosanitary treatment, the fungicide IVORY 80% WP (a.m. maneb, Manufacturer: ARYSTA Lifescience) was used (70 g in 2 litres of water applied in the pots). The cuttings were watered every 2 days with 100 ml/pot. For growth parameters measurement, out of the 20 pots of each treatment, the number of live plants was recorded at six months to quantify the survival (success) rate. Root development (length and number of roots), number of new leaves, height of seedling and aerial and root dry biomass were assessed after six months. This dry biomass was evaluated using an electronic scale after drying in the open air for two weeks.

**Statistical analysis of data**

For the parameters examined, a comparison of the means between the different factors and the different treatments was made through the analysis of simple variance (ANOVA). When a significant difference is observed between the treatments for a given factor, the ANOVA is completed by post-hoc tests, in particular the Newman-Keuls test to identify significant differences between the means at the 5% threshold. For all these tests, STATISTICA 7.1 software was used. The survival rate \((S.R)\) was calculated according to the following formula:

\[
S.R = \left( \frac{\text{number of living plants}}{\text{initial number of plants}} \right) \times 100
\]

A hierarchical bottom-up classification was carried out for structuring the culture substrates according to their ability to improve the cuttings of the kola tree.

**RESULTS**

**Assessment of the water storage capacities of the different growing substrates**

The water storage capacity of the six growing substrates was determined (Table 1). The substrate (50% Black soil + 50% Wood sawdust) has the highest water storage capacity with 800 ml/L followed by the substrates 50% Black soil + 50% Black-coco and 100% Black soil with 440 and 400 ml/L, respectively. They retain at least 40% of the water used for watering. The black-coco substrate
Substrate and genotype effects on cuttings survival rate

Substrate and genotype effects on cuttings survival rate were evaluated through an analysis of variance. The analysis (Table 2) revealed the significant effect \( p=0.0001 \) of the substrate on improving the survival rate of kola tree cuttings in nurseries. Unlike the substrate, no effects of genotype \( p=0.447 \) and “Substrate×Genotype” interaction \( p=0.745 \) were noted.

For the whole trial, the average success rate for cuttings from kola trees in nurseries regardless of the plant material was 76.2±7.6. The best success rates for cuttings from the kola tree were obtained with substrates 100% Wood sawdust, 50% Black coco + 50% Wood sawdust and 100% Black soil with 85±4.5, 80.8±6.6 and 79.7±5.8, respectively (Table 3). Mortality rates were high with substrates 50% Black soil + 50% Black coco, 50% Black soil + 50% Wood sawdust and 100% Black coco.

Table 2. Analysis of Variance (ANOVA) of the “Substrate” and “Genotype” effects on the survival rate.

| Factor                  | S.S  | D.L | M.S  | F    | p     |
|-------------------------|------|-----|------|------|-------|
| Substrate               | 1222.9 | 5 | 244.6 | 8.386 | 0.0001 |
| Genotype                | 17.4 | 1   | 17.4 | 0.595 | 0.447 |
| Substrate×Genotype      | 78.5 | 5   | 15.7 | 0.538 | 0.745 |
| Error                   | 700  | 24  | 29.2 |      |       |

SS: Sum of squares; D.L: Degree of Liberty; MS: Mean of squares; F: Fischer; p: Probability.

Table 3. Genotype survival rate (%).

| Substrate                     | Genotype survival rate (%) D9L7A1 | Genotype survival rate (%) 305 | Substrates survival rate average |
|-------------------------------|-----------------------------------|--------------------------------|----------------------------------|
| 100% Black soil               | 81.7±2.9                          | 76.7±7.6                        | 79.7±5.8ab                      |
| 50% Black soil + 50% Wood sawdust | 75±0                             | 71.7±2.9                        | 73.3±2.6bc                      |
| 100% Wood sawdust             | 83.3±2.9                          | 86.7±5.8                        | 85±4.5                         |
| 50% Black soil + 50% Black-coco | 71.7±5.8                          | 68.3±7.6                        | 70±6.3c                        |
| 100% Black-coco               | 68.3±2.9                          | 70±5                            | 69.2±3.8b                      |
| 50% Black coco + 50% Wood sawdust | 81.7±5.8                          | 80±8.6                          | 80.8±6.6ab                     |
| Substrates overall average    | 76.9±6.7                          | 75.6±8.6                        | 76.2±7.6                       |

*On the same line, data with the same letters are not significantly different at the 5% threshold. Newman-Keuls test.

Substrate and genotype effects on cuttings survival rate

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Substrate and genotype effects on the development of the kola tree root system

The substrate type and plant material used in the nursery cuttings of the kola tree has an impact on root development (Table 4). The analysis of variance highlighted the significant effect \( p=0.045; p=0.02 \) of these two factors on the number of roots formed. Indeed, except for the substrate 50% Black soil + 50% Wood sawdust with 1.9±1 roots formed (Table 5), all the other substrates favoured root production, notably the substrates 50% Black coco + 50% Wood sawdust, 100% Wood sawdust, 50% Black soil + 50% Black-coco, 100% Black-coco and 100% Black soil. It was also noted that genotype D9L7A1 produces more root than genotype 305 with 3.6±2.5 versus 2.6±1.7. The values of rooted cuttings and the largest taproots were obtained with substrates 50% Black coco + 50% Wood sawdust and 100% Black soil for the D9L7A1 clone, however no significant impact of factors on the number of rooted cuttings and taproot length could be demonstrated during this test.

Substrate and genotype effects on the development of the kola tree's aerial system (collar diameter, height and number of new leaves)

Rapid growth of height cuttings depends on the selected growing medium \( p=0.001 \) (Table 6). The best height growths (Table 7) were obtained with 100% Black soil and 100% Wood sawdust substrates with an average height of 14.7±4.2 and 13.4±3.6 cm. The growth of cuttings seems difficult on the 100% Black-coco (S5) medium. No significant genotype impact \( p=0.664 \) was found for this parameter.
Table 4. Substrate and genotype effects on the development of the kola tree root system.

| Genotype | Substrates                                      | Average number of rooted cuttings | Average length of tap-root (cm) | Average number of roots (cm) |
|----------|-------------------------------------------------|-----------------------------------|---------------------------------|-------------------------------|
| D9L7A1   | 100% Black soil                                | 15.3±2.08                         | 20.5±6.6                        | 3.6±3                         |
|          | 50% Black soil + 50% Wood sawdust              | 15±0                              | 14.5±5.4                        | 1.9±1.2                       |
|          | 100% Wood sawdust                              | 13.3±3.5                          | 17.9±7                          | 4±2.6                         |
|          | 50% Black soil + 50% Black-coco                | 14.3±1.1                          | 19.9±7                          | 4±2.2                         |
|          | 100% Black-coco                                | 10.3±2.5                          | 19.4±12.3                       | 3.2±2.6                        |
|          | 50% Black coco + 50% Wood sawdust              | 15.7±2.3                          | 21.5±8                          | 4.7±2.8                        |
|          | Total average                                  | 14±2.6                            | 18.9±8                          | 3.6±2.5^a                    |
| 305      | 100% Black soil                                | 10±6.2                            | 14.8±8.6                        | 2±1                           |
|          | 50% Black soil + 50% Wood sawdust              | 11.7±6.3                          | 17±8.4                          | 2±1                           |
|          | 100% Wood sawdust                              | 15±3.6                            | 12.9±5.8                        | 3.3±2.8                        |
|          | 50% Black soil + 50% Black-coco                | 9±2                               | 12.9±10.1                       | 2.1±0.9                       |
|          | 100% Black-coco                                | 10.3±7.1                          | 18.7±13.5                       | 2.4±2.3                        |
|          | 50% Black coco + 50% Wood sawdust              | 11.7±5.5                          | 19.1±12.9                       | 3.7±2.1                        |
|          | Total average                                  | 11.3±4.9                          | 15.9±10                         | 2.6±1.7^b                     |

Table 5. Roots average number per substrate.

| Substrate                                      | Average number of roots |
|-----------------------------------------------|-------------------------|
| 100% Black soil                               | 2.8±2.3^a               |
| 50% Black soil + 50% Wood sawdust             | 1.9±1^b                 |
| 100% Wood sawdust                             | 3.7±2.6^a               |
| 50% Black soil + 50% Black-coco               | 3.06±1.9^ab             |
| 100% Black-coco                               | 2.83±2.4^ab             |
| 50% Black coco + 50% Wood sawdust             | 4.17±2.1^a              |

For the production of new leaves, the type of substrate and genotype used for cutting the kola tree are determining factors (p= 0.026; p=0.002) (Table 6). Genotype D9L7A1 produces more leaves than genotype 305 with an average of 2.6±2.4 leaves versus 1.37±1.8. This production is more abundant in a substrate composed of 100% Black soil with 2.8±2.07 leaves.

Of all the parameters evaluated for the kola tree’s aerial system, ANOVA revealed that the diameter of the collar is a function of only the selected plant material (p=0.01).

Substrate and genotype effects on the aerial and root dry biomass of the kola tree

The impact on the aerial and root dry biomass of the substrate used and the genotype was assessed. No significant effects for root dry biomass were observed, unlike aerial dry biomass (Tables 8 and 9). For this parameter, the impact of substrate (p=0.022) (Table 8) and plant material (p=0.005) (Table 8) was noted.

Genotype D9L7A1 produces more aerial biomass than genotype 305 and this production is significant on a substrate consisting of 100% Black soil (Table 9).

A dendrogram (Figure 2) has made it possible to classify the different substrates used for tunnel cutting of the kola tree into two main classes which meet at an aggregation distance according to their efficiency on all growth parameters of nursery cuttings. The first high class mainly includes substrates 50% Black coconut + 50% Wood sawdust, 100% Black soil and 100% Wood sawdust. This first class includes 100% Black soil.
Table 6. Analysis of Variance (ANOVA) of the "Substrate" and "Genotype" effects on the aerial growth parameters of the Kola tree.

| Variable                  | Factors                  | S.S   | D.L | M.S   | F      | p     |
|---------------------------|--------------------------|-------|-----|-------|--------|-------|
| Height of the plant       | Substrate                | 218.05| 5   | 43.61 | 4.189  | 0.001 |
|                           | Genotype                 | 1.97  | 1   | 1.97  | 0.190  | 0.664 |
|                           | Genotype×Clone           | 44.00 | 5   | 8.80  | 0.845  | 0.521 |
|                           | Error                    | 999.42| 96  | 10.41 |        |       |
| Number of new leaves      | Substrate                | 56.5974| 5   | 11.319| 2.6720 | 0.026 |
|                           | Genotype                 | 41.7526| 1   | 41.752| 9.8560 | 0.002 |
|                           | Genotype×Clone           | 6.5864| 5   | 1.317 | 0.3110 | 0.905 |
|                           | Error                    | 402.4444| 95  | 4.236 |        |       |
| Diameter at the collar    | Substrate                | 0.036 | 5   | 0.007 | 0.733  | 0.6   |
|                           | Genotype                 | 0.067 | 1   | 0.067 | 6.853  | 0.01  |
|                           | Genotype×Clone           | 0.028 | 5   | 0.0056| 0.575  | 0.718 |
|                           | Error                    | 0.945 | 96  | 0.0098|        |       |

SS: Sum of squares; D.L: Degree of Liberty; MS: Mean of squares; F: Fischer; p: Probability.

Table 7. Analysis of data on the height and number of new leaves of Kola tree cuttings

| Substrate                          | Average height per plant (cm) | Average plant height per substrate (cm) |
|------------------------------------|-------------------------------|----------------------------------------|
|                                    | D9L7A1                        | 305                                    |
| 100% Black soil                    | 14.4±3.3                      | 15.02±5.3                              |
| 50% Black soil + 50% Wood sawdust  | 11.63±3.8                     | 13.3±3.9                               |
| 100% Wood sawdust                  | 13.86±4.2                     | 12.94±3.2                              |
| 50% Black soil + 50% Black-coco    | 12.79±2.6                     | 10.38±1.5                              |
| 100% Black-coco                    | 10.7±2.04                     | 10.06±1.42                             |
| 50% Black coco + 50% Wood sawdust  | 11.39±2.04                    | 11.48±3.2                              |
| Overall average                    | 12.47±3.22                    | 12.2±3.6                               |

| Substrate                          | Average number of new leaves per substrate | Average number of new leaves | Average number of new leaves per substrate |
|------------------------------------|---------------------------------------------|-------------------------------|---------------------------------------------|
|                                    | D9L7A1                        | 305                                    |
| 100% Black soil                    | 3.2±2.17                      | 2.3±2                          | 2.8±2.07*                              |
| 50% Black soil + 50% Wood sawdust  | 2.7±2.87                      | 1.8±1.92                      | 2.2±2.4a                               |
| 100% Wood sawdust                  | 3.5±2.77                      | 1.56±1.94                     | 2.47±2.5ab                             |
| 50% Black soil + 50% Black-coco    | 3.2±2.22                      | 1.56±2                        | 2.39±2.2ab                             |
| 100% Black-coco                    | 1±1.32                       | 0.44±1                        | 0.72±1.17ab                            |
| 50% Black coco + 50% Wood sawdust  | 2.1±2.47                      | 0.56±1.13                     | 1.33±2.03ab                            |
| Overall average                    | 2.6±2.4a                      | 1.37±1.8b                     | 1.98±2.2                               |

*On the same line, data with the same letters are not significantly different at the 5% threshold. Newman-Keuls test.

substrate, which has had a positive effect on aerial biomass, survival rate, height growth, number of roots and new leaves formed. It can be qualified as a class of substrates that are effective on most of the growth parameters of the kola tree. The second major class is made up of substrates 50% Black soil + 50% Wood sawdust, 100% Black-coco and 50% Black soil + 50% Black-coco can be considered as the class of substrates less effective on cuttings.

The first large class joins the second class at an aggregation distance of 6.6.

DISCUSSION

The tunnel cutting trial of the kola tree in nursery resulted in high survival rates of cuttings with an average of 76.2±7.6%. This is the first time that such a survival
Table 8. ANOVA of dry biomass data.

| Variable                  | S.S  | D.L | M.S  | F    | p     |
|---------------------------|------|-----|------|------|-------|
| Root dry matter           |      |     |      |      |       |
| Substrate                 | 0.2  | 5   | 0.04 | 0.948| 0.453 |
| Genotype                  | 0.007| 1   | 0.007| 0.162| 0.68  |
| Substrate × Genotype      | 0.144| 5   | 0.029| 0.683| 0.636 |
| Error                     | 4.05 | 96  | 0.042|      |       |
| Aerial dry matter         |      |     |      |      |       |
| Substrate                 | 5.307| 5   | 1.061| 2.757| 0.022 |
| Genotype                  | 3.146| 1   | 3.146| 8.173| 0.005 |
| Substrate × Genotype      | 0.736| 5   | 0.147| 0.382| 0.859 |
| Error                     | 36.954| 96 | 0.385|      |       |

SS: Sum of squares; D.L: Degree of Liberty; MS: Mean of squares; F: Fischer; p: Probability.

Table 9. Analysis of data on aerial and root dry biomass.

| Substrate                     | Average root dry matter (g) | Average root dry matter per substrate (g) |
|-------------------------------|-----------------------------|-------------------------------------------|
|                               | D9L7A1                      | 305                                       |
| 100% Black soil               | 0.4±0.12                    | 0.46±0.28                                 |
| 50% Black soil + 50% Wood sawdust | 0.26±0.16                  | 0.4±0.24                                 |
| 100% Wood sawdust             | 0.32±0.13                   | 0.26±0.1                                 |
| 50% Black soil + 50% Black-coco | 0.36±0.2                   | 0.3±0.22                                 |
| 100% Black-coco               | 0.33±0.24                   | 0.38±0.25                                |
| 50% Black coco + 50% Wood sawdust | 0.4±0.25                   | 0.36±0.12                                |
| Overall average               | 0.34±0.18                   | 0.36±0.21                                |

| Substrate                     | Average aerial dry matter (g) | Average aerial dry matter per substrate (g) |
|-------------------------------|-------------------------------|---------------------------------------------|
|                               | D9L7A1                       | 305                                         |
| 100% Black soil               | 2.09±0.68                    | 1.83±0.44                                 |
| 50% Black soil + 50% Wood sawdust | 1.8±0.58                   | 1.6±0.54                                 |
| 100% Wood sawdust             | 1.92±0.85                    | 1.53±0.64                                 |
| 50% Black soil + 50% Black-coco | 1.99±0.78                   | 1.3±0.51                                  |
| 100% Black-coco               | 1.46±0.62                    | 1.15±0.52                                 |
| 50% Black coco + 50% Wood sawdust | 1.5±0.48                  | 1.27±0.62                                 |
| Overall average               | 1.8±0.69ab                   | 1.45±0.57ab                               |

*On the same line, data with the same letters are not significantly different at the 5% threshold. Newman-Keuls test.

A (success) rate has been obtained for the kola tree, particularly for the species C. nitida. This high success rate may be due, on one hand, to the favourable period in which the cuttings were made. Indeed, the rainy season period (April to October) is the favourable period for the cutting of tropical species such as the kola tree, unlike the dry season (Ricez, 2008). On the other hand, the method of dressing adopted (cutting with four leaves cut in half) and the type of cutting adopted (tunnel cutting) are known to improve success rates. This result is in agreement with Wolff's (1999) work on Eucalyptus gunnii, which showed that with controlled temperature and humidity, the tunnel allows good viability of the cuttings. In addition, this type of tunnel growing has also been successfully used for the cutting of cocoa tree, a Malvaceous species (Theobroma cacao) (Koko et al., 2011). Unlike the substrate, no effect of genotype and "Substrate × Genotype" interaction on survival was found. Soil conditions, the culture medium and its water storage capacity play an important role in the development of plants, including kola trees (Haferkamp, 1988; Xie et al., 2016). The best success rates for cuttings from the kola tree were obtained with substrates 100% Wood sawdust, 50% Black coco + 50% Wood sawdust and 100% Black soil. These substrates have a sufficient water storage capacity unlike substrates with 50% Black soil + 50% Black-coco, 50% Black soil + 50% Wood sawdust and 100% Black-coco where mortality rates are high. The high storage capacity of these substrates could be the cause of the observed mortality cases. They retain at
least 40% of the water used for watering, the high water content of the substrate can cause an oxygen deficit in the substrate that is harmful to the growth of plants and cuttings in particular (Felix et al., 2007). The black-coco substrate retains less water (33%) compared to other substrates. The retention capacity of the substrate favourable to the kola tree following this test must be between 360 and 420 ml/L. A storage capacity that is not or too high, for example 800 ml/L for 50% Black soil + 50% Wood sawdust, has a negative effect on the root development of cuttings. Indeed, apart from the substrate 50% Black soil + 50% Wood sawdust all the other substrates favoured root production, notably the substrates 50% Black coco + 50% Wood sawdust, 100% Wood sawdust, 50% Black soil + 50% Black-coco, 100% Black-coco and 100% Black soil. These substrates are porous and well drained, which is a beneficial characteristic for rooting cuttings (Koyama et al., 2014).

The substrates 100% Black soil, 50% Black coconut + 50% Wood sawdust, and 100% Wood sawdust gave the best results on growth parameters especially the substrates 100% Black soil and 100% Wood sawdust according to the dendrogram. These substrates had a positive effect on aerial dry biomass (respectively 1.96±0.57; 1.73±0.76), height growth (respectively 14.7±4.2; 13.4±3.6), number of roots (respectively 2.8±2.3; 3.7±2.6) and neoformed leaves (respectively 2.8±2.07; 2.47±2.5). Apart from the porosity and good drainage of these substrates, black soil also had mineral element content necessary for mineral nutrition during plant growth, unlike sawdust. According to Van Cleve et al. (1991), the amount of biomass produced depends on nutrient absorption of culture medium. Insufficiently ground sawdust releases fewer nutrients (Sie et al., 2008). However, these results must be qualified with regard to the production of new leaves, roots and above-ground biomass, which are also due to the chosen kola tree genotype. Many researchers studied the influence of genotype on biomass and plant growth (nutrient use efficient genotypes) and found impressive results (Van Cleve et al., 1991). It appears from this study that genotype D9L7A1 has a higher vegetative development than clone 305.

**Conclusion**

The main objective of this study was to develop a method...
CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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