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# CURING-BASED MINI-NUT PROPAGATION TECHNIQUE OF KOLA (*Cola nitida*, Vent. Schott & Endlicher.): AN INNOVATIVE APPROACH FOR GERMINATION AND SEEDLING GROWTH

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## ABSTRACT

Limited information on science and technical knowledge of raising *Cola nitida* seedlings in the nursery hinders kolanut farmers to overcome challenges post-harvest dormancy. Experiment was conducted in a screen house at Federal University of Agriculture, Abeokuta, Nigeria to evaluate the effects of mini-nut techniques, curing in pyramidal device and palm basket on early seed germination and vigorous seedling growth of *Cola nitida*. Treatments consisted of mini-nut techniques (12.5% embryonic), 25 50, 75 and 100% Cotyledon Portion (CP) and curing media (Pyramid Device (PD), Palm Basket (PB) and PB+PD)). Experiment was 5 x 3 factorial layout in Completely Randomized Design, replicated 3 times. Data collected on seed germination percentage, growth and dry matter production were subjected to analysis of variance and mean comparisons done using DMRT ($P \leq 0.05$). Germination commenced at 2 WAS (Weeks After Sowing), in nuts with 25% CP in PD, with 10.0% germination. Other treatments were between 0 and 3.33%. At 8 WAS, nuts with 25% and 50% CP in PD had 100% germination while nuts with 12.5 – 50% PB and PB + PD had 80% germination. Within the same period, nuts with 25% CP in PD produced the tallest plant (34.1cm). Also, 75% CP in PD had significantly wider leaf area (49.1cm$^2$) than those in PB and PB+PD. Nuts with 25, 50 and 75% CP in PD and PB+PD had rapid and uniform germination of *Cola nitida*, and vigorous seedling obtained within the period of 8 weeks after sowing, in 25 and 75% CP in PD.

**Keywords:** *Cola nitida;* germination; curing media; mini-nuts; pyramidal device; growth

## INTRODUCTION

*Cola nitida* (Vent. Schott and Endlicher) is commonly called Kolanut, The fruit of kola tree, an evergreen tree crop in the genus *Cola* and the family *Sterculiaceae* (Okigbo and Ndubis, 2008). The genus kola consists of about 100-125 Cola species in West Africa. However, *Cola nitida* and *Cola acuminata* are species of real economic importance (Quarcoo, 1973; Daramola, 1978). The trees, which reach heights of 20 to 25 meters, produce a star-shaped fruit. Each fruit contains between two and five kolanuts, about the size of a chestnut. Botanically, kola fruit is a follicle of five to six pods, each pod containing 6 –10 kolanuts (Van Eijnatten, 1966).

The nut possesses a bud at an extreme end with varying sizes of cotyledon which is fresh to touch. Propagation of plants involves the use of the nut (Oladokun, 1988; Olaiya and Hammed, 2003; Opeke, 2005) which occurs in three distinct colored biotypes: white, pink and red. The red-biotype is predominant, while white-biotype is rarest (Ogutuga, 1975; Oladokun, 1988). Kolanut trees are mainly grown between 6 and $7^0$ North of the equator but have also been found up to $10^0$ N on the West Coast of Africa. It thrives in the hot tropical lowland forest with rainfall of 17,000 mm extending over a period of 8 months or with monthly rainfall of about 1,200 mm and a temperature of between 23 and $28^0$C (Ekanade, 1989). The species have also been cultivated in the transitional zones in savannas (Opeke, 1992). The species could be cultivated in drier areas where growth could only be made possible by the occurrence of wet land with a high water-table (Russel, 1955). The species flourishes in both heavy and light soil if they have good depth. Better girth and fruiting are obtained in well-drained, fertile soils containing much humus. According to FAOSTAT (2022), Nigeria produces 174.11 tonne (55.27%) followed by Cote D'Ivoire 58.64 tonne (18.61%), Cameroon 48.57 tonne (15.42%), Ghana 24.64 tonne (7.96%), Sierra Leone 8.45 tonne (2.68%) and Republic of Benin 611.26 tonne (0.19%) of the total world kolanut production of 315,024 tonne.

It is an important commodity crop in West Africa and other tropical regions of the World (Famaye, 2012). Also, native to tropical Africa with its centre of diversity in West Africa especially Cote d'Ivoire and Ghana (Opeke, 2005). The kolanut is a culturally significant staple in many West African countries, valued for its stimulant effects on the central nervous system. In various markets, bus depots and corner shops throughout the region, small piles of kolanuts are commonly available for sale. This nut represents an important cash crop for many rural farmers and numerous individuals consume it daily for an energy boost. Each nut contains more caffeine than two large cups of American coffee (Adeyemi and Adebisi, 2017). In Western countries, such as the United States of America and Europe, kolanut extract is more prevalent than the fresh nuts themselves. This extract serves as a popular food flavoring in beverages like Coca-Cola, Pepsi-Cola and a variety of energy drinks (Gonzalez *et al.,* 2020). The U.S. Food and Drug Administration (FDA) has classified kolanut as generally recognized safe (GRAS) for human consumption; kola extract is categorized as a natural food flavoring. The FDA has approved kola extract for use as an inactive ingredient in certain pharmaceutical products (FDA, 2018). Historically, kola extract has been utilized in some weight loss medications and over-the-counter stimulants. Kolanut extract is marketed as an herbal supplement, though these products are usually not regulated by the FDA. Warnings regarding caffeine content are often included on packaging. The American Herbal Products Association advised that kolanut should be avoided by pregnant or breastfeeding women, as well as individuals under the age of 18, due to its caffeine content (AHPA, 2021).

Kolanut seeds exhibit long post-harvest dormancy periods of between 1 and 8 months which need to be removed (Oladokun, 1988). This suspension of germination has been attributed to the physiological status of the embryo, high contents of caffeine and polyphenols (Elias and Bressani, 1979; Oladokun, 1985). This phenomenon was explained by the fact that the embryos of stored seeds were much more developed morpho-genetically than the fresh seeds (Van Eijnatten, 1966). Further investigations carried out by Van Eijnatten (1968) further buttressed this finding that immature seeds germinated poorer than the mature ones. Polyphenols have been reported to make amino acids unavailable by binding strongly to them (Elias and Bressani, 1979), while caffeine is believed to delay kolanut germination until its hydrolysis begins (Hammed *et al.*, 2013; Oladokun, 2000). Mobilization of nitrogen reserves as a source of energy and nutrient supply to the expanding new tissues is therefore proposed to take part in the control of germination (Garciarrubio *et al.*, 1997). This is because germination of seeds accompanies metabolism of food reserves in the cotyledons (Onomo *et al.*, 2010). Cotyledons of *Cola nitida* seeds, besides being a reservoir of nutrient reserves (Oladokun, 1989; Hammed *et al.*, 2013), contain high number of polyphenols and caffeine; these chemicals are more abundant in red-coloured biotype nuts (Ogutuga, 1975).

Curing is the post-harvest treatment of kolanuts after removing the testa, which involves wrapping freshly harvested nuts with banana leaves and placing them in a woven basket. Curing, before sowing reduces long dormancy period and helps in the mobilization of nutrients in which achieving early germination and even germination of the seed, their understanding of the germination process is of important ecological and agronomical relevance. Curing seeds in a pyramidal shape has been investigated to enhance not only germination but also post-germination growth vigour of crop seedlings and crop yield in some arable crops. Such investigation has not been carried out in tree crops, kola inclusive. Exposure of the internal tissue of the *Cola nitida* seeds cotyledons (as obtained in mini-nut technology), to the pyramidal effects would provide direct effects of the pyramidal energy into the seeds. Such treatments of *Cola nitida* seeds could provide an intervention technology in the series of cultivation limitations bedeviling kola production in the world. This may, therefore, positively influence seed germination, seedling growth and field establishment successes, especially at the early period, better than the previous studies. As part of the effort to increase productivity and overall production, various practices that would enhance the establishment, growth and productivity of crops deserve attention. This experiment was thus set up to evaluate the mini-nut technique, curing method of Pyramidal Device (PD) and Palm Basket (PB) and determine the effects on kola-nut early seed germination and improved morphological growth in the nursery.

## MATERIALS AND METHODS

### *Experimental location and sample source*

The study was carried out at the Federal University of Agriculture Abeokuta Ogun State, Nigeria (Latitude 7$^0$ 15' N Longitude 3$^0$ 25' E). Matured pod of the kolanut tree crop were collected from the field plots of Kola Association of Nigeria (KOLAN) farms located in Okuku, Odo Otin Local Government Area, Osun State, Nigeria.

### *De-pulping process, sample preparation and sowing*

The pods were cut open to extract the nuts and depulped to remove the seed coat (kolanut testa). Seven hundred (700) healthy fresh red kolanut seeds, with average weight between 15 and 20 g were selected and used for the experiment. The sample kolanut seeds prepared as mentioned above were placed inside each of the medium and the plastic pots filled with partially decomposed sawdust. The plastic pots were perforated underneath for draining out excess water. The selected and treated kola seeds were sown horizontally inside the 50 cl plastic pots according to the treatment above, at one seed per pot, at a sowing depth of 5 cm and covered with thin layer of sawdust. Prior to sowing, the sawdust was watered to field capacity for two weeks, to allow for adequate moisturization of the media. Ten seeds were sown per treatment giving 150 seeds per replicate which amounted to 450 seeds for the whole experiment. Watering was carried out daily using 50 ml / pot in split application, early in the morning and evening to prevent the plant from water stress.

***Storage medium condition***

Temperature and relative humidity of the curing media/device were monitored with the aid of thermometer and hygrometer for twelve weeks consecutively (Table 1). Temperature and relative humidity data at different time points in the curing media (Plate 1) and laboratory room (Palm Basket, Pyramidal Device, Lab Room) during a period of 12 weeks were observed (Table 1). Data were collected in relation to the curing and sowing of kolanut (*Cola nitida*), with a special note indicating that the measurements at week 0 were taken two weeks before this process began. The first three columns under each of the medium represent the temperature in Degrees Celsius at various weeks. The next three columns under each location represent the relative humidity percentage at the corresponding weeks. The asterisk * at week 0 indicates that these measurements were taken two weeks before the curing and sowing of kolanut (*Cola nitida*) began. This may suggest a baseline or initial environmental condition before the specific agricultural activities commence.

**Plate 1: Media used for curing kola seeds**

(a) Palm Basket

(b) Pyramidal Device

***Experimental design and treatments***

The experiment was a 3 x 5 factorial layout in a completely randomized design with three replicates. Factor 1 was curing-media: Pyramidal Device (PD), Palm Basket (PB) and combination of Pyramidal Device plus Palm Basket (PD+PB). Factor 2 was mini-nut treatment, which consisted of 12.5, 25, 50, 75, 100% cotyledon portion (CP), giving 15 treatments combinations. (Table 2). Dried palm baskets were procured, and pyramidal devices were made with the use of plywood (as described by Adetiloye, 1996), with the edge length of 11.43 cm having an angle of

$108^0$, and the sample seeds/nuts for mini-nut treatment were prepared in accordance with the reports of (Hammed *et al.*, 2019). The volume and surface area of Pyramidal Device (PD) used for the study were determined using the geometry formula derived by Mark-Boster (2022).

$$V = \frac{(15 + 7\sqrt{5})}{4} e^{3} \qquad A = 3\sqrt{25 + 10\sqrt{5}}\; e^{2}$$

**e** = Length of an edge

**Table 1: Temperature ($^0$C) and Relative humidity (%) in the curing media (Palm-basket and Pyramidal Device) and Laboratory room before sowing of *Cola nitida* nut recorded fortnightly for 12 weeks**

| Temperature ($^0$C) | | | | Relative humidity (%) | | |
|---|---|---|---|---|---|---|
| Weeks | Palm Basket | Pyramidal Device | Lab Room | Palm Basket | Pyramidal Device | Lab Room |
| 0* | 28.8 | 27.7 | 29.2 | 76 | 78 | 79 |
| 2 | 28.7 | 28.5 | 28.9 | 78 | 79 | 80 |
| 4 | 28.8 | 28.4 | 29.2 | 79 | 78 | 79 |
| 6 | 28.3 | 28.4 | 29.3 | 79 | 78 | 81 |
| 8 | 29.1 | 29.5 | 29.5 | 80 | 79 | 82 |
| 10 | 29.0 | 28.9 | 28.9 | 81 | 80 | 83 |
| 12 | 28.9 | 28.7 | 29.0 | 80 | 79 | 81 |

*Two weeks before curing and sowing of kolanut (*Cola nitida*)

**Table 2: Treatment definition**

| Treatment | Definition |
|---|---|
| 12.5% CP in PD | 12.5% embryonic portion cured in a pyramidal device |
| 25% CP in PD | 25% cotyledon portion cured in a pyramidal device |
| 50% CP in PD | 50% cotyledon portion cured in a pyramidal device |
| 75% CP in PD | 75% cotyledon portion cured in a pyramidal device |
| 100% CP in PD | 100% cotyledon portion cured in a pyramidal device |
| 12.5% CP in PB | 12.5% embryonic portion cured in a palm basket |
| 25% CP in PB | 25% cotyledon portion cured in a palm basket |
| 50% CP in PB | 50% cotyledon portion cured in a palm basket |
| 75% CP in PB | 75% cotyledon portion cured in a palm basket |
| 100% CP in PB | 100% cotyledon portion cured in a palm basket |
| 12.5% CP in PB + PD | 12.5% embryonic portion cured in a combination of palm basket plus pyramid |
| 25% CP in PB + PD | 25% cotyledon portion cured in a combination of palm basket plus pyramid |
| 50% CP in PB + PD | 50% cotyledon portion cured in a combination of palm basket plus pyramid |
| 75% CP in PB + PD | 75% cotyledon portion cured in a combination of palm basket plus pyramid |
| 100% CP in PB + PD | 100% cotyledon portion cured in a combination of palm basket plus pyramid |

**Note:** Total curing duration was kept constant for all treatments to allow comparison with another single stage. **PD =** Pyramid device – cotyledons cured in pyramid-shape curing device; **PB** = Palm basket – cotyledons cured in traditional woven palm basket; **PB+PD** = Sequential curing – initial curing in palm basket (PB), then transferred to pyramid device (PD) to complete curing.

***Data collection***

Data were collected on seed germination at two weekly observations for emergence of plumule and minimum of two leaves as an index of germination (ISTA, 2004) until no further germination was observed. At 8 WAS, observation of growth (Plant height (cm), number of leaves/plants, stem girth (mm), and leaf area ($cm^2$) of seedlings began at the nursery. Dry matter production was determined from 14 to 20 WAS.

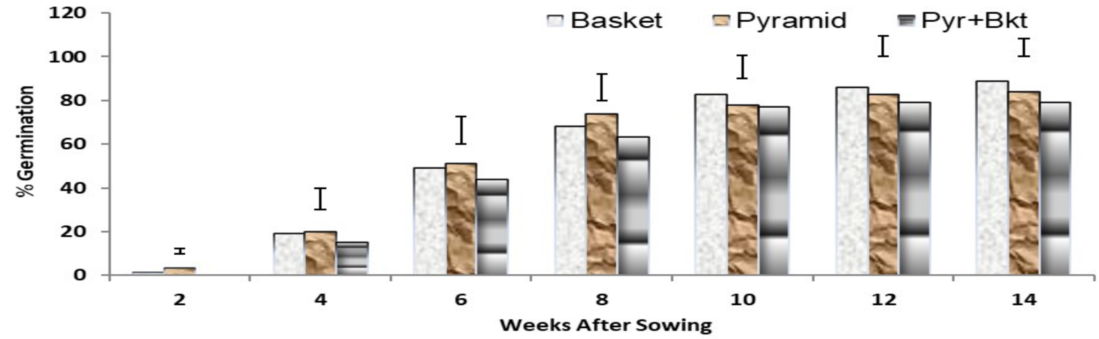
***Data analysis***

All the data collected were subjected to analysis of variance (ANOVA) procedures using Genstat 4.10.3 software (Genstat, 2019), and the treatment means were compared using Duncan's Multiple Range Test (DMRT), at the 5% level of probability ($p \leq 0.05$).

## RESULTS

Seed germination commenced at 2 WAS from nuts cured in PD with 3.33% germination percentage followed by nuts cured in PB (1.33%) and PB+PD with zero percentage germination (Figure 1). At 6 WAS nuts cured in PD had more than 50% germination while PB and PB+PD had 49.3% and 44.0% germination %, respectively. At 8 WAS, nuts cured in PD recorded the highest germination % with 74% followed by PB and PB+PD with 68% and 63.3% in that order. At 10 WAS, nuts cured in PB, PD and PB+PD had 82.7%, 78% and 77.3%, respectively, germinated (Figure 1a). Mini-nut seeds treatment germination was recorded at 2 WAS, with 25% cotyledon portion (3.33%), 12.5% CP (2.22%), 50% CP and whole nut with 1.11% germination each, while 75% CP were yet to germinate (Figure 1a). At 6 WAS, more than 50% germination was recorded for 25, 12.5, and 50% CP with 66.7%, 57.8% and 58.9% respectively, while 75% CP and Whole nuts had between 44.4 and 13.3% germination percentage; the differences were significant. At 10 WAS, 50% CP had 93.3% germination closely followed by 25% CP with 92.2% germination, while other treatments recorded between 58.9% and 72.2% (Figure 1b). With respect to the interactions, at 2 WAS, 12.5% cotyledon portion cured in PD had germinated 10.0%, whole nuts cured in PD, 12.5% cotyledon portion cured in PD, 50% cotyledon portion cured in PB, and 12.5% cotyledon portion cured in PB had 3.3% each, while none of the PB+PD treatments had germinated. At 6 WAS, more than 50% germination were observed in some treatments: 25%, 50% CP in PD, embryonic 12.5%, 25%, 50% CP in PB and 12.5%, 25% CP in PB+PD. At 8 WAS, 25% and 50% CP in PD had attained 100% percentage germination, while other treatments ranged between 33.3% and 80%, and at 10 WAS, similar trend was observed (Table 3).

**(a)**



**(b)**

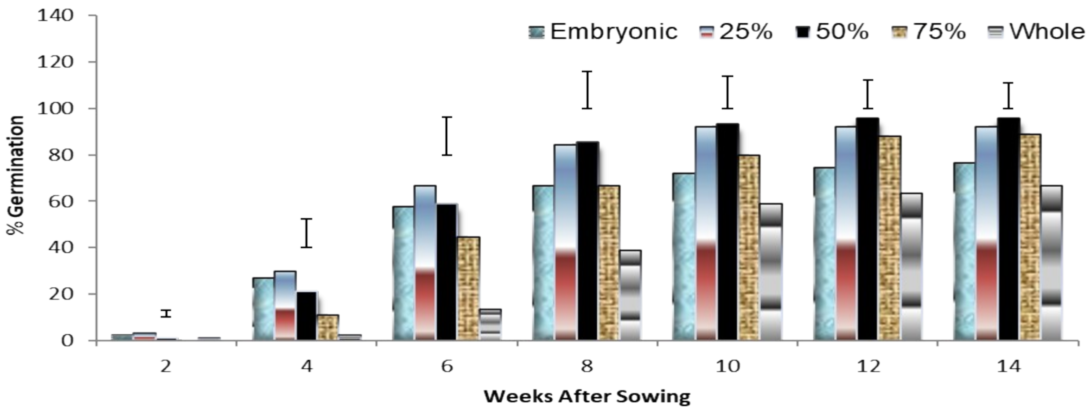



**Figure 1**: Percentage germination of kolanut seeds (*Cola nitida*) as affected by: (a) curing media; (b) mini-nut technique at 2-14 WAS.

Curing medium had no influence on the height of the seedlings. Height of the seedlings raised from nuts treated with curing in PD, PB, and PB+PD were between 17.8 and 20.4 cm at 8 WAS, 21.6 and 22.6 cm at 10 WAS, 24.9 and 25.4 cm at 12 WAS, 25.5 and 27.3 cm at 14 WAS (Figure 2a). However, mini-nut treatment influenced the height of the seedlings. At 8 WAS, seedlings raised from the nuts treated with 25% cotyledon portion had significantly tallest seedlings (26.6 cm) compared to other treatments that ranged between 13.5 and 20.9 cm; the differences were significant. At 12 and 14 WAS, nuts treated with 25% cotyledon portion had a reduced plant height (27.2 and 26.6 cm) compared to 75% cotyledon portion (27.9 and 29.9 cm), while other treatments ranged between 19.9 and 25.6 cm and between 19.5 and 28.6 cm, respectively (Figure 2b). Interactions showed that seedlings raised from nuts treated to 25% cotyledon portion cured in pyramidal device had significantly tallest seedlings compared with other treatments that ranged between 12.6 and 24.9 cm at 8 WAS. Same trend was observed at 10, 12 and 14 WAS, where 25% cotyledon portion of the nuts cured in PD had highest plant height values of 34.6, 5.4 and 35.9 cm, respectively compared to other treatments relative to each period (Table 4).

***Total Leaf Area (cm²)/plant***

The differences in the leaf area of the seedlings derived from the three-curing media (PB, PD and PB+PD) were not significant throughout the period from 8 to 14 WAS (Figure 3a). Leaf area of the seedlings raised from nuts treated with storage in PB, PD and PB+PD recorded 51.8 $cm^2$, 52.7 $cm^2$ and 52.4 $cm^2$, respectively at 14 WAS. However, at 8 WAS, development of leaf area of seedlings raised from nuts of 25% and 75% portion of cotyledon were highest, with 38.4 $cm^2$ and 39.6 $cm^2$, respectively. These were significantly higher than 50% portion of cotyledon and 100% whole nuts with leaf area of 28.2 $cm^2$ and 25.1 $cm^2$ in that order, while 12.5% portion of cotyledon recorded significantly reduced leaf area of 17.5 $cm^2$, the differences were significant. At 10 WAS, seedlings raised from nuts treated to 75% portion of cotyledon had the highest (53.5 $cm^2$) leaf area production, followed by 25% portion of cotyledon (43.9

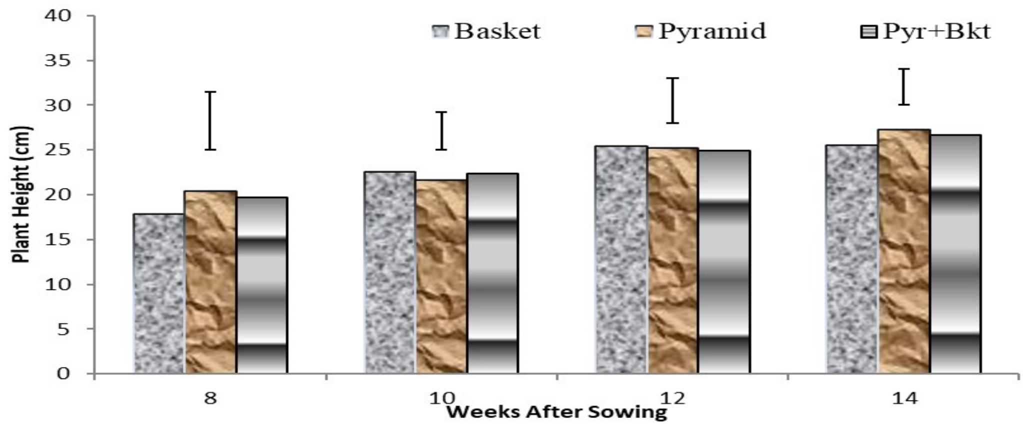
**Table 3: Seed germination (%) of kola (Cola nitida) as affected by curing medium and mini-nut technique in the Nursery.**

| Treatment | | Weeks After Sowing (WAS) | | | | | | |
|---|---|---|---|---|---|---|---|---|
| Curing medium | Mini-nut Size | 2 | 4 | 6 | 8 | 10 | 12 | 14 |
| | 12.5% Embryonic | 3.33b | 33.3a | 70.0ab | 80.0ab | 83.3abc | 83.3abc | 90.00abc |
| | 25% cotyledon | 0.00b | 36.7a | 66.7ab | 80.0ab | 90.0ab | 90.00ab | 90.00abc |
| PB | 50% cotyledon | 3.33b | 23.3abc | 56.7ab | 76.7ab | 90.0ab | 93.3ab | 93.3ab |
| | 75% cotyledon | 0.00b | 3.33bc | 40.0bcde | 60.0bcde | 80.0abcd | 90.0ab | 93.3ab |
| | 100% (Whole nut) | 0.00b | 0.00c | 13.3de | 43.3cde | 70.0bcde | 73.3bcd | 76.7bcd |
| | 12.5% Embryonic | 3.33b | 26.7ab | 43.3bcd | 53.3bcde | 53.3dde | 56.7d | 56.7de |
| | 25% cotyledon | 10.00a | 36.7a | 80.00a | 100.0a | 100.0a | 100.0a | 100.0a |
| PD | 50% cotyledon | 0.00b | 20.0abc | 73.3ab | 100.0a | 100.0a | 100.0a | 100.0a |
| | 75% cotyledon | 0.00b | 13.3abc | 43.3bcd | 76.7ab | 80.0abcd | 93.3ab | 93.3ab |
| | 100% (Whole nut) | 3.33b | 6.67bc | 16.7cde | 40.0de | 56.7cde | 63.3cd | 70.0cde |
| | 12.5% Embryonic | 0.00b | 23.3abc | 60.0ab | 66.7bcd | 80.0abcd | 83.3abc | 83.3abc |
| | 25% cotyledon | 0.00b | 16.7abc | 53.3ab | 73.3abc | 86.7ab | 86.7abc | 86.7abc |
| PB+PD | 50% cotyledon | 0.00b | 20.0abc | 46.7abc | 80.0ab | 90.0ab | 93.3ab | 93.3ab |
| | 75% cotyledon | 0.00b | 16.7abc | 50.0ab | 63.3bcde | 80.00abcd | 80.00abc | 80.00abc |
| | 100% (Whole nut) | 0.00b | 0.00c | 10.00e | 33.3e | 50.0e | 53.3d | 53.3e |

Note: PB = Palm Basket, PD = Pyramidal Device, PB+PD = Combination of a Pyramidal Device and Palm Basket
Means following by same alphabets, same column, were not significantly different at $P \leq 0.05$.

cm²) and 50% portion of cotyledon with 41.2 cm²; the differences were not significant. These were, however, significantly higher compared to seedlings derived from 12.5% portion of cotyledon and 100% whole nuts with a reduced leaf area value of 22.4 cm² and 37.1 cm², respectively. At 12 WAS, leaf area of seedlings treated to 25%, 50%, 75% portion of cotyledon and 100% whole nut were not significantly different from each other (47.1 cm², 45.2 cm², 56.5 cm² and 46.0 cm²). At 14 WAS, seedlings derived from the nuts treated to 25%, 50%, and 75% portion of cotyledon had leaf area value of 92.2 cm², 95.6 cm², and 88.9 cm² respectively and were not significantly different from each other. (Figure 3b).

**a)**




**b)**

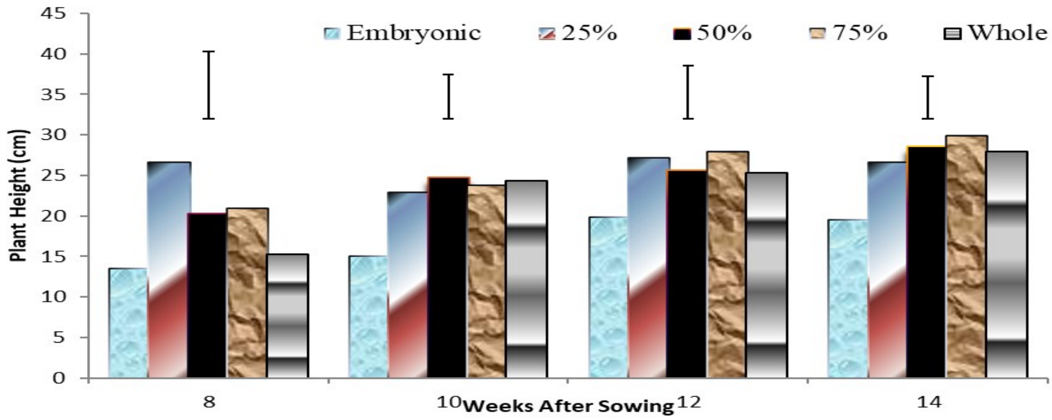
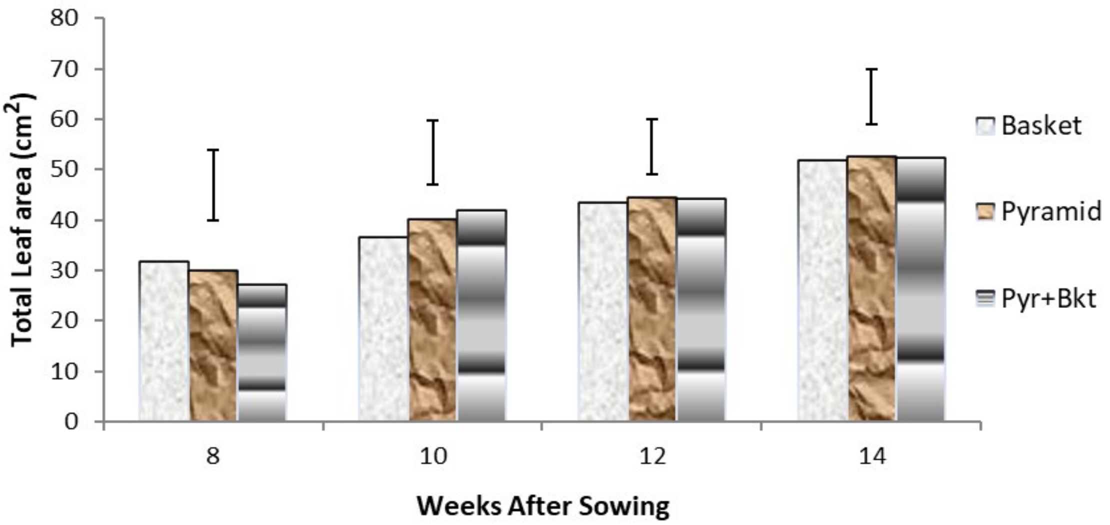



**Figure 2**: Height (cm) of kola seedlings as affected by: (a) curing media; (b) mini nut technique in the nursery.

**a)**

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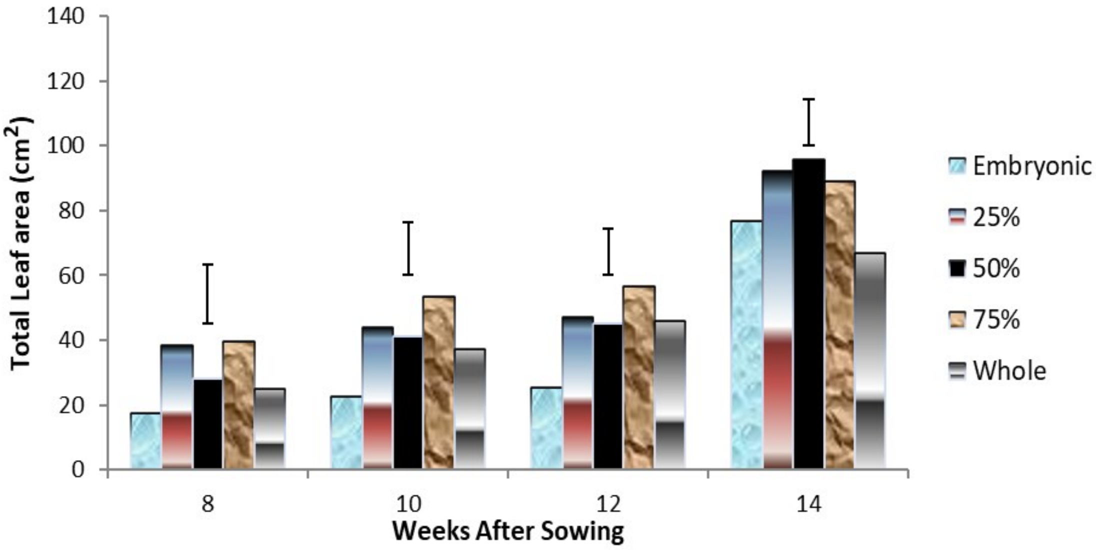



**Figure 3:** Total leaf area (cm²) per plant of kola (*Cola nitida*) as affected by: (a) curing Media; (b) mini-nut techniques at 8-14 weeks after sowing (WAS)

**Table 4: Plant height (cm) of kola seedlings as affected by curing media and mini- nut size in the Nursery**

| Treatment | | Weeks After Sowing | | | |
|---|---|---|---|---|---|
| Curing medium | Mini-nut Size | 8 | 10 | 12 | 14 |
| | 12.5% Embryonic | 12.6b | 14.8b | 23.3ab | 17.4c |
| | 25% cotyledon | 20.9ab | 22.1ab | 24.0ab | 25.5abc |
| PB | 50% cotyledon | 20.3ab | 23.1ab | 25.5ab | 26.2abc |
| | 75% cotyledon | 18.9ab | 22.3ab | 26.7ab | 28.2ab |
| | 100% (Whole nut) | 16.5b | 30.5a | 27.5ab | 30.2a |
| | 12.5% Embryonic | 13.2b | 14.1b | 17.2b | 19.3bc |
| | 25% cotyledon | 34.1a | 34.6a | 35.4a | 35.9a |
| PD | 50% cotyledon | 19.2ab | 23.7ab | 26.7ab | 32.2a |
| | 75% cotyledon | 21.7ab | 24.9ab | 27.0ab | 30.4a |
| | 100% (Whole nut) | 14.1b | 20.8ab | 24.7ab | 26.8abc |
| | 12.5% Embryonic | 14.7b | 16.2b | 19.0ab | 22.0abc |
| | 25% cotyledon | 24.9ab | 22.3ab | 26.4ab | 26.3abc |
| PB+PD | 50% cotyledon | 21.3ab | 27.5a | 25.7ab | 27.5abc |
| | 75% cotyledon | 22.3ab | 24.2ab | 30.0a | 31.3a |
| | 100% (Whole nut) | 15.5b | 20.8ab | 23.6ab | 26.5abc |

Note: Means with same letter(s), same column, were not significantly different (P ≤ 0.05).

**Table 5:** Total Leaf area ($cm^2$)/ plant of seedlings of mini-nut *C. nitida* as affected by Pyramidal Device (PD) and Palm Basket (PB) in the Nursery.

| | Weeks After Sowing | | | |
|---|---|---|---|---|
| Treatment | 8 | 10 | 12 | 14 |
| 12.5% cotyledon in PB | 19.0ab | 24.2b | 25.9bc | 90.0abc |
| 25% cotyledon in PB | 44.1ab | 49.3ab | 54.3ab | 90.0abc |
| 50% cotyledon in PB | 41.2ab | 39.4ab | 45.8abc | 93.3ab |
| 75% cotyledon in PB | 27.9ab | 37.2ab | 45.2abc | 93.3ab |
| 100% whole nut in PB | 27.1ab | 33.7ab | 46.4abc | 76.7bc |
| 12.5% cotyledon in PD | 16.8ab | 17.4b | 21.1c | 56.7d |
| 25% cotyledon in PD | 38.9ab | 42.1ab | 46.8abc | 100.0a |
| 50% cotyledon in PD | 20.0ab | 43.1ab | 48.0abc | 100.0a |
| 75% cotyledon in PD | 49.1a | 61.7a | 65.9a | 93.3ab |
| 100% whole nut in PD | 11.5b | 36.3ab | 40.5abc | 70.0cd |
| 12.5% cotyledon in PB+PD | 16.7ab | 25.6b | 29.1bc | 83.3abc |
| 25% cotyledon in PB+PD | 32.1ab | 40.6ab | 40.4abc | 86.7abc |
| 50% cotyledon in PB+PD | 23.3ab | 41.3ab | 41.8abc | 93.3ab |
| 75% cotyledon in PB+PD | 41.7ab | 61.7a | 58.5a | 80.0abc |
| 100% whole nut in PB+PD | 36.8ab | 41.3ab | 51.0ab | 53.3d |

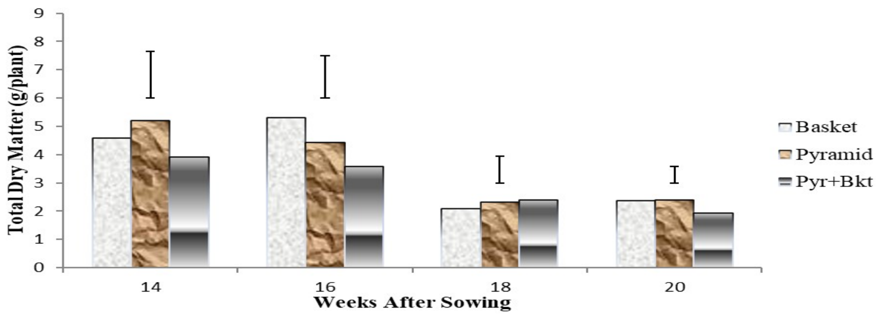
Note: Means with same letter(s), same column, are not significantly different (P< 0.05).

With respect to interactions, at 8 WAS, seedlings of 75% portion of cotyledon treated with curing in PD were significantly the largest in total leaf area (49.1 $cm^2$) compared to all other treated nuts with curing media which range between 23.3 $cm^2$ and 44.1 $cm^2$ (Table 5). However, seedlings from 12.5% portion of cotyledon treated to PB, PD and PB+PD had a reduced leaf area production with 19.0 $cm^2$, 16.8 $cm^2$ and 16.7 $cm^2$ respectively, and 100% whole nuts treated to PD with significantly reduced leaf area production of 11.5 $cm^2$; the differences were significant. At 10 WAS, seedlings raised from 75% portion of cotyledon treated to curing in PD and PB+PD recorded significantly highest leaf area of 61.7 $cm^2$. These were followed by 25% portion of cotyledon treated to curing in PB (49.3 $cm^2$), 50% and 25% portion of cotyledon treated to curing in PD with 43.1 $cm^2$ and 42.1 $cm^2$, respectively, as well as 50% portion of cotyledon and 100% whole nuts treated to curing in PB+PD with leaf area of 41.3 $cm^2$ each. All other treatments recorded leaf area between 33.7 and 40.6 $cm^2$ while 12.5% of cotyledon treated to storage in PB, PD and PB+PD had a significantly lowest leaf area production with 24.2 $cm^2$, 17.4 $cm^2$, and 25.6 $cm^2$, the differences were significant. At 12 WAS, leaf area of seedlings from 75% portion of cotyledon treated with curing in PD and PB+PD were the highest 65.9 $cm^2$ and 58.5 $cm^2$ followed by 25% portion of cotyledon treated to curing in PB and 100% whole nuts treated to curing in PB+PD with 54.3 $cm^2$ and 51.0 $cm^2$ in that order. All other treatments range between 40.4 $cm^2$ and 48.0 $cm^2$, while 12.5% portion of cotyledon treated to curing in PB, PD and PB+PD were the lowest with 25.9 $cm^2$, 21.1 $cm^2$ and 29.1 $cm^2$, respectively. The differences were significant. At 14 WAS, seedlings raised from 25% and 50% portion of cotyledon treated to curing in PD had the largest leaf area of 100 $cm^2$ each. These were closely followed by 50% portion of cotyledon treated to curing in PB, PB+PD and 75% portion of cotyledon treated to curing in PB and PD with 93.3 $cm^2$ each. Seedlings from 12.5% portion of cotyledon treated to curing in PD and 100% whole nuts treated to curing in PB+PD had significantly reduced leaf area of 56.7 and 53.3 $cm^2$, while seedlings raised from all other treatments had between the range of 70.0 $cm^2$ and 90.0 $cm^2$. The differences were significant (Table 5).

### *Total dry matter (g/plant)*

Seedlings derived from nuts treated to curing in PD had the highest (5.2 g) total dry matter content compared to other treatments which recorded 4.58 g in nuts treated to curing in PB and 3.91g in nuts treated to curing in PD and the differences were not significant ($P \leq 0.05$). At 16 WAS, seedlings derived from nuts treated to curing in PB had the highest mean dry matter leaf (5.31 g) followed by PD (4.44 g) and PB+PD (3.59 g) and the differences were significant. At 18 and 20 WAS, the nuts treated to curing in PB, PD and PB+PD had a reduced mean dry matter leaf of 2.3 g to 2.1 g and 2.3 g to 1.94 g in each year, respectively (Figure 4a). The mini-nut treatment had an increase mean dry matter content with the increase in nut sizes from 14 to 18 WAS. At 14 WAS, 100% whole nuts recorded the highest dry matter leaf of 6.49 g followed by 75% and 50% portion of cotyledon with 5.63 g and 4.50 g, while 25% and 12.5% portion had the least of 3.03 g and 3.18 g; the differences were significant. At 16 WAS, 100% whole nuts, 75% and 50% portion of cotyledon had 7.66 g, 5.49 g and 4.69 g dry matter leaf respectively and were not significantly different, however, were significantly different from 25% and 12.5% portion of cotyledon which had the least with 2.02 g and 2.38 g, respectively. Similarly, at 16, 18 and 20 WAS, seedlings derived from 75% portion of cotyledon and 100% whole nut had an increase in dry matter content of the leaf while other mini-nut treatment recorded significantly lower dry matter content of the leaf in each respective period and the differences were significant. At 18 and 20 WAS, the mini-nut treatments had a reduced dry matter leaf of between 1.36 g to 3.98 g and 1.7 g to 2.78 g in each year, respectively; the differences were significant (Figure 4b). At 14 WAS, seedlings derived from 100% whole nuts treated to curing in PB, PD and 50% portion of cotyledon treated to curing in PD having the highest mean dry matter leaf with 2.77g, 3.17g and 2.97g respectively. In the same period, 12.5%, 25% portion of cotyledon treated with curing in PB, 75% portion of cotyledon treated with curing in PD and 12.5%, 25% portion of cotyledon treated with curing in PB+PD recorded reduced dry matter leaf with 1.32 g, 1.47 g, 1.55 g, 1.58 g and 1.35 g value respectively, while all other treatments ranges between 1.80g and 2.28 g; the differences were significant. At 16 WAS, seedlings derived from 75% portion of cotyledon treated to curing in PD were the highest (3.81 g) followed by whole nuts treated to curing in PB (3.55 g) and 100% whole nuts treated to curing in PD (3.48 g) as well as 50% portion of cotyledon treated to curing in PD (3.37 g) and the differences were not significant. Subsequently, seedlings from 12.5%, 25% portion of cotyledon treated to curing in PB and 12.5%, 25% portion of cotyledon treated to curing in PB+PD recorded a significantly reduced dry matter leaf of 1.47 g, 1.48 g, 1.73 g and 1.41 g respectively, while other treatments recorded dry matter leaf from the tune of 2.00 g and 2.91 g. At 18 WAS, the dry matter leaf of seedlings derived from whole nuts treated with curing in PD, 100% whole nuts treated with curing in PB, 75% portion of cotyledon treated with curing in PD and 50% portion of cotyledon treated with curing in PB recorded 6.13 g, 5.90 g, 5.77 g and 5.57 g, respectively; the differences were not significant but significantly higher than all other treatments which ranged between 2.23 g and 5.00 g, while 12.5% and 25% portion of cotyledon treated to curing in PB recorded the least. At 20 WAS, seedlings derived from 75% portion of cotyledon treated with curing in PB+PD, 100% whole nuts treated with curing in PD, and 100% whole nuts treated to curing in PB had the highest dry matter leaf of 6.67 g, 6.40 g and 6.40 g in that order, while 12.5% portion of cotyledon treated with curing in PB (2.63 g), 12.5% portion of cotyledon treated with curing in PB+PD (2.50 g) and 25% portion of cotyledon treated with curing in PB+PD (2.30g) had significantly reduced dry matter leaf. Other treatments produced dry matter leaf from the tune of 3.53 to 5.73g; the differences were significant (Table 6).

a)



**b)**

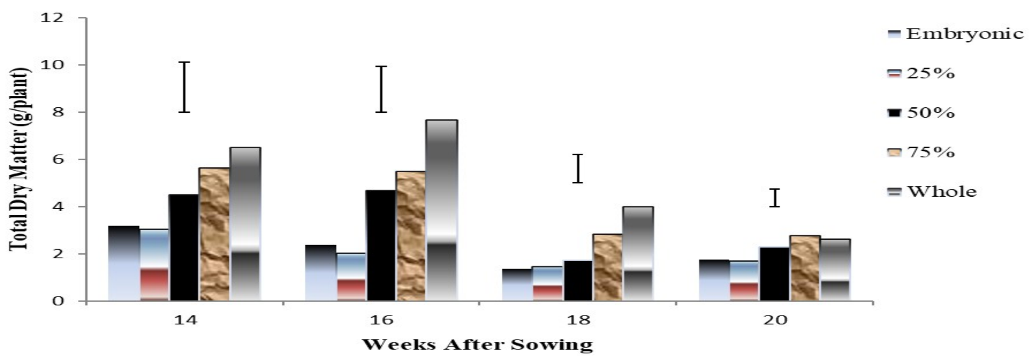



Figure 4: Total Dry matter (g/plant) of kola (*Cola nitida*) seedling as affected by: (a) curing media; (b) mini-nut techniques at 14-20 weeks after sowing (WAS)

**Table 6: Total Dry Matter (g/plant) of kola (*C. nitida)* seedling as affected by curing media (Dodecahedron Pyramidal Shape (DPS) and Palm Basket (PB)) and mini nut technique in the nursery**

| | Weeks After Sowing | | | |
|---|---|---|---|---|
| Treatment | 14 | 16 | 18 | 20 |
| 12.5% cotyledon in PB | 1.32bc | 1.47bcd | 2.10cd | 2.63c |
| 25% cotyledon in PB | 1.47bc | 1.48bcd | 1.93cd | 3.53bc |
| 50% cotyledon in PB | 2.12ab | 2.83ab | 5.57a | 5.73ab |
| 75% cotyledon in PB | 2.18ab | 2.55ab | 5.00ab | 5.63ab |
| 100% whole nut in PB | 2.77a | 3.55a | 5.90a | 6.40a |
| | | | | |
| 12.5% cotyledon in PD | 1.97b | 2.10bc | 2.60c | 4.60bc |
| 25% cotyledon in PD | 2.28ab | 2.35b | 2.90c | 3.57bc |
| 50% cotyledon in PD | 2.97a | 3.37a | 3.80bc | 4.77bc |
| 75% cotyledon in PD | 1.55bc | 3.81a | 5.77a | 6.67a |
| 100% whole nut in PD | 3.17a | 3.48a | 6.13a | 6.40a |
| | | | | |
| 12.5% cotyledon PB+PD | 1.58bc | 1.73bcd | 2.43c | 2.50c |
| 25% cotyledon in PB+PD | 1.35bc | 1.41bcd | 2.23c | 2.30c |
| 50% cotyledon in PB+PD | 1.80b | 2.00bc | 3.70bc | 4.00bc |
| 75% cotyledon in PB+PD | 2.22ab | 2.38b | 4.70b | 4.60bc |
| 100% whole nut in PB+PD | 1.97b | 2.91ab | 2.97c | 4.67bc |

Note: Means with same letter(s), same column, are not significantly different (P< 0.05).

# DISCUSSION

Generally, non-readiness of some seed/nuts to germinate when sown is a characteristic of such seeds which determines the conditions required for germination within or outside the seeds (Vleeshouwers *et al.*, 1995; Thompson, 2000; Fenner and Thompson, 2005). Curing as a form of after- ripening of kolanuts is normally enhanced under dry and warm conditions (Baskin and Baskin, 1998; Perez-Fernandez *et al.*, 2000; Schutz *et al.* 2002). Therefore, environmental factors including curing condition and seed treatments that alter the conditions required for germination are altering the readiness and / or non-readiness of seeds to germinate (Finch-Savage and Leubner-Metzger, 2006). In this finding, curing of kola-nut in pyramidal device (PD) was empirically established to alter the nut dormancy because of its alteration on the condition, within the seed, required for germination of the kola-nut. Earlier researchers on kola reported that maturity and genetic origin of kola-nut are critical determinants of the nut germination. Similarly, in the findings of Ogutuga and Daramola, (1976) and Oladokun, (1986), organic chemical contents changed from complex to simpler forms when nuts of *Cola nitida* and *Cola acuminata* were stored for about six months. Mini-nut as a form of seed treatments expose the internal tissues of the kolanut cotyledons to rapid water imbibition which in effects aid hydrolysis of food reserves and caffeine and polyphenol contents into simple sugars which instigate rapid and even germination within the period of 2- 6 weeks after sowing (Hammed *et al.*, 2018). Water imbibition not only aids the breakdown of food reserves, but Finch also-Savage and Leubner-Metzger (2006) observed that it equally aids the expansion of the cotyledons, thus aiding in germination. Curing device of pyramid (PD) further aided the breakdown of food reserves in the nuts and thus the embryo became instigated to evenly germinate at a short period of 2-6 weeks compared to other treatments. However, the pronounced germination advantage of small size cotyledon portion treated with pyramidal device (PD) could not translate into growth because of the low food reserves occasioned by reduced cotyledon size. The germination advantage could only be carried into the growth phase only if adequate nutrients are made available for the propagules in the right proportion as reported in the seedlings of oil palm (Lucas, 1977) and cashew (Hammed *et al.*, 2015). Mini-nut treatments of *C. nitida* possibly have reduced concentrations of germination-inhibiting chemicals (caffeine and polyphenol) in such a way that its effect on germination earliness infers that varying amount of polyphenol is removed by variations in the mini-nut treatment, and thus, the greater the amount removed, the better the germination. The cut surface exposes polyphenol and other germination-inhibiting phyto-chemicals to lixiviate, under curing condition in dodecahedron pyramidal shape (PD), thus relieving germination. This observation favours the following order of germination in 25%> 50%> 75%> 100% cotyledon whole nuts in dodecahedron pyramidal shape (PD), indicating that as the nut sizes reduced germination increased. Cutting off some portion of the cotyledon or breaking the adhesive force that held the two cotyledons together will remove the mechanical barrier and enhance germination (Ibikunle and Mackenzie, 1974). As part of effort for this research work with the treatment of mini-nut techniques, breaking of the nitida effect by way of cutting off some portion of the *Cola nitida* nut, cured in the pyramidal device and palm basket provided an effective instigating factor for earlier and non-staggered or even germination of the *Cola* *nitida* seeds. Apart from cutting off some portions of the nuts, the nuts curing and storage in succession under the same storage condition, such that with the energizing ability of the pyramid device used as one of the storage media. This is responsible for faster germination experienced in mini nut cured in PD compared with other curing means. The observation of 10.00% germination at 2WAS with 25% cotyledon portion of *Cola nitida* nuts treated with curing in pyramidal device, prior to sowing was an accomplishment of aim, and while at 4 WAS, same treatment had less than 40.00%. Germination that was about 80% at 6 WAS, shows in accordance with the report of Hammed *et al*., (2019), that 25% portion of cotyledon exhibit good seed vigor for propagation of *Cola nitida* and gave reasonably timely and even germination of nuts at 5 weeks of sowing. This report confirms the assertion of Oladokun (2000) that the untimely and uneven germination of *Cola nitida* is more of non-conducive intrinsic cause rather than seed coat effect. The concentration of polyphenols is suspected of the dormancy possessed by *Cola nitida*, thus, cutting off the nuts must have reduced the concentration in two ways; first, treatment of 25% nut portion cured in pyramidal device (PD) which germinated faster had more of the nuts which held much of phenols and caffeine removed. Second, when the nuts were cut, which was a form of wound, then cured in pyramidal device (PD), exposing larger surface area of the tissue to electromagnetic energy which concentrates more energy on the tissue, thus causing the germinating inhibiting hormone to erode gradually. Generally, curing kola (*Cola nitida*) nuts had pronounced effects on the proximate and alkaloids contents of kola (*Cola nitida)* (Hammed *et al*., (2018). As *C. nitida* nuts undergo natural biochemical or physical changes while in storage, this is often necessary for subsequent germination (kucera *et al*., 2005) and Bair *et al*., 2006). Cured nuts germinate faster and better than fresh nuts. This phenomenon was explained by the fact that the embryos of cured nuts were much more developed morphogenetically than that of fresh nuts (Van Eijnateen, 1966; Adebola and Morakinyo, 2005). However, earlier researchers agree that kolanut (*Cola nitida*) possesses post-harvest dormancy and that the dormancy was not seed coat induced. However, the cause of this dormancy as well as its nature was not properly understood but, research report had implicated alkaloids as a germination inhibitor (Wink, 1983; Abdel-Hady *et al*., 2008 Zhou Bing *et al*., 2009). In this study, Biochemical analysis carried out in the mini-nut cured in PB, PD and PB+PD attached to the seedlings growth were correlated with some growth parameters to observing the level of association of the two categories of variables in this findings, moisture content together with some alkaloids contents were negatively correlated where, caffeine and polyphenol of the 25% portion of cotyledon cured in PD attached to the seedling had a reduced contents and had resulted in fastest germination with high carbohydrate and sugar contents, which later support the subsequent growth of the seedlings after germination. This therefore confirms the reports of Ogutuga and Daramola (1976), Oladokun, (1986), Hammed *et al*., (2013) and Olaniyan *et al*., (2018) that the breakdown of the complex compound such as caffeine and polyphenol into their simpler and absorbable forms were responsible for the evenness and timely germination of *Cola nitida* nuts. Also, the reduction in moisture content of the already germinated 25% portion of cotyledon cured in PD to less than 50%, further confirms the earliness in germination observed from the treatment. The result from this study showed that factor responsible for quick germination which Oladokun (1986); (1988) and Van Eijnateen (1969), claim to be unknown is shown to be gradual increase in dry matter yield which was in agreement with the claim of Hammed *et al*., (2018), this indicate the corresponding reduction in moisture content of the nuts. Subsequent improvement in morphological growth observed in 50% portion of cotyledon cured in PD was an indication that irrespective of nut sizes, curing in PD proved to be more effective in seedling growth after germination when compared with different sizes of kola-nuts.

## CONCLUSION

Curing-based mini-nut propagation technique represents a breakthrough in kola (*Cola nitida*) nursery production.

Integrating reduced cotyledon portions with innovative curing media such as the pyramid device (PD) and pyramid plus basket (PB+PD) combination, achieves faster, more uniform and vigorous germination outcomes. This approach holds promise for improving kola seedling supply, enhancing farm productivity, and supporting sustainable cultivation systems.

Adopting PD and PB+PD curing systems, particularly with 25, 50 –75% reduced cotyledon portions—offers a reliable and efficient method for boosting kola propagation in the nursery.

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