



## Effect of mechanical scarification and gibberellins (GA<sub>3</sub>) on seed germination and growth of *Garcinia kola* (Heckel)

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### ABSTRACT

**Objective:** *Garcinia kola* (Heckel) in general has been shown to have a low germination and this constitutes a real obstacle to any domestication initiative. The experiment was conducted in an attempt to improve seeds germination.

**Methodology and results:** Scarification in combination with Gibberellins (GA<sub>3</sub>) application and soaking in distilled water was carried out on fresh seeds and sown in two types of soil substrate: sandy topsoil (1/1) and 100 % topsoil. Parameters related to seed germination and seedlings vigour was evaluated. Results indicated that substrate do not affect seed germination and plant vigour. However, the percentage of germination ( $92.33 \pm 3.19$ ) and germination rate ( $0.013 \pm 0.00$ ) were highest when scarified seeds were soaked in increasing concentrations of GA<sub>3</sub> ( $10^{-4}$  to  $10^{-2}$  g L<sup>-1</sup>). In addition, scarified seeds soaked in distilled water have improved percentage of seed germination. Concerning plants vigour, GA<sub>3</sub> application increased significantly plant height ( $11.22 \pm 4.31$  cm) and plant wingspan ( $13.35 \pm 6.36$  cm) against  $9.19 \pm 4.17$  cm and  $11.67 \pm 6.78$  cm for plant height and wingspan in the control.

**Conclusion and applications of findings:** Results suggest that *G. kola* seeds present seed coat dormancy. Scarification and GA<sub>3</sub> application offer possibly an alternative methodology to improve seed germination and seedling vigour for a large-scale production of *G. kola* seedlings, a prior step of plant species domestication process.

**Keywords:** *Garcinia kola*, seed coat dormancy, mechanical scarification, GA<sub>3</sub>, seed germination

### INTRODUCTION

One of the main problems that prevent sustainable use of Non timber Forest Products (NTFPs) and particularly, medicinal plants, is their low seed

germination. *Garcinia kola* or 'bitter kola' is one of the most important NTFPs in tropical Africa and specifically in Cote d'Ivoire. This plant is endemic in

the humid lowland rainforest vegetation of West and Central African sub regions. It is found in coastal areas and lowland plains up 300 m above sea level with 2000 - 2500 mm rainfall per annum and temperatures ranges from 32.15 to 21.4°C and a minimum relative humidity of 76.34% (Ntamag, 1997; Sunderland, 2001). *G. kola* has a high local market demand or export. This market demand increases the level of exploitation. This overexploitation is essentially due to a large number of applications within both traditional medicine and pharmaceutical industries (Profizi, 1999; Sunderland, 1999). These seeds are commonly eaten as a snack and used for their stimulant effect, due to high caffeine content. Seed extracts have been used as an antidote for cases of food poisoning as it is chewed to prevent the development of any infection when food is suspected to be contaminated by bacteria (Stanley *et al.*, 2014). Several researches have been reported on its medicinal uses. Denloye *et al.* (2009), Murray *et al.* (2012) and Oluwatosin *et al.* (2014) reported antimalarial potential of kolaviron, a biflavonoid from *G. kola* seeds. Farombi and Owoeye (2011) reported antiviral, antihepatotoxic and antidiabetic properties of *G. kola*. Almost all species, including *G. kola*, in Clusiaceae family can only regenerate from seeds in natural environments. Unfortunately, the low germination of seeds has been a problem for natural regeneration or for several initiatives of seedlings production in nursery (Agyili *et al.*, 2006; Yakubu *et*

*al.*, 2014). The mechanism of seed dormancy for this species is unknown. In general, there are two types of seed dormancy: exogenous and endogenous. According to Bewley (1997), seed dormancy is the failure of an intact viable seed to complete germination under favourable conditions. Seeds with exogenous dormancy usually have pericarp and/or seed coat impermeable to oxygen and/or water. Sometime, germination is inhibited by chemicals in epidermis or adjacent interior membranes (Bradford, 1995). With regard to physiological dormancy, seeds concerned have underdeveloped embryos, but simply need time to grow and germinate (Baskin & Baskin, 2004; Finch-Savage & Leubner-Metzger, 2006). According to the literature, several techniques are used to overcome seed dormancy. Gibberellic acid (phytohormone) and stratification (cold treatment) treatments are typically used for leaching out inhibitors while scarification (physical or mechanical treatment) are used to break seed coat dormancy (Ellis *et al.*, 1985; Rehman & Park, 2000; Delanoy *et al.*, 2006; Hilhorst, 2007). Concerning *Garcinia kola*, works realized on seed germination improvement gave mixed results. Germination percentages remain low and the best are around 62 % (Agyili *et al.*, 2006; Yakubu *et al.*, 2014; Koffi *et al.*, 2015). The objective of this study was to develop methods to break dormancy for achieving rapid, uniform and high germination rate of *G. kola* seeds.

## MATERIAL AND METHODS

**Seeds collection and experimental sites:** Fruits of *G. Kola* were collected in August 2014 from trees growing in a mixed forest in Ahouabo, Adzope department, Cote d'Ivoire. Ahouabo site, situated to 100 km from Abidjan, has similar weather characteristics as the University research station. Both sites are located in forest zone where the rainfall pattern is bimodal with 4 distinct seasons, two dry seasons (from December to March and from July to August), and two rainy seasons (from April to June and from September to November). Mean annual rainfall varies between 1800-2000 mm. Mean monthly temperature varies between 27 and 30°C, whereas mean relative humidity ranges between 70-84%. Research was carried out from September 2014 to March 2015 at University research station. Trees were about 10 to 30 m tall and 30 to 80 cm in diameter at breast height (1.5 m

above ground). At the time of collection, fruits were judged mature when their colour changed to greenish to yellow-orange. After collection, fruits were crushed and then seeds were extracted and transported in plastic bags at the University Nangui Abrogoua (former University Abobo-Adjame), in Abidjan for germination tests. Plastic bags with seeds were kept under laboratory climatic conditions (22–25 °C, 50–60% RH) during 72 h before seed germination test.

**Seed germination tests:** After 72 h of fermentation in plastic bags, seeds were washed abundantly with tap water. Cleaned seeds were then subjected to different treatments (mechanical scarification, chemical and soaking). Treatments applied were based on previous results of germination experiments in several plant species (Bradbeer, 1988; Yang *et al.*, 2007; Kouakou *et*

al., 2009; Hang et al., 2010). The mechanical scarification consisted to remove carefully with a sharp blade a part of the pericarp of seeds. For the chemical and soaking treatments, seeds were soaked under total darkness in beakers containing 300 ml of distilled water or the testing solutions (gibberellins-GA<sub>3</sub>). After soaking, seeds treated chemically were rinsed in distilled water prior to be sown in plastic bags filled with substrates in the nursery. Each treatment consisted of three replicates, each one including 50 seeds. Plastic bags were filled with two types of substrates. The first was constituted of a sandy topsoil mixed with soil taken from the forest undergrowth with vegetation mainly composed *Pueraria phaseolides* (Benth) and *Chromolaena odorata* (L.) in a proportion 1:1 (V/V) with a pH= 5.6. The second, with a pH= 5.7, was made of 100% of soil taken from the forest undergrowth. Both substrates were disinfected with a fungicide (maneb 80 %®) before filling the bags. The nursery was covered with a shade built with bamboo and palm fronds. Temperature varied between 27 °C and 29 °C. Nursery was watered every 2 days. Germination was evaluated every 2 days after the first germination was registered. Seeds were classified as germinated when the emerging shoot appear on the surface of substrate in the

bag. Two parameters of germination were evaluated:- germination percentage (evaluated at the end of germination test); - germination rate (R), equal to  $\Sigma n / \Sigma(tn)$ , where  $t$  is the time in days and  $n$  is the number of seeds germinated the day  $t$  (Bewley & Black, 1994; kouakou et al., 2009). Another trait analysed was seed viability. Indeed, at the end of the test, non germinated seeds were cut open using a pruner and classified in two categories following Ellis et al. (1985): (1) fresh seeds (firm and healthy); (2) dead seeds (showing mould, empty and decay).

**Data analysis:** Mean values and standard deviations were calculated for each character with respect to substrate, mechanical scarification and chemical treatment. Combined multivariate analysis of variance (MANOVA) appropriate to three factors was performed to compare these characters, as well as their interaction. This allowed the identification of significant factors based on a vector of dependent variables. The General Linear Models (GLM) procedure of the R statistical version 9.1 was used to identify traits contributing to difference when MANOVA revealed significant difference for a factor. Least Significant Difference (LSD) multiple range-tests were used to identify differences between means.

**RESULTS**

**Effect of treatments on seeds germination:** Table 1 presents multivariate analysis of variance (MANOVA) appropriate to three factors performed to compare different treatments effect, as well as their interaction. This test shows that mechanical scarification, chemical treatment, soaking and their interaction have a significant effect on seeds germination ( $P < 0.05$ ).

**Table 1:** Multivariate analysis of variance (AMOVA) performed to test the overall effect of substrate type, gibberellins application, mechanical scarification and soaking on seeds germination

Sources	Statistical	
	F	P
Substrate	1.268	0.248
Scarification	25.534	0.000
Gibberellins	5.008	0.000
Soaking	3.787	0.017
Substrates*Scarification	2.172	0.002
Substrate*Gibberellins	1.411	0.140
Scarification*Gibberellins	1.895	0.023
Substrate*Soaking	0.749	0.664
Scarification*Soaking	1.353	0.209
Gibberellins*Soaking	2.101	0.003
Substrate*Scarification*Gibberellins	1.205	0.267
Substrate*Scarification*Soaking	0.620	0.780
Substrate*Gibberellins*Soaking	1.137	0.307
Scarification*Gibberellins*Soaking	1.849	0.0128
Substrate*Scarification* Gibberellins*Soaking	1.198	0.289

**Kouakou et al. J. Appl. Biosci. 2016 Effect of mechanical scarification and gibberellins (GA3) on seed germination and growth of *Garcinia kola* (Heckel)**

The mean values of different characters with respect to substrate type, gibberellins application, mechanical scarification and distilled water imbibitions are indicated in table 2. The combined effect of the three parameters had affected significantly seed viability, germination percentage and rate. Significantly, higher viabilities were

registered in all the unscarified seeds contrary to scarified seeds, whatever the treatment. The lowest percentages of viability  $61.67 \pm 5.69\%$  and  $78.33 \pm 9.95\%$  were obtained for scarified seeds soaked in distilled water for 12 h and control 1, respectively against  $98.78 \pm 1.67$  to 100% for unscarified seeds.

**Table 2:** Combined effect of scarification, gibberellins (GA<sub>3</sub>) application and soaking duration on *Garcinia kola* seeds germination.

Treatments			Traits		
Seed type	Gibberellins (GA <sub>3</sub> , mol l <sup>-1</sup> )	Soaking (h)	Seed viability (%)	Germination percentage (%)	Germination rate (R)
Scarified seed	10 <sup>-2</sup>	12	97.00 ± 2.14 <sup>a</sup>	81.67 ± 3.19 <sup>abc</sup>	0.010 ± 0.00 <sup>d</sup>
		24	96.3 ± 1.67 <sup>ab</sup>	<b>90.00 ± 1.92<sup>a</sup></b>	0.010 ± 0.00 <sup>d</sup>
		48	94.00 ± 3.23 <sup>bc</sup>	78.33 ± 9.20 <sup>abc</sup>	0.011 ± 0.00 <sup>c</sup>
	10 <sup>-3</sup>	12	93.33 ± 0.00 <sup>bc</sup>	90.00 ± 3.33 <sup>ab</sup>	0.011 ± 0.00 <sup>c</sup>
		24	90.00 ± 4.30 <sup>c</sup>	<b>92.33 ± 3.19<sup>a</sup></b>	<b>0.012 ± 0.00<sup>b</sup></b>
		48	95.00 ± 1.67 <sup>b</sup>	<b>90.00 ± 4.30<sup>a</sup></b>	<b>0.013 ± 0.00<sup>a</sup></b>
	10 <sup>-4</sup>	12	93.33 ± 3.85 <sup>bc</sup>	86.66 ± 3.85 <sup>abc</sup>	0.010 ± 0.00 <sup>d</sup>
		24	96.67 ± 3.33 <sup>ab</sup>	<b>91.67 ± 5.00<sup>a</sup></b>	<b>0.012 ± 0.00<sup>d</sup></b>
		48	95.00 ± 3.19 <sup>b</sup>	<b>88.67 ± 4.71<sup>ab</sup></b>	0.010 ± 0.00 <sup>d</sup>
	10 <sup>-5</sup>	12	95.00 ± 3.19 <sup>b</sup>	83.33 ± 6.38 <sup>abc</sup>	<b>0.012 ± 0.00<sup>a</sup></b>
		24	98.33 ± 1.67 <sup>a</sup>	85.00 ± 7.39 <sup>abc</sup>	0.011 ± 0.00 <sup>c</sup>
		48	90.00 ± 7.93 <sup>c</sup>	80.00 ± 8.16 <sup>abc</sup>	0.010 ± 0.00 <sup>d</sup>
	H <sub>2</sub> O	12	61.67 ± 5.69 <sup>e</sup>	<b>58.33 ± 9.95<sup>d</sup></b>	0.010 ± 0.00
		24	90.00 ± 3.33 <sup>c</sup>	80.00 ± 5.44 <sup>abc</sup>	0.010 ± 0.00 <sup>d</sup>
		48	95.00 ± 3.19 <sup>b</sup>	<b>88.33 ± 4.19<sup>ab</sup></b>	0.010 ± 0.00 <sup>a</sup>
Control 1		78.33 ± 9.95 <sup>d</sup>	<b>40,00 ± 5,44<sup>e</sup></b>	0.010 ± 0.00 <sup>d</sup>	
unscarified seeds	10 <sup>-2</sup>	12	100.00 ± 0.00 <sup>a</sup>	71.67 ± 8.77 <sup>cd</sup>	0.010 ± 0.00 <sup>d</sup>
		24	100.00 ± 0.00 <sup>a</sup>	75.00 ± 4.19 <sup>bc</sup>	0.010 ± 0.00 <sup>d</sup>
		48	100.00 ± 0.00 <sup>a</sup>	78.33 ± 5.00 <sup>abc</sup>	0.010 ± 0.00 <sup>d</sup>
	10 <sup>-3</sup>	12	98.33 ± 1.67 <sup>a</sup>	80.00 ± 4.71 <sup>abc</sup>	0.010 ± 0.00 <sup>d</sup>
		24	99,66 ± 1,92 <sup>a</sup>	<b>91,66 ± 1,67<sup>a</sup></b>	0,011 ± 0,00 <sup>c</sup>
		48	100,00 ± 0,00 <sup>a</sup>	80,00 ± 8,16 <sup>abc</sup>	0,010 ± 0,00 <sup>d</sup>
	10 <sup>-4</sup>	12	100.00 ± 0.00 <sup>a</sup>	86.66 ± 3.85 <sup>abc</sup>	0.010 ± 0.00 <sup>d</sup>
		24	100.00 ± 0.00 <sup>a</sup>	85.00 ± 1.67 <sup>abc</sup>	0.010 ± 0.00 <sup>d</sup>
		48	100.00 ± 0.00 <sup>a</sup>	81.67 ± 5.69 <sup>abc</sup>	0.010 ± 0.00 <sup>d</sup>
	10 <sup>-5</sup>	12	100.00 ± 0.00 <sup>a</sup>	83.33 ± 6.38 <sup>abc</sup>	0.011 ± 0.00 <sup>c</sup>
		24	100.00 ± 0.00 <sup>a</sup>	88.33 ± 5.00 <sup>ab</sup>	0.010 ± 0.00 <sup>d</sup>
		48	99.66 ± 1.92 <sup>a</sup>	81.67 ± 7.39 <sup>abc</sup>	0.010 ± 0.00 <sup>d</sup>
	H <sub>2</sub> O	12	100.00 ± 0.00	69.33 ± 6.87 <sup>cd</sup>	0.010 ± 0.00 <sup>d</sup>
		24	99.67 ± 1.19 <sup>a</sup>	72.64 ± 3.19 <sup>cd</sup>	0.010 ± 0.00 <sup>d</sup>
		48	100.00 ± 0.00 <sup>a</sup>	73.86 ± 4.71 <sup>cd</sup>	0.010 ± 0.00 <sup>d</sup>
Control 2		98.78 ± 1.67 <sup>a</sup>	<b>55.87 ± 8.67<sup>d</sup></b>	0.010 ± 0.00 <sup>d</sup>	
	<b>F</b>		2.68	2.10	2.07
	<b>P</b>		0.010	0.042	0.044

Control 1: Scarified seeds untreated; Control 2: Unscarified seeds untreated, Different letters in columns indicate statistical difference ( $P < 0.05$ ).

For the percentage of germination, best results were obtained when scarified seeds were treated with GA<sub>3</sub> (contents varying from 10<sup>-2</sup> to 10<sup>-4</sup> g L<sup>-1</sup>) for 24 h or 48 h. Similarly, results were obtained with the germination rate. Indeed, scarified seeds treated with GA<sub>3</sub> (10<sup>-4</sup> and 10<sup>-3</sup> g L<sup>-1</sup>) had accelerated seed germination. Germination rates were 0.012 ± 0.00 and 0.013 ± 0.00 for 10<sup>-4</sup> g L<sup>-1</sup> and 10<sup>-3</sup> g L<sup>-1</sup> of GA<sub>3</sub>, respectively, against 0.010 ± 0.00 for control 1 (scarified seeds untreated).

**Effect of treatments on seedlings vigour:** Table 3 presents multivariate analysis of variance (MANOVA)

appropriate to three factors performed to compare different treatments effect, as well as their interaction. This test shows that mechanical scarification, gibberellins application and interactions Substrate\*Gibberellins and Gibberellins\*Soaking duration affect significantly seedlings vigour (*P* < 0.05). Therefore, results interpretation were made with these simple interactions (Substrate\*Gibberellins and Gibberellins\*Soaking duration) and with individual effect of scarification and gibberellins on seedling vigour.

**Table 3:** Multivariate analysis of variance (AMOVA) performed to test the overall effect of substrate type, gibberellins application, mechanical scarification and soaking on seedlings vigour

Sources	Statistical	
	<i>p</i>	<i>P</i>
Substrate	0.708	0.586
Scarification	3.742	0.005
Gibberellins	6.333	0.000
Soaking	0.273	0.993
Substrate*Scarification	0.596	0.665
Substrate*Gibberellins	0.893	0.743
Scarification*Gibberellins	0.877	0.616
Substrate*Soaking	0.311	0.987
Scarification*Soaking	0.847	0.600
Gibberellins*Soaking	1.876	0.002
Substrate*Scarification*Gibberellins	0.577	0.930
Substrate*Scarification*Soaking	0.589	0.852
Substrate*Gibberellins*Soaking	0.826	0.742
Scarification*Gibberellins*Soaking	0.757	0.834
Substrate*Scarification*Gibberellins*Soaking	1.170	0.235

**Effect of mechanical scarification on seedling vigour** : Results of table 4 indicate that for the four parameters characterizing plant vigour, three were significantly affected by scarification (*P* < 0.05). Mechanical scarification significantly increased plants height (9.27 ±

3.97 cm), Plant collar diameter (2.85 ± 1.49 cm) and growth rate 1.6 ± 0.14 cm month<sup>-1</sup> against 8.48 ± 4.36 cm, 2.16 ± 1.57 cm and 1.5 ± 0.1 for plant height, collar diameter and growth rate, respectively, for plants produced from non scarified seeds.

**Table 4:** Effect of mechanical scarification on *Garcinia kola* seedling vigour

Seed type	Plant height (cm)	Plant collar diameter (cm)	Plant wingspan (cm)	Growth rate (cm month <sup>-1</sup> )
SC	9.27 ± 3.97 <sup>a</sup>	2.85 ± 1.49 <sup>a</sup>	11.66 ± 6.32 <sup>a</sup>	1.6 ± 0.14 <sup>a</sup>
NSC	8.48 ± 4.36 <sup>b</sup>	2.16 ± 1.57 <sup>b</sup>	10.94 ± 6.88 <sup>a</sup>	1.5 ± 0.13 <sup>b</sup>
<i>F</i>	5.82	5.75	1.91	5.51
<i>P</i>	0.016	0.017	0.167	0.013

SC: Scarified seeds; NSC: Unscarified seeds

Different letters in columns indicate statistical difference (*P* < 0.05).

**Effect of gibberellins applications on seedling vigour:** 0.001). Plant size and plant growth rate seem significantly increased with GA<sub>3</sub> content (Table 5).  
 Apart of the collar diameter, all other parameters studied were significantly influenced by GA<sub>3</sub> treatments ( $P <$

**Table 5.** Effect of gibberellins applications on *Garcinia kola* seedling vigour.

Gibberellins (GA <sub>3</sub> mol l <sup>-1</sup> )	Traits			
	Plants height (cm)	Plant collar diameter (cm)	Plant wingspan (cm)	Growth rate (cm month <sup>-1</sup> )
10 <sup>-2</sup>	10.53±4.54 <sup>a</sup>	2.56±1.72 <sup>a</sup>	11.94±6.20 <sup>ab</sup>	1.8±0.11 <sup>a</sup>
10 <sup>-3</sup>	9.89±4.11 <sup>ab</sup>	2.81±1.53 <sup>a</sup>	12.20±6.78 <sup>a</sup>	1.8±0.14 <sup>a</sup>
10 <sup>-4</sup>	9.58±3.84 <sup>b</sup>	2.83±1.44 <sup>a</sup>	12.57±6.59 <sup>a</sup>	1.7±0.14 <sup>b</sup>
10 <sup>-5</sup>	9.17±4.27 <sup>c</sup>	2.64±1.51 <sup>a</sup>	11.13±6.39 <sup>c</sup>	1.6±0.12 <sup>c</sup>
H <sub>2</sub> O	9.19±3.86 <sup>c</sup>	2.76±1.52 <sup>a</sup>	11.55±6.55 <sup>b</sup>	1.6±0.14 <sup>c</sup>
Control	9.02±4.17 <sup>c</sup>	2.59±1.44 <sup>a</sup>	11.67±6.78 <sup>b</sup>	1.6±0.13 <sup>c</sup>
<i>F</i>	4.18	0.58	4.59	4.46
<i>P</i>	0.001	0.719	0.001	0.001

Different letters in columns indicate statistical difference ( $P <$  0.05).

**Combined effect of Gibberellins (GA<sub>3</sub>)-soaking duration on seedlings development:** The mean values observed for plant collar diameter, whatever the treatment, were statistically equal to the control (Table 6). However, gibberellins (GA<sub>3</sub>) application and soaking interacted significantly in affecting plant height, plant wingspan and plant growth rate ( $P <$  0.05). GA<sub>3</sub>

concentrations superior to 10<sup>-5</sup> g L<sup>-1</sup> improved plant growth rate up to 1.8 cm month<sup>-1</sup> against 1.5 cm month<sup>-1</sup> for control plants, whatever soaking duration. In addition, 10<sup>-2</sup> g L<sup>-1</sup> of GA<sub>3</sub>, has greatly, promoted plant growth: 11.22±4.31 cm, for seed soaked 12 h and 24 h, against 9.19±4.17 cm month<sup>-1</sup> in the control.

**Table 6.** Combined effect of gibberellins (GA<sub>3</sub>) and soaking duration on *Garcinia kola* seedlings development.

Treatments		Traits			
Gibberellins (GA <sub>3</sub> mol L <sup>-1</sup> )	Soaking (h)	Plant height (cm)	Plant collar diameter (cm)	Plant wingspan (cm)	Growth rate (cm month <sup>-1</sup> )
10 <sup>-2</sup>	12	10.53±4.54 <sup>a</sup>	2.80±1.68 <sup>a</sup>	9.94±6.30 <sup>e</sup>	1.8±0.13 <sup>a</sup>
	24	11.22±4.31 <sup>a</sup>	2.57±1.72 <sup>a</sup>	8.90±6.22 <sup>f</sup>	1.7±0.12 <sup>b</sup>
	48	9.65±4.90 <sup>b</sup>	2.32±1.76 <sup>a</sup>	7.97±6.07 <sup>g</sup>	1.8±0.13 <sup>a</sup>
10 <sup>-3</sup>	12	9.57±4.58 <sup>b</sup>	2.76±1.56 <sup>a</sup>	12.48±7.00 <sup>b</sup>	1.7±0.14 <sup>b</sup>
	24	9.10±4.03 <sup>c</sup>	2.76±1.64 <sup>a</sup>	11.48±6.95 <sup>cd</sup>	1.7±0.14 <sup>b</sup>
	48	9.80±3.75 <sup>ab</sup>	2.90±1.41 <sup>a</sup>	12.65±6.50 <sup>ab</sup>	1.8±0.15 <sup>a</sup>
10 <sup>-4</sup>	12	9.07±3.96 <sup>c</sup>	2.78±1.58 <sup>a</sup>	12.33±6.90 <sup>bc</sup>	1.8±0.13 <sup>a</sup>
	24	9.67±3.82 <sup>b</sup>	2.76±1.44 <sup>a</sup>	12.02±6.57 <sup>c</sup>	1.7±0.14 <sup>b</sup>
	48	10.01±3.78 <sup>ab</sup>	2.95±1.31 <sup>a</sup>	13.35±6.36 <sup>a</sup>	1.8±0.16 <sup>a</sup>
10 <sup>-5</sup>	12	9.86±4.08 <sup>b</sup>	2.82±1.48 <sup>a</sup>	11.64±6.16 <sup>cd</sup>	1.6±0.13 <sup>c</sup>
	24	8.89±4.51 <sup>c</sup>	2.58±1.45 <sup>a</sup>	11.72±6.65 <sup>cd</sup>	1.6±0.14 <sup>c</sup>
	48	8.39±4.32 <sup>d</sup>	2.51±1.60 <sup>a</sup>	10.04±6.39 <sup>e</sup>	1.7±0.13 <sup>b</sup>
H <sub>2</sub> O	12	8.59±3.91 <sup>cd</sup>	2.84±1.60 <sup>a</sup>	10.68±6.28 <sup>de</sup>	1.5±0.14 <sup>d</sup>
	24	8.88±4.01 <sup>c</sup>	2.61±1.47 <sup>a</sup>	10.97±6.49 <sup>d</sup>	1.6±0.13 <sup>ac</sup>
Control	12	10.11±3.57 <sup>ab</sup>	2.82±1.50 <sup>a</sup>	12.99±6.79 <sup>a</sup>	1.6±0.15 <sup>c</sup>
	48	9.19±4.17 <sup>c</sup>	2.59±1.44 <sup>a</sup>	11.67±6.78 <sup>cd</sup>	1.5±0.15 <sup>d</sup>
<i>F</i>		2.13	0.46	2.07	2.41
<i>P</i>		0.007	0.961	0.01	0.001

Different letters in columns indicate statistical difference ( $P <$  0.05).

## DISCUSSION

**Effect of treatments on seeds germination:** In the present study, mechanical scarification, GA<sub>3</sub> application and soaking duration were also equally effective in increasing seeds germination and seedlings vigour in *Garcinia kola*. As mechanical scarification and GA<sub>3</sub> application promoted percentage of germination and germination rate, *G. kola* could present both seeds coat dormancy and physiological dormancy probably imposed by the chemicals in the seed (Bradber, 1988, Baskin & Baskin, 2003; Debeaujon *et al.*, 2007). However, given that scarified seeds untreated with GA<sub>3</sub> presented higher percentage of germination contrary to unscarified seed, it can be deduced that dormancy was certainly exogenous (Robert & king, 1980; Bewley & Black, 1994; Rehman & Park, 2000). Dormancy imposed by seed coat was broken by scarification and soaking the seed in distilled water. These techniques were efficient for leaching out the inhibitory phyto-compounds present in seed coat. These results confirm those of Yakubu *et al.* (2014) which have revealed that the duration of soaking of fresh decoated seeds increased significantly the percentage of germination of *Garcinia kola* seeds. Soaking seed in water or Hydropriming allows the seeds to quickly reach a high level of moisture with a sufficient content supply of oxygen. Therefore, it can increase the level of some metabolites associated with the germination process and enzymes implicated in energy production (Afzal *et al.*, 2012; Zulueta-Rodríguez *et al.*, 2015). The lowest percentages of viability obtained with scarified seed could be explained by the fact that damage could have occurred during mechanical scarification. Indeed, *G. kola*

possesses an almost microscopic embryo merged with the albumen, given that the micropylar region is very sensitive (Kouakou *et al.*, 2009). In addition, GA<sub>3</sub> has accelerated seed germination rate and percentage. Gibberellic acid treatment is still used for leaching out inhibitors present in seeds (Ellis *et al.*, 1985; Rehman & Park, 2000; Delanoy *et al.*, 2006; Kouakou *et al.*, 2009). It can be concluded that both treatments (scarified seeds and co-applying GA<sub>3</sub>) and soaking were able to overcome dormancy.

**Effect of treatments on seeds vigour:** In the present study, scarification, GA<sub>3</sub> application and soaking duration were effective to increase the seed germination and seedling vigour (height, collar diameter and plant wingspan). These treatments are priming techniques, as well as several other priming methods (osmopriming, halopriming and the biopriming) employed to increase the germination and improve the uniformity in emergence of problematic seeds and also to promote the growth and development of many plant species: soybean (Rouhi *et al.*, 2011); tomato (Nawaz *et al.*, 2011); Mexican fir tree (Zulueta-Rodríguez *et al.*, 2015). According to McCourt *et al.* (2005), GA<sub>3</sub> can be described as a 'transitional' hormone, promoting growth and changes in developmental status. GA<sub>3</sub> is able to influence physiological and morphological development of seedlings from seed treated with gibberellins. For example, leaf emergence time and the time to flowering are strongly influenced by Gibberellins (Evans & Poethig, 1995; Bewley, 1997; Bentsink & Koornneef, 2002).

## CONCLUSION

*Garcinia Kola* presents seed coat dormancy and the presence of some inhibition in this tegument can explain the low and not uniform seed germination. Treatments using seeds scarification combined with soaking in GA<sub>3</sub> or in distilled water gave the highest germination percentages. These techniques were efficient for leaching out the inhibitory phyto-compounds present in seed coat. GA<sub>3</sub> application also improves seedlings development and growth. This study can be adopted as a good alternative of other treatments, which produced low

germination percentages and irregular germination. Further investigations are needed to improve the seedlings mass-production. This can concern other priming methods such as halopriming (treatment of seed with salt in order to improve germination) and osmopriming (seed soaked in a solution of polyethylene glycol in order to improve germination). Biochemical analysis can be performed to better apprehend the physiological mechanisms that take place in germinating seed.

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