## PAPER FOR PUBLICATION

# (HECKEL)

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# ABSTRACT

The aim of this study was to determi ent for the optimum germination of Garcinia Kola Heckel and also to in. \_\_\_\_\_ ... , \_\_... vth characteristics. The study was conducted using 5 pre-treatments, these involved soaking 20 seeds of each treatment in cold water at room temperature for periods of 48, 72, 96 hours, 1 and 2 weeks respectively, before sowing, subjecting seeds to hot water treatment at 50°c, 70°c and 90°c for 30minutes respectively, treating seeds in concentrated sulphuric acid for 10, 30 and 60 minutes respectively; removal of the seed coat of seeds and the control seeds (no treatment). Top soil was used as germination medium. The results showed that removal of seed coat and also soaking of seeds in cold water for 96 hours respectively proved very effective for germinating the seeds of G.kola and were thus recommended for its propagation. Its growth rate (17cm in 20 weeks) was quite slow, by all standards, and this gives an indication of what it could be in nature, especially given the high population density in the tropical rain forest area of Nigeria where it naturally dominates. This possibly accounted for the species being almost extinct. Thus, this study has provided better understanding of its propagation which is needed by farmers, as this will help this all important threatened species from going into extinction. Key words: germination, pre-treatments, G.kola.

## INTRODUCTION

Garcinia kola Heckel, often called bitter kola is an indigenous medicinal tree belonging to the family Clusiaceae, formerly Guttiferae.

years, and found in moist forests throughout west and central Africa (Vivien and Faure, 1985).

Garcinia kola has been proved as one of the many non-timber forest products that are of high

socio-economic importance (Adebisi, 2004).

The many uses of the plant have resulted in its over exploitation which now threatens it with extinction in several west and central African countries such as Coted'Ivoire (FAO, 1996), Togo (Ministry of Environment and Forest Resources, Republic of Togo, 1998) Congo and Cameroon (Tchatat, 1999).

In Nigeria, low populations of the plant are found in home gardens and few stands are found in the wild due to rapid deforestation and heavy exploitation in the natural forests (Ajayi, 2016).

The major difficulty in propagating *G.kola* and other *Garcinia* genus is related to seed germination (Yakubu, 2014).

Due to dormancy, *Garcinia kola* seeds can take 18 months to germinate (Aduse-Poku et al, 2003).

Researchers have studied the germination problems of *Garcinia kola* seeds and suggested various means of breaking its dormancy (Anegbeh et al, 2006);(Oboho and Urughu, 2010; Oboho and Ogana, 2011).

Garcinia kola seeds have both seeds coat dormancy and physiological dormancy probably imposed by the chemicals in the seed (Oboho and Urughu, 2010).

Nzegbule and Mbakwu,2001 had reported the presents of a substance associated with germination inhibition, phenol, in the seed of *G.Kola* 

However, there is still a need to investigate simple and practicable methods that farmers could easily adopt with low technological input, to improve the germination of *Garcinia kola* which is not available now in the South Eastern Nigeria.

The main objective of the study therefore was to investigate the best methods to break dormancy and improve germination of *Garcinia kola* seeds within a short time using pre-treatments and to investigate its early growth characteristics.

# **MATERIALS AND METHODS**

**Study Area-** The study was conducted around Agu-Awka, near Esther Obiakor estate, behind government house ,Awka, Anambra state Nigeria, Awka is a town that lies between latitude (7°00' and 7°10')N and longitudes (6° 05' and 6° 15')E in the rain forest of Nigeria(Richards, 2005) Representative seed samples were collected from the market at Awka in November. The bulked seed lot was thoroughly mixed and transported to the study area. The containers used for collecting the seeds gave an open packing arrangement that ensured good flow of air around the seeds and thereby facilitated drying and also reduced the hazards of mould attack and decay. Simple float test and a cut test were used to determine the internal state of the seeds(generally full. shriveled or empty). Shriveled seeds were removed before the dormancy-breaking

**Seed pre-treatment-** five treatments were used.

**Treatment one-** 20 seeds of each treatment were soaked in clean cold water at room temperature for periods of 48, 72 and 96 hours respectively and also for 1 and 2 weeks.

**Treatment two-** this involved subjecting 20 seeds in each to hot water treatment ranging from 50°c, 70°c and 90°c for 30 minutes in each case.

**Treatment three-** this involved treating 20 seeds with concentrated tetraoxosulphate IV acid for varying periods of time ranging from 10 minutes, 30 and 60 minutes. The seeds were properly rinsed with distilled water before sowing in poly bags.

Treatment four- this involved removal of the seed coats before sowing.

**Treatment five-** was the control where 20 untreated seeds were sown into the poly bags.

Manually, water was applied daily so that the medium was kept moist at all times without getting water-logged.

The choice of the different treatment levels for temperature, cold water and concentrated acid were randomly taken.

Simple viability tests were conducted on the seeds before the dormancy breaking treatments.

# DATA COLLECTION AND ANALYSIS

In the nursery, the criterion for germination was a visible protrusion of the shoot apex or epicotyls on the surface of the soil.

Germination was recorded daily until no further germination occurred.

Data on percentage germination, cumulative germination, complete dormancy period, total dormancy period, differential dormancy period, germination rate (Djavanshir and Pourbeik, 1976) and germination speed were collected.

Also weekly measurements of the height of the seedlings, leaf area and number of branches were recorded to determine the early growth characteristics.

Photographs of the seedlings were taken at different developmental stages.

**Complete dormancy period-** The duration (days) from the date of sowing to the date of first germination was recorded, for each treatment.

**Total dormancy period-** The duration (days) from the date of sowing to completion of germination for each treatment was recorded.

**Differential dormancy period**. The difference between the total dormancy period and the complete dormancy period (days) was recorded for each treatment.

**Germination speed-** The number of days it took for 50% of the sown seeds to germinate was recorded for each treatment.

**Cumulative germination-** The total number of seeds that germinated throughout the period of experiment was recorded for each treatment.

**Percentage germination-** The total number of seeds that germinated divided by the number of seeds sown multiplied by 100 recorded for each treatment.

**Germination rate**- A record of the total number of seeds that germinated per number of days in the germinator was taken for each treatment.

The height of the seedlings was measured using metre rule; also length and width of the leaf were measured.

The leaf area was calculated by leaf length a leaf width.

The mean seedling height and leaf area were calculated. The number of leaves was counted and recorded.

The experiment lasted for seven months. (212 days).

# **RESULTS**

The seeds of *Garcinia kola* showed hypogeal germination (Plate 1)

Treatments studied had profound effects on the germination periods of *Garcinia kola*. Germination commenced after 62 days of sowing with seeds whose seed coat was removed.

For the intact seeds soaked in cold water for durations of 48 hours, 72 hours, 96 hours, 1 week and 2 weeks, germination commenced at 85, 84, 110, 88 and 97 days respectively (table1)

Germination was faster and more abundant when the seed coat was removed than soaking of intact seeds for various time durations.

(Table 1). Seeds whose seed coats were removed equally recorded the highest germination percentage of 95% while the control seeds (no treatment) and seeds soaked in hot water (50°c) had the lowest germination percentage of 55% (Table 1).

It is equally worth pointing out that seeds soaked in cold water for 96 hours recorded a germination percentage of 85%.

All the seeds treated in hot water at 70°c and 90°c and all seeds treated with concentrated sulphuric acid at various time durations failed to germinate (table 1).

Table 1 also shows that seeds whose seed coats were removed had low total dormancy period, hence germinated first, intact seeds soaked in cold water between 72-96 hours had comparatively moderated complete dormancy period, hence were the next to germinate while seeds soaked in water for 48 hours, 1 week, 2 weeks and seeds soaked in hot water (50°c) and

the control seeds (no treatment) had maximum total dormancy period in this study, hence delayed germination.

Complete dormancy period ranged from 62-114 days in this investigation.

# Mean height of the seedlings, leaf area and number of leaves and branches.

There was a steady increase in seedling height with time (table 2) (plates 1 and 2).

The leaf area also increased with time, then stopped at 14 weeks (table 2). The number of the bifoliate leaves equally increased with time (table 2, plates 1 and 2). There was no development of branches observed within the period of twenty weeks of study (table 2) (plates 1 and 2).

# **DISCUSION**

The complete dormancy period obtained in this investigation ranged from 62- 114 days. These values were lower than 70-109 days obtained by Oboho and Ogana (2011) in the germination of *Garcinia kola* seeds and also lower than 95 days for 1<sup>st</sup> germination obtained by Eyog-Matig (2007), for decoated *Garcinia kola* seeds soaked in cold water (30°c) for 24 hours but higher than 12-62 days obtained by Yakubu et al, (2014) in the germination of decoated *Garcinia kola* seeds and also higher than 35-63 days obtained by Bolanle-Ojo et al, (2014) in the germination of decoated and soaked *Garcinia Kola* seeds.

Seed coat removal yielded the best result in this investigation suggesting that part of the dormancy in *G.Kola* is seed coat induced. This agrees with the report of Thomlinson et al.(2000) that seed cot may be overcome by peeling off the coat.

Soaking of coated seeds in cold water for 96hours proved effective in his investigation also. This could be due to the water dissolving the impermeable seed coat, hence allow water and oxygen into the embryo of the seed and enhance germination. Soaking of seeds one and two weeks yielded poor germination in this investigation. This could be due to microbial attack and subsequent destruction of the embryo.

When the coated seeds were soaked in hot water for 30 minutes at various temperatures, germination occurred at 50°c, and not above it since seeds treated at 70°c and 90°c failed to germinate. Germination failure was previously reported when soaking *Garcinia kola* seeds in warm water (60°c) for 8 hours (Nzegbule and Mbakwu, 2001) and this confirms that warm water treatment affected negatively the germinations of *Garcinia kola* seeds.

Coated seeds in hot water (50°c) germinated showing that seed coat may protect, to a certain extent, the embryo from destruction by warm water.

When the seeds were treated with concentrated tetraoxosulphate VI acid  $(H_2SO_4)$ , at various time durations, they all failed to germinate.

It appeared the concentrated acid charred the thin seed coat and destroyed the embryo.

Similarly, Aliero (2004), reported that prolonged emersion of seeds in  $H_2SO_4$  injured the seeds since the acid could destroy vital parts of the embryo.

The untreated seeds (the control) recorded the longest complete dormancy period of 114 days and also the least percentage germination of 55%. This reflects the dormancy in *Garcinia kola* seeds.

It appeared that seed coats of *Garcinia kola* restricted water supply and gaseous exchange into the embryos and inhibited germination in the control treatment.

## CONCLUSION

This work has revealed certain important aspects of the ecology and biology of Garcinia kola.

- Its seeds showed seed coat dormancy problems which can be broken by removing the seed coat and also soaking intact seeds in cold water at room temperature for 96 hours or there about.
- Soaking treated seeds in hot water and concentrated sulphuric acid for different periods negatively affected the germination of the seeds.
- The high percentage germination recorded in this research work can be attributed to regular supply of water to the seeds in the nursery from sowing to germination.
- It shows hypogeal germination
- The cotyledons are not separated (eudicot)
- The bifoliate leaves show alternate leaf arrangement
- Water in the soaked seeds were changed every 24 hours hence no problem of microbial infection.
- The seedlings exhibit a very slow growth rate (17cm in 20 weeks)
   (Table 2)

Because of these attributes of the seed, it is difficult to germinate and grow profusely in the wild.

A better understanding of its propagation is needed by farmers through good nursery and agroforestry practices.

This will help this all-important, threatened species from going into extinction.

# Disclaimer:

This paper is based on preliminary dataset. Readers are requested to consider this paper as preliminary research article, as authors wanted to publish the initial data as early as possible. Authors are aware that detailed statistical analysis is required to get a scientifically established conclusion. Readers are requested to use the conclusion of this paper judiciously as statistical analysis is absent. Authors also recommend detailed statistical analysis for similar future studies.

Treatments	Comple te dorman cy period 1st germin ation (days)	Total dormanc y period (TDP) (days)		Germina tion speed (G S) days	Cumm ulative germin ation (CG) (days)	Percentag e (%) germinati on (GP) (days)	Germination rate(GR) (No. per day)
Seeds soaked in cold water (48hrs)	85	212	127	85	15	75	0.07

# TABLE 1

reatment for 30 nins ( $50^{\circ}c$ )  Hot water	Seeds soaked	84	164	80	42	15	75	0.07
Seeds soaked   110	in cold water							
in cold water       (96hrs)       88       212       124       74       12       60       0.05         in cold water       (1 week)       212       115       71       15       75       0.07         Seeds soaked in cold water       97       212       115       71       15       75       0.07         Hot water       105       212       117       82       11       55       0.05         reatment for 30 mins (50°c)       -       -       -       -       -       -       -         Hot water treatment for 30       -       -       -       -       -       -       -         Hot water treatment for 30       -       -       -       -       -       -       -	(72hrs)							
(96hrs)	Seeds soaked	110	183	73	24	17	85	0.08
Seeds soaked   88   212   124   74   12   60   0.05     In cold water (1 week)   Seeds soaked   97   212   115   71   15   75   0.07     In cold water (2 week)   Iot water reatment for 30   105   212   117   82   11   55   0.05     Hot water treatment for 30 mins (70°c)   Hot water treatment for 30   -	in cold water							
in cold water (1 week)       97       212       115       71       15       75       0.07         Seeds soaked in cold water (2 week)       105       212       117       82       11       55       0.05         Hot water for 30 mins (50°c)       -<	(96hrs)							
(1 week)	Seeds soaked	88	212	124	74	12	60	0.05
Seeds soaked in cold water (2 week)         97         212         115         71         15         75         0.07           Hot water reatment for 30 mins (50°c)         105         212         117         82         11         55         0.05           Hot water treatment for 30 mins (70°c)         -	in cold water							
in cold water       (2 week)         Hot water       105       212       117       82       11       55       0.05         reatment for 30 nins (50°c)       -	(1 week)							
(2 week)       105       212       117       82       11       55       0.05         reatment for 30 nins ( 50°c)       -	Seeds soaked	97	212	115	71	15	75	0.07
Hot water       105       212       117       82       11       55       0.05         reatment for 30 mins (70°c)       -	in cold water							
reatment for 30 nins ( $50^{\circ}c$ )  Hot water	(2 week)							
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	Iot water	105	212	117	82	11	55	0.05
50°c)         - <td>reatment for 30</td> <td></td> <td></td> <td></td> <td></td> <td></td> <td></td> <td></td>	reatment for 30							
Hot water $         -$	nins (							
treatment for 30 mins (70°c)       - <td< td=""><td><math>50^{\circ}c)</math></td><td></td><td></td><td></td><td></td><td></td><td></td><td></td></td<>	$50^{\circ}c)$							
mins (70°c)         - <th< td=""><td>Hot water</td><td>-</td><td>-</td><td>-</td><td>-</td><td>-</td><td>-</td><td>-</td></th<>	Hot water	-	-	-	-	-	-	-
mins (70°c)         - <th< td=""><td></td><td></td><td></td><td></td><td></td><td></td><td></td><td></td></th<>								
treatment for 30								
	Hot water	-	-	-	-	-	-	-
$mins (90^{\circ}c)$	treatment for 30							
	$mins (90^{\circ}c)$							
Conc. Sulphuric	Conc. Sulphuric	-	-	-	-	-	-	-
acid treatment								
for 10 mins	for 10 mins							

TABLE 2  for  Rev  Coa Table 2: Mean height of seedlings, leaf area and number of leaves and branches  Cotation seeds   117   212   70   10   11   100							
	n seeus 114 eatm <b>gré</b> dling height (1	cm) Leaf area	$(cm)^2$	No.of	No. of branches		
V	Svv IIIg IIvigIIv (	<del></del>	(•)	leaves			
1							
2.	9.0	4.50		2	-		
3.	9.5	11.25		4			
4.	10.0	21.44		4	-		
5.	10.05	34.03		6	-		
6.	12.0	37.84		6	-		
7.	13.0	40.50		6	-		
8.	13.2	40.50		6	-		
9.	13.5	54.00		6	-		
10.	15.0	54.90		6	-		
11.	15.5	56.25		8	-		

12.	16.0	56.25	8	
			0	-
13.	16.0	56.25	8	-
14.	16.0	58.50	10	-
15.	16.2	58.50	10	-
16.	16.5	58.50	10	-
17.	16.5	58.50	10	-
18.	16.7	58.50	10	-
19.	16.7	58.50	10	-
20.	17.0	58.50	10	-

Mean(x) = 14.147 mean(x) = 46.22 mean(x) = 7.368  
S.E = 0.629 S.E = 3.97 S.E = 0.573  
Con. Limit = 14.147 con. Limit = con limit = 
$$\frac{+}{0.629}$$
 46.22 $\frac{+}{3.97}$  7.368 $\frac{+}{0.573}$ 

at 95%



Plate 1: Hypogeal germination in G. kola

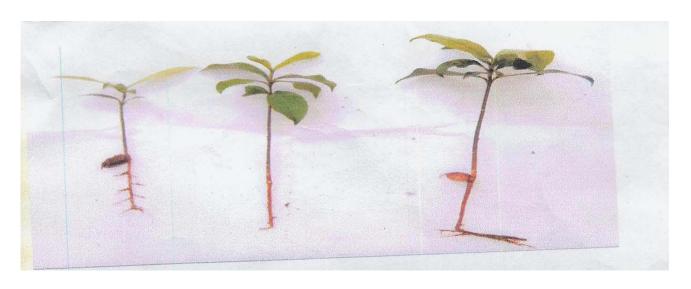


Plate 2: Early developmental stages in G. kola

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