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EFFECTS OF LONG STORAGE PERIOD AND WATER SOAKING DURATION ON GERMINATION OF *Terminalia ivorensis* A. Chev

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ABSTRACT: *Terminalia ivorensis* A. Chev. is an important timber species reported to be threatened by poor regeneration which is through seed. Mature dewinged dried seeds of *T. ivorensis* were subjected to different storage periods (0, 3, 6, 9 and 12 months at room temperature, respectively). Stored seeds were also subjected to 6 different hours of soaking in cold water (0, 12, 24, 48, 72 and 96 hours, respectively). The seeds were sown in sterilizedriver-sand and watered three times a week in the morning. Daily germination count was recorded while Mean Germination, First Day of Germination (FDG), Last Day of Germination (LDG), and Time Spread of Germination (TSG), Germination Index (GI) and Coefficient of Velocity of Germination (CVG) were calculated. Data obtained were subjected to analysis of variance at 5% probability level. At 21 days after sowing (DAS), germination commenced. Highest mean germination (0.47 seeds/day) was recorded from seeds stored for a period of 6 months. Followed by freshly collected seeds and seeds stored for 3 months with 0.27 seeds/day. Seeds stored for 3 and 6 months commenced germination earlier (22 days) than freshly collected seeds and the rest of storage treatments. It was observed that GI and CVG decreased with and was significantly affected by the length of storage as fresh seeds had the highest values (1.10 and 0.62, respectively) while lowest was observed in 12 month storage (238.94 and 74.38, respectively). Water soaking duration had no significant effect on germination of *T. ivorensis* seeds but, soaking for 12 hours had highest values for TSG (10 days), GI (1.06) and CVG (238.34). Long term storage above a period of 6 months under an ambient condition is not recommended for *T. ivorensis* seeds while 12 hours water soaking is encouraged to germination.

Keywords: *Terminalia ivorensis*, storage period, water soaking, germination index, dormancy

1. INTRODUCTION

Terminalia ivorensis of *Combretaceae* family is a large deciduous forest tree ranging in height from 15 to 46 m, with a very straight with small buttresses and sometimes fluted. According to Orwa *et al.*, (2009) the matured trees have very flat topped with a wide horizontal canopy of

evenly distributed foliage arising from the apex of the straight bole. In young trees, the branches are whorled; deciduous, young shoots and foliage falling a few years after initial growth, leaving sockets to mark their original position on the bole (Richter and Dallwitz, 2009). The species is generally called black afara, French (framiré); Spanish (terminalia); Swahili (mwalambe) and in Nigeria

(idigbo). It bears winged fruit with single seed. This important timber species has been reported to be under threat by loss of habitat and poor regeneration thereby, classified as ‘vulnerable’ by IUCN (Hawthorne, 1998). Additionally, this species, whose storage behavior is classified as orthodox, is having difficulty producing seedlings due to chemical and physical seed coat dormancy (Somade, 1985; Schaeffer, 1990).

Seed storage is an important step towards the preservation and conservation of plant genetic resources (mostly seed). Storing the seeds of economic important plants is necessary to preserve planting stocks till next planting season or when there is failure in seed production. The successful storage of orthodox seeds such as *Terminalia* species requires the control of seed moisture content and storage temperatures although, other factors do also influence seed longevity in storage (Dadjoet *al.* 2019; De Vitiset *al.* 2020). Most orthodox seeds can be stored for long periods in refrigerated conditions (0°C to 5°C or even down to -20°C) (FAO, 2018).

Storage conditions have great impact on seed quality and its germination. Baskin and Baskin, (2020) opined those seeds of many species are dormant at maturity although freshly collected seeds of some are non-dormant (Russell, 2011). Many researchers have indicated that soaking in water can influence breaking of dormancy in seeds of various plant species (Usman *et al.* 2010; Odoiet *al.* 2019; Dadjoet *al.* 2019). Water treatment can be used to brake the dormancy imposed by water-impermeable, hard seed coat like that of *T. ivorensis* seeds. Therefore, this study focused on obtaining the appropriate storage period at room temperature and water soaking duration necessary in germination of *Terminalia ivorensis* seeds with the aim of reducing seeds wastage as a result of storage and to enhance seedlings production.

2. MATERIAL AND METHODS

2.1. Experimental Site

The experiment was carried out in the Sustainable Forest Management Department (SFM), Forest Research Institute of Nigeria (FRIN), Jericho Hills, Ibadan, Oyo State, Nigeria. The area lies between Latitude 7° S and 9° N and Longitude 3° W and 58° E of Greenwich Meridian Time (GMT). The climate of the area is dominated by wet and dry seasons with average annual rainfall ranging from 1300 to 1500mm. The average temperature is about 37.2° C and average relative humidity of about 75.8%. The dry season spreads between November and March while the rainy season is from April to October (Forestry Research Institute of Nigeria, 2022).

2.2. Seed Collection and Processing

Seed collection was done in January 10th, 2021. Matured seeds of *Terminalia ivorensis* were manually collected from a standing tree with good phenotypical features (plus tree) from the arboretum, FRIN. Collection was done with the assistance of an expert tree climber after the seeds had relatively dried on the mother plant and about to dispersing. The seeds were brought to the nursery for sun drying for a period of three days. This was necessary for seed wings removal (dewing).

2.3. Seed Storage and Pre-sowing Treatment Method

Five thousand (5000) seeds were carefully selected and arranged into five (5) different storage treatments. The treatments include A: fresh seeds (Control), B: seeds stored for 3 months, C: seeds stored for 6 months, D: seeds stored for 9 months and E: seeds stored for 12 months. Each treatment consists of one thousand (1000) seeds which were spread on black polythene, placed on the floor and stored at room temperature. Fresh seeds were sown after due process and subjected to soaking treatments while other treatments were sown at the end of each storage periods.

From the seeds subjected to different storage periods, total number of three hundred (300) seeds were counted out and subjected to different water soaking treatments. Six (6) varying hours of soaking in cold water were considered which include 96 hours, 72 hours, 48 hours, 24 hours, 12 hours and 0 hour (control). At the end of each soaking durations, the seeds were removed from the water, drained in a plastic sieve and air-dried for 24 hours. They were sown in plastic germination baskets of dimension 10 x 10 x 5 cm, filled with river-sand to 3/5 height of the baskets. The baskets were placed in the screen house and watered three times a week in the morning.

2.4. Data Collection

Data collection commenced after 21 days of sowing when first germination was noticed. A seed is considered to have germinated as soon as the radicle is clearly seen and no more in the seed coat (Auld *et al.*, 1988). Daily germination was recorded while and overall germination percentage, Mean Germination, First Day of Germination (FDG), Last Day of Germination (LDG), and Time Spread of Germination (TSG), Germination Index (GI) and Coefficient of Velocity of Germination (CVG) were considered.

FDG: day on which the first germination event occurred (Kader, 1998)

LDG: day on which the last germination event occurred (Kader, 1998).

Time spread of germination: the time in days between the first and last germination events occurring in a seed lot (Kader, 1998).

$$CVG = N_1 + N_2 + \dots + \frac{N_x}{100} \times N_1 T_1 + \dots + N_x T_x$$

.eq. 1

N- Number of seeds germinated each day: T- No. of days from seeding corresponding to N (Jones and Sanders, 1987).

$$\text{Germination index } -GI = (10 \times n_1) + (9 \times n_2) + \dots + (1 \times n_{10}) \text{ eq. 2}$$

n_1, n_2, \dots, n_{10} = Number of germinated seeds on the first, second and subsequent days until the 10th day; 10, 9 ... and 1 are weights given to the number of germinated seeds on the first, second and subsequent days respectively (Bench *et al.*, 1991).

2.5. Experimental Design and Data Analysis

The experiment consists of 5 by 6 factorial treatments arranged in Completely Randomized Design (CRD) with 5 replicates (10 seeds per replicate per treatment). Total number of one thousand five hundred (1500) seeds was used for the experiment. Data recorded were subjected to analysis of variance (ANOVA) while the means was separated using Duncan Multiple Range Test.

3. RESULTS

3.1. Effects of Long Seed Storage on Germination of *Terminalia ivorensis*

Overall germination percentage: percentage germination of seeds both fresh and stored ranged from 34 to 60%. Seeds stored for 3 months had the highest

germination percentage (60%) followed by freshly collected ones (59%) while those stored for 12 months recorded the least germination percentage with 34% (Figure 1).

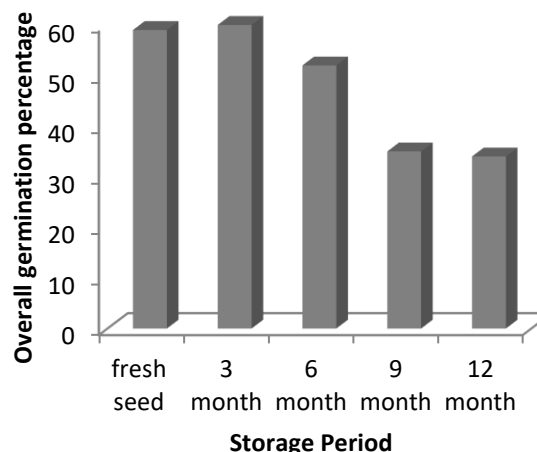


Fig 1: Overall germination percentage of *Terminalia ivorensis* seeds subjected to storage periods.

Mean Germination: according to the Table 1 below, the highest mean germination of 0.47 seeds/day was recorded from seeds stored for a period of 6 months at 21 Days After Sowing (DAS). This was followed by freshly collected seeds and seeds stored for 3 months with 0.27 seeds. At 23 and 25 DAS, seeds stored for 3 months recorded the highest value (0.87 and 1.40, respectively). However, at 27DAS, 9-month storage had the highest mean germination (1.03) followed by 12-month storage (0.97) but each of these treatments were not significantly different from mean germination of those of other periods of seeds storage. It was also observed that at 29, 31 and 33 DAS, fresh seeds had highest mean germination while those stored for 9 and 12 months had the least. These results implied that as storage period increases, the stored seeds germination decreases.

Table 1: Mean germination of *Terminalia ivorensis* seeds subjected to different storage periods

Periods of storage (Months)	Mean germination of <i>Terminalia ivorensis</i> seeds, days after sowing						
	21	23	25	27	29	31	33
12	0.03b	0.47b	0.53b	0.97a	0.70b	0.63b	0.33b
9	0.03b	0.43b	0.60b	1.03a	0.73b	0.63b	0.00b
6	0.47a	0.63ab	1.23a	0.83a	0.83b	0.97a	0.37a
3	0.27ab	0.87a	1.40a	0.80a	0.90ab	1.00a	0.47a
Fresh seeds	0.27ab	0.63ab	1.23a	0.90a	1.23a	1.13a	0.50a

* Mean values with the same letters along the columns are not different significantly at $p \leq 0.05$

First Day of Germination: the result of analysis of variance showed that length of storage considered in the study did not significantly affect germination of *T. ivorensis* seeds (Table 2). Nonetheless, it was observed that seeds stored for 3 and 6 months commenced germination earlier (average of 22 days) than freshly collected seeds and the rest of storage treatments (Table 3).

Last Day of Germination: storage periods did not significantly affect the last day of germination (Table 2). Nonetheless, germination stopped earliest in seeds stored for 9 months (average of 31 days) while freshly collected seeds germinated till 32 days on the average, after sowing (Table 3).

Time Spread of Germination: there was significant effect of storage on time spread of germination of *T.*

ivorensis seeds. Seeds stored for 3 months had the highest value of 11 days while those stored for 9 and 12 months had the lowest (average of 8 days) (Table 3).

Germination Index (GI): it was observed that GI decreased with and was significantly affected by the length of storage. Freshly collected seeds had the highest GI mean value (1.10) while the lowest (0.62) was observed in seeds stored for 12 months (Table 3).

Coefficient of Velocity of Germination: this also followed similar trend with GI as the values decreased with length of storage (Table 3). Freshly collected seeds had highest mean value (238.94) and significantly differ from 9- and 12-month storage periods while those stored for 3 and 6 months were not significantly different. The least value for CVG was observed in 12 months of storage.

Table 2: Analysis of variance for the effects of storage periods on germination of *Terminalia ivorensis* seeds

Source of variations		Df	Sum of squares	Mean Square	F	Sig
First day	Storage periods	4	7.20	1.800	1.570	.213
	Error	25	28.667	1.147		
	Total	29	35.867			
Last day	Storage periods	4	6.667	1.667	2.083	.113
	Error	25	20.000	.800		
	Total	29	26.667			
Time of spread	Storage periods	4	35.200	8.800	6.875	.001
	Error	25	32.000	1.280		
	Total	29	67.200			
CVG	Storage periods	4	150374.528	37593.632	3.196	.030
	Error	25	294104.837	11764.193		
	Total	29	444479.366			
GI	Storage periods	4	1.275	.319	5.294	.003
	Error	25	1.505	.060		
	Total	29	2.779			

CVG: coefficient of velocity of variation; GI: germination index

Table 3: Mean effect of storage and soaking durations on selected parameters of seed germination of *Terminalia ivorensis*

Treatments		First day germination (day)	Last day germination (day)	Time spread of germination (day)	Germination index	Coefficient of velocity of germination
Seed Storage	Fresh	23a	32a	10a	1.10a	238.94a
	3 month	22a	32ab	11a	1.05a	214.80ab
	6 month	22a	32ab	10a	0.93a	210.19ab
	9 month	23a	31b	8b	0.63b	82.15bc
	12 month	23a	31ab	8b	0.62b	74.38c
Soaking duration (hours)	Control	22a	32a	10a	0.95a	192.55a
	12	22a	32a	10a	1.06a	238.34a
	24	23a	32a	9a	0.72a	94.73a
	48	22a	32a	10a	0.81a	134.08a
	72	22a	32a	10a	0.83a	157.38a
	96	23a	31a	9a	0.83a	146.36a

* Mean values with the same letters along the column are not significantly different ($p \leq 0.05$)

3.2. Effects of Water Soaking Durations on Seed Germination of *Terminalia ivorensis*

Overall germination percentage: figure 2 showed the overall germination percentage as affected by soaking

duration and the highest overall germination percentage (56%) was recorded in seeds treated with 12 hours water soaking duration while least (41%) was observed in 24 hours.

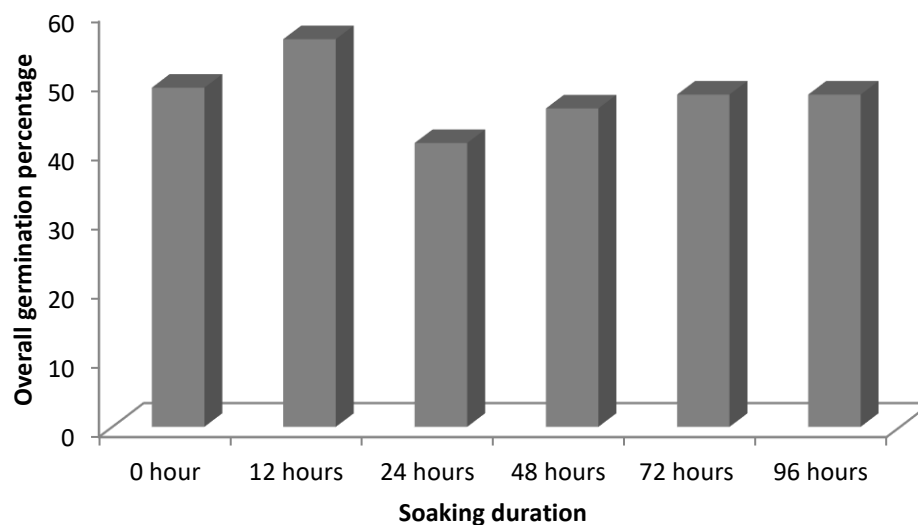


Figure 2: Overall germination percentage of *Terminalia ivorensis* seeds subjected to water soaking durations.

Mean Germination: seeds soaked for 24 hours had highest mean germination value (0.8) at 21 DAS but had least at 29 and 31 DAS (0.6 and 0.56 respectively). At 23 DAS, 12 hours soaking had highest mean germination value with 1.0 while 98 hours soaking had the least. At

25 and 27 DAS, 98 hours soaking and control recorded highest mean germination values. Seeds soaked for 48 hours and 72 hours recorded mean germination values at 31, 29 and 33 DAS (Table 4).

Table 4: Mean of seed germination of *Terminalia ivorensis* subjected to different water soaking durations

Period of soaking (hours)	Mean of <i>T. ivorensis</i> seed germination, days after sowing (DAS)						
	21	23	25	27	29	31	33
96	0.04cd	0.28c	1.32a	0.80b	0.92ab	1.00ab	0.20c
72	0.32ab	0.48bc	0.88a	0.64b	1.08a	0.76bc	0.56a
48	0.48a	0.44bc	1.08a	0.76b	0.88ab	1.08a	0.00d
24	0.80d	0.72ab	0.88a	0.80b	0.60b	0.56c	0.28bc
12	0.28abc	1.00a	0.96a	1.08ab	0.92ab	0.92ab	0.40ab
Control	0.16bcd	0.72ab	0.88a	1.36a	0.88ab	0.92ab	0.20c

* Mean values with the same letters along the column are not significantly different ($p \leq 0.05$)

First Day of Germination: the results of analysis of variance showed that soaking duration considered in the study did not affect germination of *T. ivorensis* seeds (Table 5). However, seeds soaked for 12, 48 and 72 commenced germination earlier (average of 22 days) than other treatments while 24 hours soaking commenced germination later than others (Table 3).

Last Day of Germination: soaking duration had no significant influence on last day of germination (Table 5). Seeds soaked for 72 hours germinated till 32 days while those soaked for 96 hours germinated till 31 days and 48 hours soaking germination stopped at 31 days (Table 3).

Time Spread of Germination: soaking duration had no significant influence on time spread but 12 and 48 hours of soaking had highest value of 10 days while seeds

soaked for 24 and 96 hours had least value of 9 days (Table 3).

Germination Index (GI): germination Index did not follow any trend or pattern as soaking duration had no significant effect on the parameter. Seeds soaked for 12 hours had the highest GI value followed by control (0.95) while 24 hours-soaked seeds recorded the least value of 0.72 (Table 3).

Coefficient of Velocity of Germination: soaking duration had no significant effect on CVG and no trend was followed. Table 3 showed that seeds subjected to 12 hours soaking recorded the highest value (238.34) followed by control (192.55) while 24 hours soaking had the least value (94.73).

Table 5: Analysis of variance for the effects of soaking durations on germination of *Terminalia ivorensis* seeds

Source of variations		Df	Sum of squares	Mean Square	F	Sig
First day	Soaking durations	5	10.267	2.053	1.925	.127
	Error	24	25.600	1.067		
	Total	29	35.867			
Last day	Soaking durations	5	4.267	.853	.914	.488
	Error	24	22.400	.933		
	Total	29	26.667			
Time of spread	Soaking durations	5	13.467	2.693	1.262	.312
	Error	24	51.200	2.133		
	Total	29	64.667			
CVG	Soaking durations	5	61585.640	12317.128	.763	.585
	Error	24	387404.245	16141.844		
	Total	29	448989.885			
GI	Soaking durations	5	.360	.072	.714	.619
	Error	24	2.419	.101		
	Total	29	2.779			

CVG: coefficient of velocity of variation; GI: germination index

4. DISCUSSION

Different approaches of breaking seed dormancy in order to enhance germination rate and increase the germination processes have been suggested by many authors (Airi *et al.* 2009). These include physical scarification of the seed coat by nicking (Azad *et al.* 2010), piercing with needles and knives, and or using abrasion paper (Schmidt, 2000). Method such as cold-water soaking can also be used to overcome physical seed dormancy (Sabongari and, 2003). Hossain *et al.* (2005) reported that seeds with hard and impermeable seed-coats were noted to establish germination after pre-sowing treatments. However, breaking of seed dormancy varies from species to species. Therefore, it is very important to determine method and condition suitable for germination of different plant species.

The results of seed germination of *T. ivorensis* for the first week germination commenced indicated that both freshly collected seeds and seeds stored for 3 and 6 months germinated well compared to seeds stored for 9 and 12 months. Kader (1998) explained that if lower FDG is obtained, it is an indication that initiation of germination in the seed lot is faster while lower LDG indicate a faster ending. From this study, seeds stored for 3 and 6 months-initiated germination faster than others but freshly collected seeds and seeds stored for 3 months continued to germinate till 33rd day after sowing making 3 month stored seeds to have highest TSG (11 days). TSG, GI and CVG were lower for 9 and 12 months when compared to other treatments. GI expresses germination percentage and germination speed of seed and the higher the GI the higher the vigour and germination ability of the seed lot (Bench *et al.* 1991).

In this study, higher GI values were observed in fresh seeds and those stored for 3 and 6 months. This trend was observed by Palomeque *et al.* (2020) in storage experiment carried out on *Cedrela Montana Weinmannia fagaroides* and *Oreocallis grandifolia* seeds stored for 12 months. This suggests that the maximum threshold for storing *T. ivorensis* is between 1-6 months after harvesting. This clearly indicated that *Terminalia ivorensis* seeds can be stored under a room temperature and remain viable for seedling production only within a period of 6 months as efforts to increase the seeds storage period may damage and reduce the seed germination potentials. Long term storage above a period of 6 months under an ambient condition may encourage infestation of pests which may damage the seeds by burrowing into the cotyledon eaten up the embryonic portion of the seeds. Asomaning and Sacandé (2019) opined that seed germination is affected by the time of storage as freshly collected seeds have the greater potential of producing maximum seedlings germination than storing for a longer

time. This is because the germinating embryo remains active and seed oil is still intact and not sealed up the germination pores of the seeds by the cotyledon (Airi *et al.* 2009).

Water soaking of seeds before sowing has been effective for germination of some seeds. At 21 DAS, seeds soaked for 96 hours and control started germination with low vigour which changed at 25 and 27 DAS respectively. Seeds soaked for 24 and 48 hours performed relatively well from 21 till 33 DAS where 48 hours soaking recorded zero germination. Soaking for 72 hours gained germination vigour at 23 DAS and performed well till 33 DAS. This suggests that soaking in water plays good role on germination of *Terminalia ivorensis* seeds. The result is in line with Owonubi *et al.* (2005) who opined that seed germination of some forest tree species respond well with soaking in water for some days.

5. CONCLUSION

1. This study has shown that seeds of *Terminalia ivorensis* harvested and stored for different periods respond differently during germination.
2. It was observed that both freshly collected seeds and seeds stored for 3 to 6 months germinated well.
3. Decline in germination was observed as period of storage increased to 12th months. Moreover, soaking influenced *T. ivorensis* seeds germination as soaking for 24 and 48 hours in water enhanced better germination though, other treatments did not perform poorly.
4. Sowing freshly collected seeds of *T. ivorensis* is ideal and germination will be attained with or without water pretreatment.

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