Original Research Article

Effects of Desiccants on Seed Quality of Three Tree Species in the Moist-Decidous Forest Ecotone

ABSTRACT

A study was carried out to determine the effects of seed desiccants on seed quality of three very important indigenous forest tree species. The experimental period was December, 2015 to February, 2016. Seeds were collected from the Bobiri Forest Reserve. Seed desiccation experiment was set up in a Complete Randomized Design (CRD) with three (3) replications. Germination percentage, seed vigour, 1000 seed weight, moisture content, seed health analysis, carbohydrate, protein and oil contents were determined before and after seed desiccation. The study revealed that the Zeolite beads® dried the seeds of *Pericopsis elata* within 2 days and 3 days for *Sterculia rhinopetala* but *Guarea cedrata* seeds were dried within 12 days. This rate of drying was much faster than the rest of the desiccants without any deleterious effect on seed quality. *P. elata* and *S. rhinopetala* showed orthodox seed storage behavior by surviving drying to a lower moisture content which can enhance their long term storability. *G. cedrata* seeds however, exhibited recalcitrant seed behaviour and lost viability significantly after desiccation. *G. cedrata* seeds unlike *P. elata* and *S. rhinopetala* cannot be dried to lower moisture contents and stored for longer period under ambient conditions.

Keywords: ***storage, orthodox, germination, conservation, recalcitrant***

1. INTRODUCTION (ARIAL, BOLD, 11 FONT, LEFT ALIGNED, CAPS)

Tree planting is undoubtedly known to be an effective measure to protect the climate and mitigate the effects of change. This is possible due to the role trees play in greenhouse gas carbon dioxide sequestration, counteract erosion and desertification among others [1]. There is a growing concern about the uncontrolled exploitation and d of trees especially indigenous species in the tropics that are threatened with extinction. Studies have shown tha plant species are in danger of extinction, while some have already become extinct [2]. On a global basis, the IU estimated that about 12.5% of the world's vascular plants, totaling about 34,000 species, are under varying deg threat. In Ghana, three indigenous trees of economic importance included *Peripcopsis elata* "Kokrodua" is class endangered species [3], *Sterculia rhinopetala* "Wawabima" and *Guarea cedrata* "Kwabohoro" have been described as vulnerable, according to the IUCN Red List of Threatened Species [4]. There is therefore an urgent need to conserve these species in any way practicable. This can be achieved by in situ or by ex-situ conservation technologies. One of the ex situ conservation of plant germplasm that is safe, effective and inexpensive is conventional seed storage. This method does not only maintain its viability but also its vigour without hampering the genetic makeup [2] In storage, the seed longevity is influenced by the seed moisture content, temperature and type of container used. Among these factors, the seed moisture content plays a significant role in determining seed longevity. There are various forms of drying methods that have been used for drying seeds of all kinds to reduce seed moisture content. Methods such as sun drying, forced air drying, modified solar drying [5] and desiccant drying [6,7]. Since seed is a material used for regeneration purposes, it must be dried in a manner that does not affect its germination and vigour during storage. To effectively conserve these

Comment [A1]: Effects of Desiccants on the Quality and Conservation of Seeds of Tree Species in a Ghanaian Forest. Just a suggestion.

Comment [A2]: Inform the appropriate location. City, country, geographical coordinates. The reader should be able to locate himself when reading his work, right from the start.

Comment [A3]: The first time it is mentioned, it is interesting to insert the name of the author and family to which it belongs.

Comment [A4]: I suggest that you first introduce species, and not start talking about them directly. We still do not know in which species the desiccants were used. First, introduce us, then talk about the outcome.

Comment [A5]: It is good but lacked a more specific conclusion on the use of dessicants in the four species. What is the main conclusion? This is still a result.

Comment [A6]: Good choice. But I suggest putting some word synonymous with forest

Comment [A7]: Very well written! But I suggest talking about the main characteristics of the studied plant species. Wood use, non-timber, growth, social value ... Do not extend much, but only the character of importance, showing us the reason for choosing the trees of your research. Also speak about which are orthodox, which are recalcitrant. Figures are welcome tropical tree seeds, it is essential that we have basic knowledge about their seed drying sensitivity, seed physiology, responses to desiccation and their storage potential. Desiccant drying in a closed container is often suggested as a low-technology method to reduce the moisture content of seed germplasm. Suitable desiccants include silica gel (sodium silicate), lithium chloride, calcium chloride, molecular sieve, charcoal and rice which have been widely used on agricultural seeds with quite an appreciable success[7, 8, 6]. However, there is little information known on how the desiccants perform on tree seed species, particularly the tropical species. This study was aimed at drying the seeds of these tropical tree species to lower moisture using different desiccants for subsequent storage.

2. MATERIAL AND METHODS

2.1 Seed Collection and seed desiccation experiment.

The seed samples were collected from the Bobiri Forest Reserve in December, 2015. This Forest Reserve is located in the south-east sub-type of moist semi-deciduous (MSSE) forest in Ghana, covering an area of about 5,445 ha [9]. It is located on the main Accra - Kumasi Highway at the village of Kubease, about 30 kilometres (19 miles) from Kumasi. It is about 25 minutes' drive from the Kwame Nkrumah University of Science and Technology (KNUST). The Reserve was created in 1931 and has an area of 54.65 km$^{2.}$ After seeds were collected, they were put in plastic seed bags, tightly sealed and sent to the experimental station of the Department of Horticulture, Kwame Nkrumah University of Science and Technology, Kumasi, Ghana. The seed desiccation and other laboratory experiments were conducted at the Department of Horticulture, KNUST.

Comment [A8]: Insert in Abstract. Is it possible to make a simple map of the study area, or enter the geographical coordinates, or both? If yes, better

2.2 Experimental Procedure

The seed to desiccant ratio used was 1:1. 100g each of the seeds of the three species were weighed using an electronic scale and put in an airtight transparent plastic container. 100g each of the desiccants were weighed, put in gauze and held above the seeds in the container to prevent the desiccants from having direct contact with the seeds. The treatments were laid in a completely randomized design and replicated three times. The desiccants used were Zeolite Bead®, Charcoal, Biochar, Paddy rice and no desiccant (as control).

Comment [A9]: I believe you made a Factorial 3 x 5. Try to run your data in a factorial and see what information it will have as a result.

2.3 Data Collected

Data collected include time taken (days) for seeds to be completely dried,1000 seed weight (g), seed germination percentage (SGP), seed vigour (relating to total leachates) and seed moisture content (%) determined according to ISTA Rules, 2007[10]. The chemical seed composition; percentage oil, moisture content, protein and carbohydrate were determine using the rules as set out in AOAC, 2007 [11]. The Seed Vigour Index (SVI) was determined according to the formula proposed by Abdul-Baki and Alderson (1973) as: Seed Vigour Index = (Shoot length + Root length) X Germination Percentage [12]. Data collected from the laboratory experiments were subjected to analysis of variance using Statistix Student Version 9.0. Tukey's HSD (Honest Significant Difference) was used for mean separation at probability level 0.01.

3. RESULTS

3.1 Seed initial quality characteristics

There were significant differences among the treatments for seed moisture content, seed vigour, seed vigour index, thousand seed weight and germination percentage (Table 1). *Guarea cedrata* had the highest moisture content (27%) and thousand seed weight (1089.7g). On the other hand, *Pericopsis elata* recorded significantly the highest vigour index (2689.7) but the least moisture content (7.5%) and thousand seed weight (254.67g). There were significant differences ($p \leq 0.01$) between the treatments for germination percentage. *P. elata* recorded significantly the highest germination (96%) percentage followed by *S. rhinopetala* (95%). There were however, no significant differences ($p \leq 0.01$) between the treatments for seed vigour (Table 1).

Table 1. Initial seed quality characteristics of *G. cedrata, S. rhinopetala* and *P. elata*

Species	Moisture Content %	Vigour ($\mu S\ cm^{-1} g^{-1}$)	Vigour Index	1000 SW (g)	Germination (%)
P. elata	7.5	23.0	2689.7	254.7	96.3
S. rhinopetala	10	22.5	2376.7	779.7	95.4
G. cedrata	27	25.4	2251.7	1089.7	90.7
HSD (0.01)	3.7	4.36	27.96	5.59	3.66

Comment [A10]: Although I have entered the p-value, I suggest inserting the "letters" of the Tukey test for better understanding.

3.2 Seed initial proximate composition

There were significant differences between the treatment for P. elata, S. rhinopetala *and* G. cedrata. P. elata *recorded the highest seed oil (31.25%) and protein (37.41%) contents but the least carbohydrate (1.93%) content. The least oil (23%) and protein (9.1%) contents were recorded by* G. cedrata *but recorded the highest carbohydrate (19.43%) content.*

Table 2: The initial proximate composition of the three tree species

Species	Oil (%)	Protein (%)	Carbohydrate (%)
P. elata	31.3	37.4	1.9
S. rhinopetala	23.0	19.2	17.4
G. cedrata	13.5	9.1	19.4
HSD (1%)	10.85	3.23	3.81

3.3 Number of days taken for seeds to attain dryness

There were significant differences between the treatments for the number of days taken for each of the seeds species to attain dryness at a moisture content of 3.5% for all the species (Table 3). It took 2 and 3 days for the Zeolite Bead® to dry *P. elata* and *S. rhinopetala* significantly less in time than the other desiccant treatments. The number of days to dry the same species using charcoal or Biochar was not significantly different. The longest time for the attainment of dryness was experienced under the control treatment (no desiccant) and use of Rice (paddy) (13 to 82 days). The rice desiccant treatment took 6.5 times more days than the Zeolite beads to dry *P. elata* (Table 3). It took 12.3 days for the Zeolite Bead® to dry *G. cedrata* to steady moisture content, significantly less in time than the other desiccant treatments. It however, took 37 and 39 days respectively using charcoal and biochar to dry the same species under the same conditions. The number of days increased further to 82 when rice was used as a desiccant or no desiccant was applied.

Table-3

	Number of Days		
Desiccant/Species	***P. elata***	***S. rhinopetala***	***G. cedarata***
Zeolite Bead®	2	3.3	12.3
Charcoal	6	9.8	36.8
Biochar	6.3	9.8	38.6
Rice (paddy)	13	21.1	79.6
No desiccant	13.5	21.9	82.7

HSD (0.01)	***3.55***	***3.55***	***3.55***

Comment [A11]: I suggest to present this data in a graph

3.4 Proximate composition, vigour and 1000 seed weight of the species

For all three seed species, there were no significant differences between the treatments for the constituents of the proximate composition. For *P. elata*, carbohydrate content ranged from 1.21% to 1.25%; oil content ranged from 31.52% to 31.58% and protein content ranged from 38.06% to 38.08%. There were also no significant differences between the treatments for seed vigour such that it ranged from 24.56 μS $cm^{-1}g^{-1}$ to 24.65 μS $cm^{-1}g^{-1}$. For *S. rhinopetala*, carbohydrate content ranged from 16.53% to 16.95%; oil content ranged from 23.32% to 23.68% and protein content ranged from 20.55% to 20.57%. There were also no significant differences between the treatments for seed vigour such that it ranged from 26.08 μS $cm^{-1}g^{-1}$ to 26.14 μS $cm^{-1}g^{-1}$. *G. cedrata,* carbohydrate content ranged from 18.07% to 18.28%; oil content ranged from 6.52% to 6.58% and protein content ranged from 11.06% to 11.08 %. There were also no significant differences between the treatments for seed vigour such that it ranged from 27.23 μS $cm^{-1}g^{-1}$ to 32.43 μS $cm^{-1}g^{-1}$.

Comment [A12]: Standardize the way you speak of the species. Previously you write the full name

Table 4: effect of desiccants on the proximate composition, vigour and 1000 seed weight of the three species

Species	**Desiccants**	**Carbohydrate %**	**Oil %**	**Protein %**	**Vigour μS $cm^{-1}g^{-1}$**
	Beads	1.25	31.58	38.07	24.58
	Charcoal	1.22	31.57	38.06	24.64
P. elata	Biochar	1.23	31.56	38.08	24.56
	Rice	1.24	31.53	38.06	24.60
	No desiccant	1.21	31.52	38.08	24.64
	HSD (1%)	1.77	8.92	5.42	0.18
	CV (%)	4.65	7.98	4.01	0.21
	Beads	16.53	23.38	20.56	26.08
	Charcoal	16.73	23.37	20.55	26.14
S. rhinopetala	Biochar	16.33	23.63	20.57	26.09
	Rice	16.93	23.33	20.55	26.1
	No desiccant	16.95	23.32	20.57	26.14
	HSD (1%)	5.4	8.92	5.41	0.18
	CV (%)	9.15	10.78	7.43	0.2
	Beads	18.28	6.58	11.07	32.43
G. cedrata	Charcoal	18.28	6.57	11.06	30.63
	Biochar	18.18	6.56	11.08	29.63
	Rice	18.07	6.53	11.06	27.53

Comment [A13]: I keep the suggestion to enter the letters of the statistical test, for better visualization of your data.

No desiccant	18.22	6.52	11.08	27.23
HSD (1%)	5.4	8.92	5.42	5.4
CV (%)	8.39	8.39	13.79	5.18

3.5 Effects of desiccants on 1000 seed weight (g) for the tree species

For all three seed species, there were significant differences in the treatments for 1000 seed weight (Table 5). For *P. elata,* the highest weight was recorded by the control and the least weight was recorded for Zeolite bead. *S. rhinopetala,* the heaviest seeds were recorded for biochar, rice desiccants and no desiccation treatment. For *G. cedrata*, the highest weight was recorded by Zeolite bead and the least was recorded by the control.

Table 5: Effects of desiccants on 1000 seed weight (g) for the tree species

	1000 Seed Weight (g)		
Desiccant/Species	***P. elata***	***S. rhinopetala***	***G. cedarata***
Zeolite Bead®	254.3	781.0	1099.0
Charcoal	257.0	781.4	1098.9
Biochar	258.0	781.7	1098.1
Rice (paddy)	258.8	782.0	1097.1
No desiccant	258.9	768.1	1094.4
HSD (0.01)	**3.57**	**0.74**	**3.58**

3.6 Effects of desiccants on germination (viability) of *G. cedrata* after desiccation

Comment [A14]: Try to put this information in a single chart and see if it gets better the view. I believe that the information will be passed more clearly, in a more dynamic way, in addition to reducing space.

There were no significant differences among the beads on germination percentage of *G. cedrata* seeds after desiccation but the highest germination was recorded by rice (12.33%) and the lowest was recorded by beads (8.32%) as shown in Table 6.

Table 6. Effects of desiccants on germination (viability) of *G. cedrata* after desiccation.

Dessicants	Germination %
Beads	8.32
Charcoal	10.31
Biochar	11.42
Rice	12.33
No dessicant	9.20
HSD (0.01)	5.42

3.7 Effects of desiccants on germination (viability) of *S. rhinopetala* after desiccation.

There were no significant differences among the beads on germination percentage of *S. rhinopetala* seeds after desiccation but the highest germination was recorded by beads (88.30%) and the lowest was recorded rice (85.33%) as shown in Table 7.

Table 7. Effects of desiccants on germination (viability) of *S. rhinopetala* after desiccation.

Dessicants	Germination %
Beads	88.30
Charcoal	88.10
Biochar	87.33
Rice	85.33
No dessicant	86.20
HSD (0.01)	5.30

3.8 Effects of desiccants on germination (viability) of *P. elata* after desiccation.

There were no significant differences among the beads on germination percentage of *P. elata* seeds after desiccation but the highest germination was recorded by beads (95%) and the lowest was recorded by no desiccant (92%) as shown in Table 8.

Table 8 Effects of desiccants on germination (viability) of *P. elata* after desiccation.

Dessicants	Germination %
Beads	95.00
Charcoal	94.00
Biochar	93.00
Rice	93.00
No dessicant	92.00
HSD (0.01)	3.55

4. DISCUSSION

The differences observed in the initial seed quality could be attributed to the high genetic variations that existed between the species. Seeds of *P. elata* and *S. rhinopetala* were shed with relatively lower moisture contents of 7.5% and 10%, respectively, which is characteristic of orthodox seeds. According to Berjak and Pammenter (2004), viability of orthodox seeds can be maintained even when the moisture content is reduced and can also be dried further to enhance their longevity [13]. The results of the present study showed that *P. elata* and *S. rhinopetala* seeds could remain viable for a long period of time when moisture was reduced. *G. cedrata* seeds, however were shed at very high moisture content (27%) and the seeds were metabolically active and also recorded high germination which is characteristic of recalcitrant seeds. Hay (2003) reported that recalcitrant seeds are metabolically active and would have high germination capacity when planted immediately after seed collection [14]. The results of the present study clearly confirm that *G. cedrata* had an initial high seed moisture but with a high initial germination probably showing recalcitrant seed storage behaviour.
The initial vigour index was highest (2689.7) whilst the initial vigour (in terms of solute leakage) were low and within the recommended leakage levels as reported by Milosevic et al., (2010) that seeds with leakage below 25 µS $cm^{-1}g^{-1}$ were of high vigour whilst those with vigour more than 35 µS $cm^{-1}g^{-1}$ were of low vigour [15].

The Zeolite Bead® were significantly able to dry the seeds at a faster rate as compared to charcoal, biochar, rice and the control. This could be attributed to the presence of aluminum silicates that fill the micropores which have high affinity to hold water in these micro molecular pores for a longer duration. The results of the current study confirms the findings of Nassari *et al.* (2014) who investigated the drying ability of beads on the quality of tomato seeds and reported that the beads were significantly effective to reduce absorb seed moisture at the fastest rate [16]. Hay *et al.* (2012) also reported on the advantages of using the beads as desiccant including their greater affinity for water, especially at low humidity; more rapid drying; and no hysteresis effect, which lowered the amount of water that could be adsorbed after regeneration [6]. Buady (2002) reported that charcoal was a good drying agent and was found to keep stored seeds viable quite better as compared to dried rice used as a desiccant [17]. Moreover, Nyarko (2006), indicated that rice was a poor desiccant as compared to charcoal just as was found in the present study [18]. Additionally, for *P. elata* and *S. rhinopetala*, the desiccants did not have any deleterious effect on the vigour (solutes leakage), vigour index, germination percentage, seed protein, oil content and carbohydrate. This could be due to the fact that the two species are orthodox seeds and that desiccation to a lower moisture content rather improved viability thereby confirming Harrington's principle that for every 1% reduction in seed moisture there was a doubling of the viability of the seed [19]. McDonald, (2004) also reported that desiccation-sensitive seeds cannot be dried to lower moisture content without deleterious effect on viability as compared to desiccation-insensitive seeds [20].

The deleterious effects of desiccation on *G. cedrata* seeds which was evident in the significantly reduced germination percentage, confirmed their high sensitivity to drying. According to Pritchard (1991), seeds that are desiccation-sensitive lose their viability considerably after dehydration [21]. Hoekstra *et al*., (2001) also indicated that desiccation results in reduced cellular volumes and causes the compaction of cytoplasmic components [22]. This compaction increases molecular interactions leading to protein denaturation and membrane fusion. Furthermore, Chin (1988) opined that death of recalcitrant seeds was due to reduction in moisture and was basically due to the loss of membrane integrity and nuclear disintegration [23]. The results of the present study for *G. cedrata* confirm these findings.

Comment [A15]: Very well! Speaking of the advantages and disadvantages of using desiccants you act with impartiality. Well written.

5. CONCLUSION

Results obtained from this study has shown that among the four desiccants used in drying *P. elata, S. rhinopetala* and *G. cedrata,* beads had the fastest drying time without any deleterious effect on the physical and chemical properties of seeds. *G. cedrata* seeds lost viability considerably after desiccation and therefore could not be stored.

Comment [A16]: Good. I missed that in the abstract.
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COMPETING INTERESTS

"Authors have declared that no competing interests exist.".

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Comment [A17]: Only 13.04% of the references are based on the literature available in the last ten years. I suggest updating. Not forgetting the classics (pioneering publications).

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