EFFECTS OF SEED SIZE AND PHYTOHORMONE ON THE GERMINATION OF SEEDS OF TWO FOREST TREE SPECIES, *Prosopis africana* Guil AND *Dialum guineensis* Wild.

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Abstract

Studies were conducted on the effect of seed sizes and phytohormone (GA_3) on seed germination of two savanna tree species: *Prosopis africana* (Guil) and *Dialum guineensis* (Wild). The selection of the seeds was based on the role they play in the savanna community and nutrient recycling of soil. The trees, like many others are fast going into extinction. The seeds of these species were found to be dormant while the seedlings also have high mortality rates. The two tree species produce two seed types, large and small. The Gibberellic acid at 3000mg/l had significant effect ($P \leq 0.05$) on seed germination in both *P. africana* and *D. guineensis*. The large and small seeds of two tree species evaluated had up to 80 and 30 to 50% germination; respectively. Termination of hard seed coat was obtained under 10-15 minutes treatments, and gave up to 70-80 percentage germination in *D. guineensis* and *P. Africana* seeds. Gibberellic acid at 1000mg/L-3000mg/L gave maximum percentage seed germination of 60-80% while untreated seeds did not germinate at all. The experimental design used was the randomized block type. All treatments were in five replicates.

Introduction

The Nigerian savannas contain some general utility tree species including *Sena spp, Parkia biglobossa, Afzelid Africana, Daniella oliveri* and *Vittellaria paradoxa* which are faced with many problems including annual bush burning and over exploitation, hence the need for continuous attention to solve the problems (Agboola, 1991; Agboola 1995). Some scarification methods such as the use of concentrated sulphuric acid, dry heat and wet heat treatments have been known to improve seed germination. However, the choice of chemical scarification agent for a given species is influenced by seed size, seed coat thickness and seed susceptibility to oxidative damage (Rosner *et al.*, 2002).

The genus *Prosopis* include about 44 species among which *P. farcata, P. alba, P. chitensis, P. cineraria, P. juliflora, P. pallida, P. tamaruye* found mainly in the arid regions of the world. *Prosopis africana* is a perennial tree legume which belong to the family leguminosae and sub-family mimosoidae,class-magnolipsoida.This species is a small to large tree with 4-2m tall and up to 1min girth with an open canopy and dropping foliage resembling *Tamarindus indica. P.africana* is the only native to intertropical Africa.In Nigeria, the trees are found between latitude 7^0N and 10^0N(Agboola,1995).*P.africana* is frequently found in fallow land,on various textured and lateritic soils.There are two varieties,one with narrow cylindrical pods (2.5cm in diameter) and one with broader flattened pods(3cm in width).Pods remain on the tree long after maturity.

Dialum guineensis is the commonest and most widespread species in Nigeria. The small black velvety fruits are very conspicuous and distinctive. The plant is from the class Leguminosae. The seed which is rich in vitamin C,can be used to supplement the content in food. The tree is used in making furniture and other household equipment,since the wood responds to polish. The distribution of the plant extends from Senega to Nigeria, also occurring in the Island of Sao Tome (Keay,1989). The plant occurs most in the forest and forest outliers in savanna agroecological zones of the country. The tree is about 20m high, but often shrubby with a densely leafy crown that could serve as shade. The tree could also serve as wind-break against heavy storm. The bark is smooth, grey, yielding red gum, sapwood white and with distinct ripple marks (Keay, 1989). In most African countries, *P. africana* and *D. guineensis* are used for diverse purposes such as carving pestle and mortar, furniture, angled hoe from stems with the auxiliary branch, tobacco pipes. In Europe, the woods are used for cabinet works. They are also used to manufacture carpentry tools, turnery and planes among others. Charcoals for cooking and various

commercial and domestic uses are made from the tree. In Northern Nigeria, several forms of single membrane drums are made from the trees.

There is little or no known evidence of propagation of *P. africana* and *D. guineensis* seeds by indigenous people of Southwest Nigeria. These species grow wild in the Guinea savanna in Nigeria. The information obtained showed that the farmers find it difficult to propagate the plant through the seeds.

In an attempt to enhance seed germination and seedling growth in economic plants of savanna agroecological zones, consideration had been given to seed sizes and hormone treatments. The plant growth hormones that have been associated with seed germination and seedling physiology included Gibberellic acid (GA_3), Indole Acetic Acid (IAA), Ethylene, Kinetin/Cytokinine. (Agboola, 1998; Fasidi *et al.*, 2000).

The present studies have focused on what effect would seed size differences have on seed germination of forest tree species.

Materials and Methods

Fruit collection and seed processing

Pods of *P. africana* seeds were collected directly from the tree stands after fruit fall in December 2007 from the savanna vegetation within the campus of the University of Ilorin, Ilorin (8.32^{oN} and 4.34^{oE} latitude) Nigeria. Seeds of *D. guineensis* were collected from seeds store of Federal Forestry Research Institute Nigeria (FRIN) Jericho, Ibadan, Nigeria. (7^0N and 4^0E longitude).

The seeds of *P. africana* were obtained by cracking the pods with a stone of moderate size while those of *D. guineensis* were obtained by soaking the fruit overnight and washed. The seeds were hand-picked while the damaged ones discarded.

Preliminary germination studies

Viability test of seeds were carried out using the floating method of ISTA. Fresh seeds were surface-sterilised with 0.1% mercuric chloride solution for 30 seconds and rinsed several with distilled water. About twenty seeds were placed in 9cm diameter Petri dishes on filter papers moistened with distilled water. All the germination trials were done at room temperature of 30 ± 2°C in five replicates.

Effect of chemical scarification treatments on germination:

Ten seeds were divided into lots and immersed in 100% Conc. H_2SO_4, for the period of 5, 10, 15 and 20 minutes before placement on filter paper in petridishes. Untreated seeds were used as control. Seeds were observed for germination while percentage germination was recorded. The experiment was subjected to completely randomized design (CRD).

Effect of seed size and chemical scarification on germination:

Linear scale and thread were used to measure the seeds of *P.africana* and sorted into small (1.1x0.6cm) and large sizes (0.7x0.5cm).While that of *D.guineensis* ranges from 0.7x0.6cm and 0.6cmx0.8cm for small and large seeds respectively, using the same method of measurement. The seeds were then scarified chemically, as stated above before preparing for germination.

Preparation of Phytohormone (GA_3)

The GA_3 of 100, 200, 300mg was dissolved into 50mls of 95% ethanol in 250mls volumetric flask. The solution was marked up to 100ml by adding distilled water to give solution of 1000mg/l, 2000mg/l and 3000mg/l concentration. A solution of 95% ethanol solution with distilled water mixture was used as control. In order to prepare GA solutions of 1-5ppm,0.01g of GA_3 was dissolved in 50ml distilled water to give stock solution of 200ppm.This was used to prepare GA solutions of 1ppm,2ppm,3ppm,4ppm and 5ppm concentration by measuring 0.25ml,0.50ml,0.75ml,1.00ml and 1.25ml respectively into measuring cylinder and adding distilled water to make up to 50ml. Scarified seed lots were moistened in 1000, 2000 and 3000mg/l of GA_3 solution and slated for germination in Petridishes. Untreated seeds served as control.

Results

A pod of *P. africana* contains about 14-15 seeds and a pulp of *D. guineensis* contains 1-2 seeds. The *D. guineensis* seeds appear in two distinct sizes (0.7 x 0.6cm for small seeds and 0.6cm x 0.8cm for large seeds). The *P. africana* seeds also occur in two distinct sizes (1.1 x 0.6cm for small seeds and 0.7 x 0.5cm for large seeds).

Table 1: Effect of 98% Conc. H_2SO_4 on dormancy breaking in *P. africana* seed

Days to germination	Level of treatments				
	5mins	10mins	15mins	20mins	Control
2* germination	10.0	0.0	40.0*	0.0	0.0
4*	10.0	20.0	50.0*	0.0	0.0
6*	10.0	20.0a	50.0a	0.0	0.0
10*	10.0	20.0a	80.0a	0.0	0.0
SDV ±	15.81	3.87	10.00	12.24	12.24
SEM ±	7.07	1.94	5.78	4.31	4.31

Table 2: Effect of 100% Conc. H_2SO_4 on dormancy breaking in *D. guineensis* seed

Days of germination	Level of treatments				
	5mins	10mins	15mins	20mins	Control
2* germination	10.0a	10.0	20.0	0.0	0.0
4*	20.0a	30.0	50.0	0.0	0.0
6*	20.0a	30.0	60.0	0.0	0.0
10*	20.0a	30.0	70.0	0.0	0.0
SDV ±	3.87	20.00	2.21	12.24	12.24
SEM ±	9.94	11.55	15.00	4.3	4.31

SDV = standard deviation
SEM = standard error mean

Immersion of seeds in concentrated H_2SO_4 for the period of 15mins resulted in the highest germination of 80% in *P. africana* and 70% in *D. guineensis* at 10 days after treatment (DAT) (Table 1).Although there was no germination when the seeds of the two species were immersed in H_2SO_4 for 20mins.Through out the experiment,20 and 30% germination were recorded for seeds of *P.africana* and *D. guineensis* immersed in H_2SO_4 treatments for 10min at 10 DAT (Table 1). Germination was 0% in control experiment all through (Table 1).
Germination percentage for *D. guineensis* seeds was 60-70% at 15 minute treatment within 6-10 DAT (Table 2).Having 10-30% germination as the least percentage germination for treatment under 5-10min within 2-10 DAT. Whereas, under 15min treatment of concentrared H_2SO_4, 50% germination was recorded and observed for 4 days.However,untreated seedswhich serve as control gave 0% germination for all the days of the experiment (Table 2).

Fig. 1 shows the effect of seed sizes on the germination of *P. africana* seeds. The large seed size had highest percentage germination of about 80 to 90% between 12 and 20 DAT.The least germination occurred at 4-5 DAT while about 60% germination percentage was recorded for the seed at 10 DAT (Fig. 1). Untreated large seeds which served as control did not germinate.The small seed-size of P.*africana* initially had a better germination performance than the large-size(30%),within the first 5DAT.The highest germination attainable did not however, exceed 70% for the small seeds(Fig.1). Untreated small seeds did not germinate. The large seeds of *D. guineensis* had the highest germination of 80% in 10 DAT (Fig.2).Germination was low earlier on (4-8) days of germination (Fig. 2). Small seed-size of *D. guineensis* also had a better germination performance (30%) at 1-4 days. The control experiment did not germinate.

Gibberellic acid (GA_3) effected 70% percentage germination at 3000mg/l in *P. africana* seeds within 20-25 days (Fig. 3). At 2000mg/l concentration, GA_3 germination effect on *P africana* seeds was 50% within 5-25 days (Fig.3). Fig. 4 showed the significant effect of GA_3 at 3000mg/l, with germination at 50% in *Dialum guineensis* seeds within 25 days (Fig.4). Untreated seeds showed 0% germination.

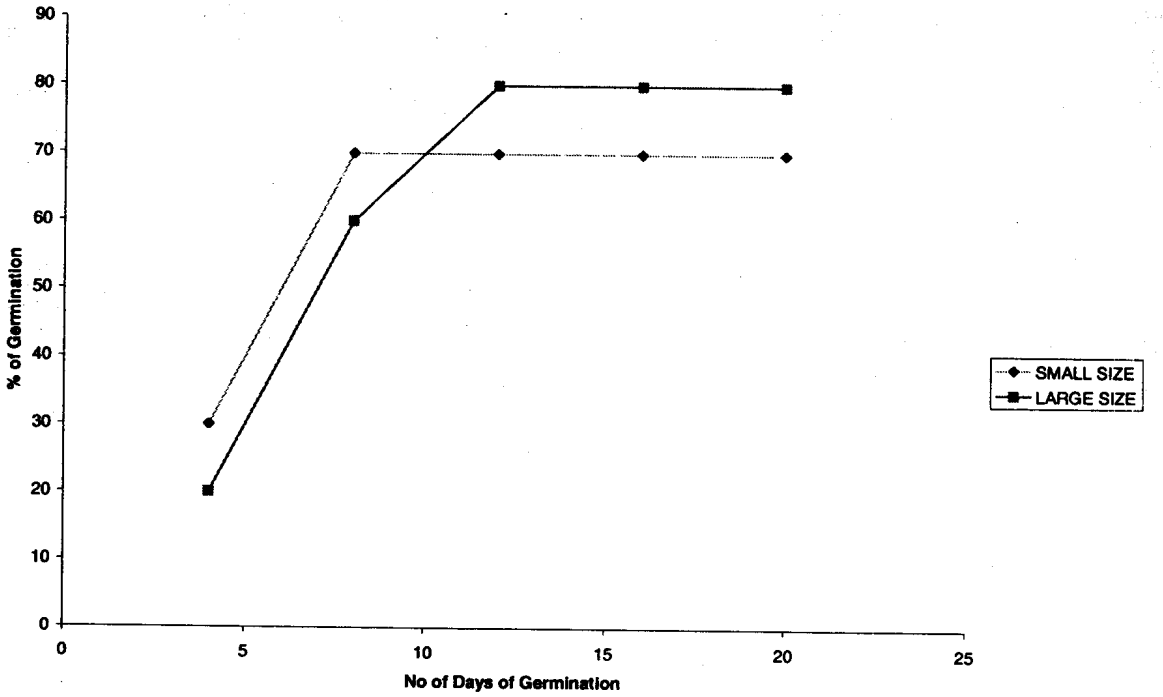

Fig 1 Effect of Seed Sizes on Germination of *P .africana* Seeds

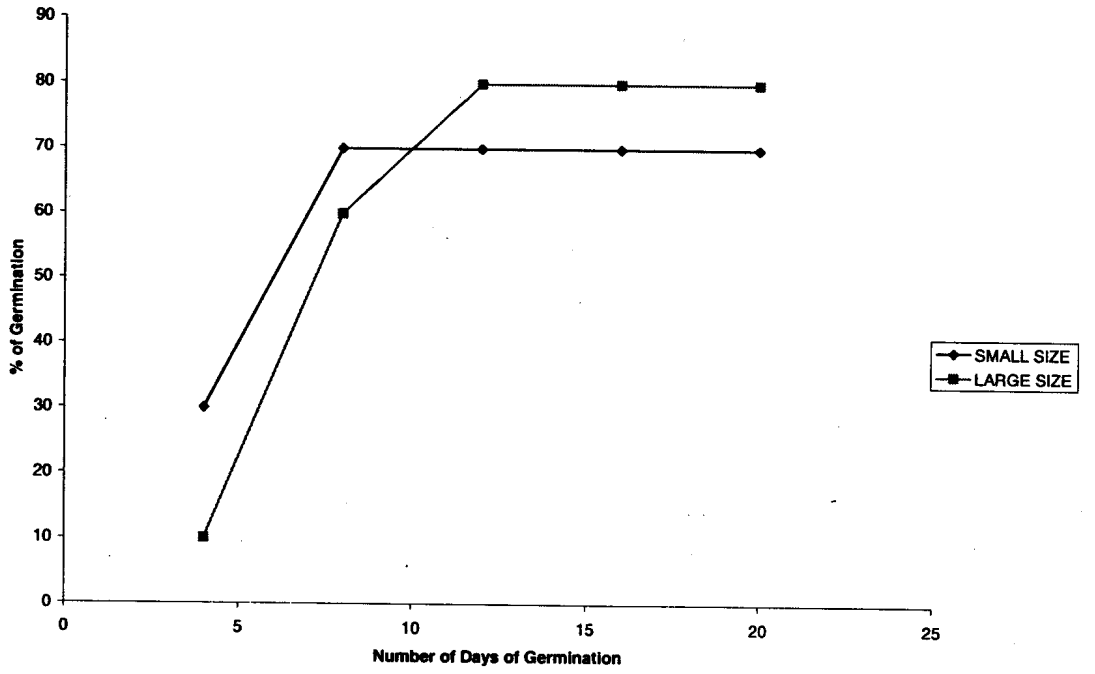

Fig 2 Effect of Seed Sizes on Germinatioin of *D .quneessis* Seeds

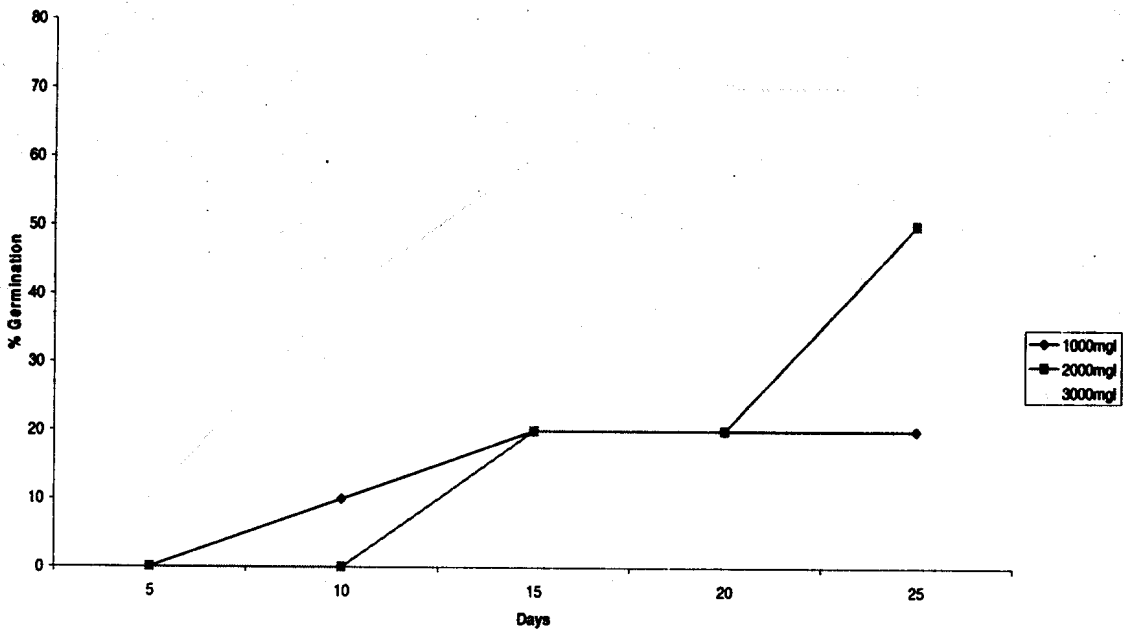

Fig 3 :Effect of Gibberellic Acid (GA_3) on Germination of *P. africana* Seeds.

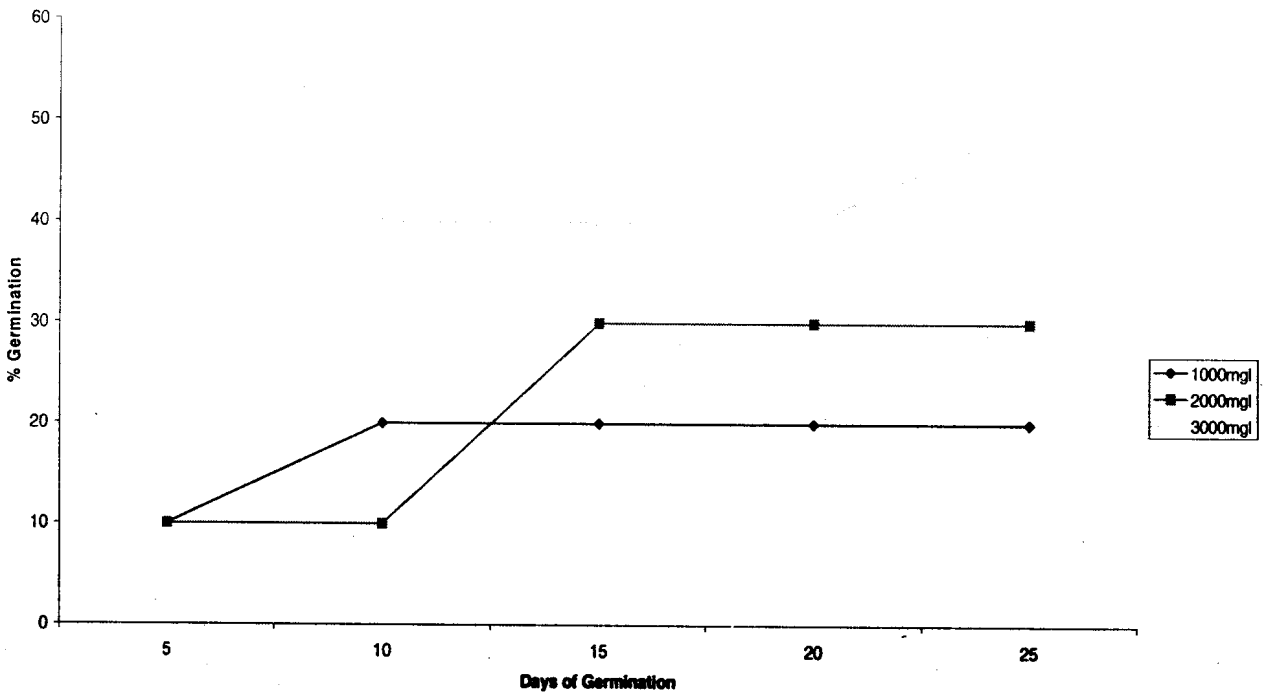

Fig 4 : Effects of GA_3 on Germination Of *D. guineensis* Seeds

Discussion

Prosopis africana and *Dialum guineensis* have two seed types based on observable size difference. It was observed in the study that large seed had higher germination percentage in both species. Occurrence of colour and size types in seeds of some tropical and savanna trees such as *Parkia biglobossa, Uraria picta* and *Leucaena leucocephala* are well documented by Etejere *et al.*, (1982) and Agboola, *et al.*, (1992, 1995). The observable differences in sizes may be due to genetic attributes, genetic constitution and size of the embryo. The seeds of *P. africana* and *D. guineensis* exhibit physical dormancy due to hard seed coat. This was terminated using chemical scarification method which involved the soaking of the seeds in concentrated H_2SO_4 for various periods which include 5, 10, 15 and 20 minutes before planting.
This result showed that seeds soaked for 15 minutes had the highest germination percentage of 80 and 70% at 10 DAT (Table 1) for *P. africana* and 70% in *D. guineensis* respectively (Table 2).

Chemical scarification has been used by previous researchers to break seed coat dormancy in various plant such as *Ceiba petandara, Cercocarpus montamis, Centrosema pubescens, Haranyana madagascariensis, Albezia lebbeck, Tamarindus indica* and *Parkia biglobossa* (Etejere *et al.*, 1982, Agboola, 1995). Hard seed coatings have been found to be impervious to water and gases. The proper enzymatic actions and proper mobilization of food materials for growth of the embryo are hampered due to the impervious nature of their seed coats. However, seeds of *P. africana* and *D. guineensis* germinate readily after reducing the thickness of the seed coats by chemical scarifications.

Gibberellic acid had significant effect ($P \leq 0.05$) on the germination of *P. africana* and *D. guineensis* especially at 3000mg/L (Fig. 3 & 4). The maximum percentage germination with the treatments was 70%.
Gibberellic acid (GA_3) is one of the major plant hormones involved in the control of processes of mobilization of food reserves from the endosperm or cotyledon (Black, 1972). Hence acceleration of the rate of germination by 3000mg/l GA_3 in the seeds was due to the unhindered entry of GA_3 into the seed following the softening of the seed coat by acid scarification. The absorbed GA actual complimented the inherent hormone in the seeds. Gibberellic acid has been reported to affect various processes in germinating seeds, including metabolism of amino-acids and respiration, increase amylase content (Stuart and Cathey, 1961). This probably explains improved germinability and peeling off of hard testa observed in *P. africana* and *D. guineensis* seeds treated with gibberellic acid.
The seed size had significant effect on seed germination. Large-size seed of *D. guineensis* seeds showed had higher germination percentage of 80 than the small size seeds with 60%. In contrast, with the seed size of *Cassia siamea* and *D. biglobossa* favoured by small-size seeds. Agboola (1996) found that seed size had significant effect on growth performance of seedling of *Gmelina arborea.* Seedlings emerging from larger seeds may enjoy a lot of food supply from the larger cotyledons of their seeds than those from small seeds (Agboola, 1996). However as soon as seedlings from both seed sizes start to photosynthesise, the favourable effect of large size seeds with larger cotyledons would have been diminished (Ebofin, 2003).

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