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GERMINATION POTENTIALS OF *Synsepalum dulcificum* (Schumach. and Thonn.) DANIELL SEEDS TO PRE-TREATMENT METHODS

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ABSTRACT

The study was carried out to determine the germination intensity of *Synsepalum dulcificum* seeds to various pre-treatment methods. This research was carried out at the forest nursery of the Federal University of Agriculture Abeokuta, Ogun State, Nigeria. This present study aimed to determine the most effective mechanism of breaking dormancy to enhance germination intensity in seeds of *Synsepalum dulcificum* using mechanical scarification at three point on seeds namely microplye, distal end and around the circumference. Also, seeds were soaked in water of temperatures of 20°C, 40°C and 80°C for 10, 30 and 60 mins respectively, in 5,10 and 15% H_2SO_4 acid for 5, 10 and 15 mins respectively and, in GA_3, IAA and IBA hormones of 0.01, 0.02 and 0.03ppm. Obtained data was analyzed using ANOVA and Fisher's Least Significant Difference was used to separate significant means. Results showed that highest germination percentage was obtained in seeds mechanically scarified around the circumference (50%) which was not significantly different (p>0.05) from the control. Seeds soaked in 20°C water for 30 min was significantly (*p<0.05*) higher than other treatments in category with germination percentage of 62.5% while highest germination were also obtained in seeds soaked in 5% H_2SO_4 for 5mins (31.25%) and seeds soaked in 0.02 ppm GA_3 (62.5%). The study concludes that *S. dulcificum* seeds will not have a problem of germination with the presence of suitable germination conditions.

Keywords: *Synsepalum dulcificum*, seeds, pre-treatment, germination intensity

INTRODUCTION

Seed dormancy is one of the most limiting factors for plant propagation is germination suspension that is caused by ABA in seed development which often persists till maturity (Bewley and Nonogaki, 2017). During seed dormancy, germination is prevented dur to unfavouable environmental conitions (such as water and oxygen) long enough for seedlings establishment and survival. Seed dormancy can be exposed to several treatments that will stimulate the embryo metabolism and increase the level of water and oxygen and reduce the mechanical resistance to the growth of the embryo (Kozlowski and Pallardy, 1997). Seed germination stage is very important in the life of a plant because it plays a major determinant of plant productivity (Ali and Elozeriri, 2017). Seed germination is the critical stage for species survival (Huang *et al.*, 2003; Yang *et al.*, 2008) therefore, the control of seed germination and growth is crucial to the survival of the next generation, and there are critical checkpoints at the transitions from dormancy to germination and from germination to growth (Kermode, 2005). Also, Increased productivity at reduced or minimum cost is important to forest productivity with careful maximization of time in production. Also, the most important issue in successful vegetative production is to grow fast, strong and healthy seedlings in the shortest possible time (Rodo and Filho, 2003).

Synsepalum dulcificum is an indigenous plant particularly to West and Central tropical Africa (Shapiro, 2012). The fruit pulp was traditionally used to sweeten palm wine and it also improve the

flavor of maize bread gone sour (Oliver-Bever, 1986; United State Department of Agriculture, 1919). It is an alternative to the conventional sweetener (sugar) and where used appropriately, the species has the ability to improve diet and health of its user (Cannon, 2004). Slater (2007) reported that there is a small demand of this species by cancer patients and creating a commercial sweetener for the diabetes patients. In addition to the value of the fruit as a taste modifier, the skin and pulp of *Synsepalum dulcificum* may also have antioxidant properties with possible benefits for human health (Inglett and Chen 2011).

In Nigeria, few stands of this species are found particularly in the South-Western Nigeria. According to Adebayo *et al.* (2002), there is lack of awareness and proper information on the potential uses of this species which is a major hindrance to its establishment. Although, *Synsepalum ducificum* can be propagated through stem cutting, air layering or grafting but these do not assure a high percentage of its survival hence, a need to propagate seedlings from seeds. Access to quality seedlings which is a major breakthrough to reforestation is limiting and success of direct sowing in the field has been inconsistent. In addition to lack of awareness and information on the uses of this specie, there is dearth of information on the appropriate nursery propagation for the plant species.

MATERIALS AND METHODS

The study was carried out at the forest nursery unit of the Federal University of Agriculture, Abeokuta situated North-East of Abeokuta, in Odeda Local Government of Ogun State, latitude $7^0 10^1$N and $7^0 58^1$N, and longitude $3^0 20^1$E and $4^0 37^1$N. This site falls within the tropical lowland with two distinct seasons: the longest- wet season last for eight months and the shortest-dry season lasted four months. It is characterized with mean annual rainfall of 1250- 1500mm, and mean monthly temperature ranges between 25.7^0C in July and 30.2^0C in February (OgunOsun River Basin Development Authority, 2015).

Seeds Collection and Soil Media

Seeds of *Synsepalum dulcificum* were collected from the mother trees at Akinyele Local Government Area in Oyo State, and were raised in germination boxes. River sand was collected, washed in acid and sieved with 2mm mesh size to remove any other debris and to ensure that the sands is of uniform in sizes. Forty (40) germination boxes were constructed for the seeds to be sown. Four (4) boxes were used for germinating the mechanically scarified seeds, while twelve (12) boxes each were used for germinating hot water treated seeds, acid treated seeds and hormone treated seeds. Boxes were filled with river sand which had been washed in acid and rinsed thoroughly in flowing water to remove any nutrients or microbes that may be present. Four (4) seeds were planted per planting hole in each box and watered evenly till germination. Germination counts were made on each box, and transformed to percentages by dividing the product of 100 and number of germinated seeds by total number of planted seeds.

$$GP = (NGS/NPS)\ x100$$

Where:

GP = Germination Percentage; NPS = Number of Planted seeds;

Experiment 1: Effect of Mechanical Scarification on seed germination

Seeds of *Synsepalum dulcificum* were mechanically scarified at three different parts of the seed namely 2mm from the distal end (S_1), around the circumference (S_2), 2mm from the micropyle (S_3) with the aid of a sand paper and non-scarified seeds as the control. Seeds were sown at the rate of 4 seeds per hole with 4 replicates in each treatment with a total of 16 seeds. The experiment was laid out in a Complete Randomized Design (CRD) was analyzed using One way Analysis of Variance.

Experiment 2: Effect of soaking in different water temperatures and duration of soaking

The effect of temperatures of water and duration of soaking treatment on germination was investigated by treating the seeds of *Synsepalum dulcificum* in three temperatures of water vis-a-vis 20^oC, 40^oC and 80^oC for 10mins, 30mins, 60mins respectively and untreated seeds represented the control treatment. Seeds were sown four per hole giving a total of 192 seeds with 4 replicates in each treatment. This experiment was laid out in a 3x4 factorial in CRD and was analyzed using ANOVA.

.**Experiment 3: Effect of concentrations of tetraoxosulphate (vi) acid and duration of soaking on *Synsepalum dulcificum* seeds**

The effect of concentration of tetraoxosulphate (vi) acid and duration of soaking in acid on germination of *Synsepalum dulficicum* where the seeds were treated with in 10%, 50% and 90% for a period of 5mins, 10mins, 15mins respectively and untreated seeds were the control treatment. The seeds were carefully removed according to the time required for soaking and were immersed immediately in cool running water to terminate the chemical reactions and remove every trace of acid from the seed. Seeds were sown four seeds per hole giving a total of 192 seeds with four replicates in each trial. This experiment was laid out in a 3x4 factorial in CRD and was analyzed using ANOVA.

Experiment 4: Hormonal effect on *Synsepalum dulcificum* seeds

Synsepalum dulficicum seeds were experimented upon using Indole Acetic Acids (IAA), Indole Butyric Acid (IBA) and Gibberelic Acid (GA_3) hormones at 0.01ppm, 0.02ppm, 0.03ppm and control concentrations respectively. Seeds were sown four per hole giving a total of 192 seeds with four replicates in each trial. This experiment was laid out in a 3x4 factorial in CRD and was analyzed using ANOVA. Significant means were further separated using Fisher's Least Significant Difference (LSD)

RESULTS

Effect of mechanical scarification on seed germination in *Synsepalum dulcificum*

Effect of seeds mechanically scarified as presented on Table 1 shows that, seeds scarified around the circumference and that unscarified seeds (control) had the highest germination value of 50 % respectively. This was significantly different ($p<0.05$) from seeds scarified at the distal end (25%) and micropyle which had the least germination of 0 %.

Table 1: Effect of mechanical scarification on seed germination in *Synsepalum dulcificum*

Scarification Site	Germination (%)
Control	50.00 [a]
Circumference	50.00 [a]
Distal end	25.00 [b]
Micropyle	0.00 [b]

Means within a column with the same superscripts are not significantly different (p>0.05), LSD p=0.05

Effect of water temperatures and duration of soaking on germination in *Synsepalum dulcificum* seeds

Germination recorded for seeds soaked in water irrespective of the duration of soaking as shown on Table 2 revealed that, seeds soaked in 20°C water had the highest germination value of 53.25 % followed by seeds soaked in 40°C water 50 %. The effects were significantly different ($p<0.05$) from seeds soaked in 80°C water which had the least germination value of 14 %.

Table 2: Effect of temperature of water in seed germination of *Synsepalum dulcificum*

Temperature of Water	Germination (%)
20°C	53.25[a]
40°C	50.00[a]
80°C	14.00[b]

Means within a column with the same superscripts are not significantly different (p>0.05), LSD p=0.05

Results on Table 3 showed the effect of duration of soaking seeds in water irrespective of water temperature. It was recorded that untreated seeds (control treatment) had significantly ($p<0.05$) highest germination percentage (54.25%) while seeds soaked for 30 mins, 60 mins and 10 mins had germination values of 39.50 %, 35.50 % and 27 % respectively.

Table 3: Effect of duration of seeds soaking in seed germination of *Synsepalum dulcificum*

Duration	Germination (%)
10 mins	27.00[b]
30 mins	39.50[b]
60 mins	35.50[b]
Control	54.25[a]

Means within a column with the same superscripts are not significantly different (p>0.05), LSD p=0.05

The effect of water temperature and duration of soaking in water has seen on Table 4 revealed that seeds soaked in 20°C water for 30 mins had significantly (*p<0.05*) highest germination of 62.5 %. This effect was not significantly different (*p>0.05*) from seeds soaked in 20°C water for 60 mins (56.25%) and the untreated seeds (50%). However, the least value 0 % was observed in seeds soaked in 80°C for 10 mins and 30 mins respectively.

Table 4: Interactive effect of temperature of water and duration of soaking in seed germination of *Synsepalum dulcificum*

Water temperature (°C)	Duration of soaking (mins)	Germination (%)
20	10	37.50[b]
	30	62.5[a]
	60	56.25[ab]
40	30	56.25[ab]
	60	43.75[ab]
80	30	0.00[c]
	60	6.25[c]
	Control	50.00[ab]

Means within a column with the same superscripts are not significantly different (p>0.05), LSD p=0.05

Effect of concentration of H_2SO_4 acid and duration of soaking in seed germination of *Synsepalum dulcificum*

The result shown on Table 5 depicts that there was no significant difference (*p>0.05*) in seed germination with respect to concentration of the acid. However, seeds treated in 5% acid had the highest germination percentage of 28.25% while the least (17.25%) was observed in seeds treated in 15% acid.

Table 5: Effect of concentration of H_2SO_4 acid in seed germination of *Synsepalum dulcificum*

Concentration of Acid (%)	Germination (%)
5	28.25[a]
10	18.75[a]
15	17.25[a]

Means within a column with the same superscripts are not significantly different (p>0.05), LSD p=0.05

However, seeds untreated in acid had the significantly (*p<0.05*) highest germination percentage (54.25%). The former was different from seeds treated in acid for 5 mins which had a percentage of 23% and was significantly different from seeds soaked in acid for 10 mins (4.25 %) and 15 mins (4.25 %) as shown on Table 5. There was no significance difference (p>0.05) on the seed germination due to and the effect interaction of duration of soaking in acid and concentration of acid.

Table 6: Effect of duration of soaking seeds in H_2SO_4 acid in seed germination of *Synsepalum dulcificum*

Duration (mins)	Germination (%)
5	23.00[b]
10	4.25[c]
15	4.25[c]
Control	54.25[a]

Means within a column with the same superscripts are not significantly different (p>0.05), LSD p=0.05

Effect of concentration and types of growth regulators in seed germination of *Synsepalum dulcificum*

The results showed that, there was significantly no difference (*p>0.05*) in the effect of the concentrations of the hormone (growth regulator) on seed germination, irrespective of the type of hormone used (Table 7). However, untreated seeds had the highest germination percentage (54.25%) while 43.75% germination was observed in 0.01ppm, 0.02 ppm and 0.03 ppm.

Table 7: Effect of concentration of growth regulators in seed germination of *Synsepalum dulcificum*

Concentration of growth regulator (ppm)	Germination (%)
0.01	43.75[a]
0.02	43.75[a]
0.03	43.75[a]
Control	54.25[a]

Means within a column with the same superscripts are not significantly different (p>0.05), LSD p=0.05

Furthermore, types of hormone and the interaction between concentrations of hormone and types of hormone had no significant effect (*p>0.05*) on seed germination as shown on Table 8 and Table 9. However, GA_3 had the highest germination percentage 48.25% while the least germination 39% was observed in seeds treated with IBA. Also, seeds treated in 0.02 ppm GA_3 had the highest germination percentage 62.5% while the least germination 25% was observed in seeds treated in 0.03ppm GA_3 and 0.02ppm IBA.

Table 8: Effect of types of growth regulators on seed germination in *Synsepalum dulcificum*

Type of growth regulator	Germination (%)
GA_3	48.25[a]
IAA	43.75[a]
IBA	39.00[a]

Means within a column with the same superscripts are not significantly different (p>0.05), LSD p=0.05

Key: GA_3 = Gibberelic Acid, IAA = Indole -3-Acetic Acids, IBA = Indole-3- Butyric Acid

Table 9: Interactive effect of concentration and types of growth regulators on seed germination in *Synsepalum dulcificum*

Types of Hormones	Concentration of Hormone (ppm)	Germination (%)
IAA	0.01	37.50 [a]
	0.02	43.75 [a]
	0.03	43.75 [a]
	0.00	50.00 [a]
IBA	0.01	43.75 [a]
	0.02	25.00 [a]
	0.03	31.25 [a]
	0.00	50.00 [a]
GA_3	0.01	50.00 [a]
	0.02	62.50 [a]
	0.03	25.00 [a]
	0.00	50.00 [a]

Means within a column with the same superscripts are not significantly different (p>0.05), LSD p=0.05

Key: GA_3 = Gibberelic Acid, IAA = Indole -3-Acetic Acids, IBA = Indole-3- Butyric Acid

DISCUSSION

According to Kolotelo *et al.,* (2001) seeds processed for regeneration is pretreated with the appropriate techniques to maximize germination capacity and rate before sowing. This was evident as highest germination was recorded in seeds mechanically scarified around the circumference and in unscarified seeds of *Synsepalum dulcificum.* This could mean seeds that the embryo was not injured or damaged during the process of scarification as this may be far from the site of scarification and, enhanced germination might be due to larger surface area for absorption of water and respiration in the seeds. Also, opening through the circumference allowed for ready imbibtion of water hence, the higher germination rate (Zubairu, 2014). This observation agrees with the findings of Aduradola and Shinkafi (1999), Yisau *et al.,* (2015) Muhammad (2014) and Zubairu, (2014) who reported that scarification around the circumference gave the highest germination in seeds of *Tamarindus indica, Albizia zygia, Leucaena leucocephala* and *Acacia senegal* seeds respectively. According to Zubairu, (2014) mechanical scarification stimulates high rate of respiration as which could contributes to the higher germination rate of *Synsepalum dulcificum.* Mechanical scarification also increased germination rate in *Citrullus colocynthis* seeds (Saberi, 2011) and enhanced earlier germination in *Pouteria campachiana* seeds (Amoakoh *et al.,* 2017).

The water temperature (20^oC) and duration of soaking (30 minutes) seeds in water increased the germination rate of the species. Higher germination rate were recorded in of *Azadirachta indica* (Owonubi *et al.,* 2005) and *Adansonia digitata* (Ibrahim and Otegbeye, 2004) when the seeds were soaked in water for longer hours. It is possible that this combination best enhanced by the rupture of seed coat wall hence, permitting air and water through the tissues to enhance physiological changes and the germination of the embryo and also, the leaching out of chemical inhibitors (Sabongari, 2001). Muhammad (2014) and Amusa (2010) however made contrary submissions on the effect of cold water scarification on germination of *Leucaena leucocephala* and *Afzelia africana* seeds respectively. According to Amusa (2010), a lower germination which occurred in *Afzelia africana* seed might be due to insufficient oxygen for the seeds. *Synsepalum dulcificum* seeds soaked in hot water (80^0C) did not germinate. This shows that effect of the water temperature on seed embryo. On a contrary view, Saberi (2011) observed that the treatment of *Citrullus colocynthis* seeds in hot water enhanced its germination. The observable difference might be the difference in the coats of these species as that temperature might be too high for *Synsepalum dulcificum* hence, destroying the embryo.

Duration of soaking seeds in acid revealed that the control treatment had the highest germination mean while soaking duration of 5 minutes also influenced

the germination percentage. This is however contrary to the findings of Zubairu, (2014), Shinkafi, (2006) and Osman *et al.* (2004) who reported the positive effect of H_2SO_4 on *Acacia senegal* and *Lupinus varius* seeds. This difference may be possible due to the difference in hardness of the coats and their response to acid treatment. Irrespective of the significance difference, seeds soaked in 10% and 15% concentration of acid for both 10 and 15 minutes respectively did not germinate. A probable condition might be that the acid concentrations were too high for the embryo hence, its mortality. The effect of hormones on germination showed that 0.02 ppm GA_3 produced the highest germination percentage. This effect was supported by the findings of Christian (2013) who discovered that gibberellic acid and kinetin are better for acquiring germination percentage in stored seeds of *Withania somnifera.* It was also confirmed by the findings of Tsai *et al.* (1997), that gibberellins are important in seed germination as they affect enzyme production which mobilizes food production used for growth of new cells. According to Golmohammadzadeh and Rezvani (2015) as cited by Muhammad (2014) that synthesis of protein and other metabolites needed by the embryo necessary for germination can be activated by Gibberellic acid.

CONCULSION

The study therefore concludes that, scarification around the circumference increased the germination rate of *Synsepalum dulcificum* seedlings in mechanically scarified seeds while seeds soaked in 20^0C of water for 30 minutes resulted in the highest germination percentage for the water treated seeds. Also, seeds soaked in 5% acid for 5 minutes and the control resulted in highest germination percentage while seeds soaked in 0.02ppm GA_3 increased the germination rate in the hormonal treated seeds.

Recommendation

To enhance seed germination of *Synsepalum dulcificum*, mechanical scarification around the circumference, soaking in water of 20^0C for 30 minutes because seeds were destroyed at higher temperatures and GB_3 of 0.02ppm is therefore recommended.

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