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Effects of Hydro-Priming and Potassium Nitrate Priming on the Germination of *Balanites aegyptiaca*

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ABSTRACT

To meet the current demand of forest products through domestication, there is need to embrace cheap, fast and adoptable modern physiological techniques as priming that increase the seed germination percentages, reduce mean germination time and increase seedling growth of agro-forestry tree species. There is dearth of quantified information on the effect of hydro and potassium nitrate priming on the seeds of agro-forestry tree species compared to arable crops. Most of methods of breaking seed dormancy do not promote rapid and uniform germination as priming. In this regards, these experiments were conducted to assess the effect of hydro-priming hours (0, 6, 8, 12 and 14hours) on the germination of B. aegyptiaca seeds; to investigate the effect concentrations of potassium nitrate (0.2, 0.3, 0.4 and 0.5 ppm) and treatment times (0, 12, 24, 36 and 72hours) and to calculate the mean germination time of each experiment. Hydro-priming experiment was laid down in completely randomised design with four (4) replicates. A split-plot experimental design with four (4) replicates was employed for potassium nitrate experiment. Concentrations of potassium nitrate and treatment times constituted main and sub-plot treatments respectively. Results revealed that a significant increase in percentage germination was recorded with the increasing number of hours the seeds were subjected to hydro-priming. The percentage germination was ranged from 18.75% to 57.50% for the control (0) and 14hours hydro-priming. A significant decrease in percentage germination was recorded with the increasing number of concentration of potassium nitrate treatments on seeds. The percentage germination was ranged from 0.10% to 30.07% for 0.5 ppm to 0.2 ppm treatments. Germination percentage of seeds treated in 0.2 ppm concentration of potassium nitrate significantly decreased with the increasing treatment time. Germination percentage was ranged from 10% to 59.83% for the 0hour (control) and 12hours. Mean germination time ranged from 19.25 days to 23.62 days for all concentrations of potassium nitrate and treatment times. The least mean germination time that ranged from 19.25 days to 23.37days was recorded for seeds treated in 0.4 ppm of potassium nitrate for overall treatment time. Highest germination percentage of 30.07% was recorded for seeds soaked in 0.2 ppm of potassium nitrate and was recommended for mass production of its seedling for agro-forestry programmes.

Keyword: *Hydro-priming, Potassium nitrate (KNO_3), Germination, Mean germination time*

INTRODUCTION

Balanites aegyptiaca is an important tree crop of the savannah zone and semi-arid tropical region of Africa that belongs to the Balanitaceae family (Nour-El-Diu *et al*., 2010). It is native to much of the Africa and parts of the Middle East. It is most common in Senegal and Mauritania. It is a potential agro-forestry tree species. The fruit has been called Desert date in English. In Hausa, it is called Aduwa (Wikipedia, 2014). The flesh pulp of both unripe and ripe fruit of *B. aegyptiaca* is edible and eaten dried or fresh. The fruit is processed into a drink and sweet meats in Ghana, alcoholic liquor in Nigeria and a soup ingredient in Sudan. Young leaves and tender shoots are used as a vegetable. The flowers are a supplementary food in West Africa and an ingredient of "dawa dawa" flavouring in Nigeria. The fresh and dried leaves, fruit and sprout are all eaten by livestock, and contributed up to 38% of the dry matter intake of goats in the dry season (WAC, 2014). It also contains 20% - 30% protein (Nour-El-Diu *et al*., 2010). Its various parts are used for nutritional (Nour-El-Diu *et al*., 2010), medicinal, constructional, industrial and heat generating purposes (WAC, 2014).

Despite the economic values of *B. aegyptiaca* the population of the plant is declining at an alarming rate, and there are low conservation measures engaged in. Overgrazing, bush burning and dormancy of its seeds reduce the domestication rate of it. Schelin *et al*. (2003) reported that *Balanites aegyptiaca* seeds possess physical, physiological or combined dormancy which made them to be subjected to different dry heat treatment at 60°C, 80°C and 100°C for 15, 30 and 60 minutes. Sowing *Balanites aegyptiaca* seeds with mesocarp resulted in delayed germination (Elfeel, 2012). The treatment of *Balanites aegyptiaca* seeds with different methods of scarification and gibberellic acid was not significantly increased germination over control (untreated seeds) (Schelin *et al*., 2003). Since little information is available on the breakthrough of the use of other methods on breaking the dormancy of *B. aegyptiaca.* This calls for embracing cheap, simple, fast and adoptable modern physiological techniques as priming that increase the seed germination percentages, seed and seedling vigour of agro-forestry tree species to meet population demand of its products.

Seed priming is a method to promote rapid and uniform germination of seeds, by controlling imbibitions to an extent where germination is initiated, but insufficient to cause radical emergence (Schmidt, 2000). Seed priming is an efficient technique for improvement of seed vigor, increasing germination and seedling growth (Dastanpoor *et al.,* 2013). Seed priming is one of the physiological methods used to increase the uniformity and rate of seed emergence of crops (Sivritepe *et al.,* 2003; Bakht *et al.,* 2010, 2011) and successful results have been obtained for many crops such as melon (Sivritepe *et al*., 2005) and wheat (Iqbal *et al*., 2006). The technique is simple, cheap and safe (Mariem *et al.,* 2013). The excellent performances of priming in enhancing germination percentage, mean germination time and yield of the plant species have been reported (Hossain *et al.,* 2006; Farooq *et al.,* 2006; Ghassemi-Golezani *et al.,* 2008 and Moosavi *et al.,* 2009). Similar observation has been reported by Kalpana *et al.* (2015**)** who stated that among different treatments, KNO_3 (3%) was found superior in priming *Triticum aestivum* and significantly higher than rest of the treatments.

There is dearth of quantified information on the potential of hydro and halo-priming in improving the mean germination time and germination percentage of seeds of forest tree species as *B. aegyptiaca*. In view of this, investigations were conducted to assess the effect of hydro-priming and potassium nitrate on the germination of *B. aegyptiaca* seeds. Overcoming dormancy of *B. aegyptiaca* seeds by priming will help in meeting the needs of Nigerian without jeopardising environment. New initiatives in agro-forestry are seeking to promote poverty alleviation and environmental rehabilitation through efficient information on overcoming constraints in seed germination and seedling growth (Adelani *et al.,* 2014c) for domestication purposes.

MATERIALS AND METHODS

Experimental Site

The research was conducted in the screen house of the Federal College of Forestry Mechanization, Afaka, Kaduna. The college is located in the Northern Guinea Savannah ecological zones of Nigeria. It is situated are about 30 km from Kaduna along – Lagos express road in Chikun Local Government Area of Kaduna state, Nigeria. The Garmin GPS 72 model was used to determine latitude $10^o\ 35^1$ and $10^o\ 34^1$ and longitude $7^o\ 21^1$ and $7^o\ 20^1$ of the college (Adelani, 2015). The rainfall is approximately 1000 mm annually with the lowest mean monthly relative humidity of about 29%. The vegetation is open woodland with tall broad leave trees (Otegbeye *et al.,* 2001).

Fruit Collection and Materials

The fruits of *B. aegyptiaca* were sourced from the mother tree in Kaduna. The seeds were extracted from the fruits and air dried. The viability of the randomly selected seed samples was assessed with the cutting method (Schmidt, 2000). The sand from a 2mm sieve was collected from the floor of the college dam and sterilized for 24 hours. The polypots of 20 x 25 x 25cm^3 filled with the sterilized sand in the nursery were used (Adelani *et al*., 2014a). Potassium nitrate was collected from chemistry laboratory of the college and each quantity in gram was dissolved in 1 litre of distil water to prepare the solutions (Adelani *et al.,* 2014b; Kalpana *et al.,* 2015**)**.

Germination Percentage and Mean Germination Time

Percentage was computed using the following formula

$$\text{Germination Percentage} = \frac{\text{Total seed germinated}}{\text{Total seed sown}} \times 100$$

Mean germination time was calculated using the relation

$$\text{MGT} = \frac{\sum(f_x)}{\sum x}$$

Where *x* is the number of newly germinated seeds is on each day and f is the number of days, after seeds were set to germinate. X is the total number of seeds that germinated at the end of the experiment. Germination percentage and mean germination time were recorded at two days' interval for 8 weeks.

Experimental Procedure

Experiment 1**:** Effect of hydro-priming on the germination of *B. aegyptiaca.* Seeds

To investigate the effects of hydro-priming on the germination of *B. aegyptiaca* seeds, a completely randomized design with four replications was involved. Forty seeds were soaked water for different times durations (0, 6, 8, 12 and 14 hrs). Two hundred (200) *B. aegyptiaca* seeds were extracted from their fruits. The seeds were washed and air dried. The initial moisture content of the samples of the seeds was determined by weighing the seeds on Mettler Top Loading Weighing Balance (Model-Mettler PM 11-K) before and after drying to constant weight. Forty seeds were soaked in water for 0, 6, 8, 12 and 14 hours. The temperature of the water during priming was 28ºC. Stirring or bubbling was done to ensure uniform treatment and aeration. After priming seeds were removed, washed and air dried for 30 minutes and treated with fungicides (vinclozolin). The seeds were sun dried back to the initial moisture content of the seeds. Treated seeds were planted in 4cm depth of the sterilized sand and 200ml of water was applied regularly at two days interval for eight weeks (Adelani *et al.,* 2014b). Means of eight weeks data were used for tabulations. Seeds that were not soaked in water served as control. Germination count was recorded after the emergence of the plumule. Final germination count was taken when no further germination took place for several days.

Experiment 2**:** Effect of potassium nitrate priming on germination of *B. aegyptiaca* seeds

The effect of concentration and time of potassium nitrate on germination of *B. aegyptiaca* seeds was assessed using a split-plot design with four replications. Four concentrations of potassium nitrate (0.2, 0.3, 0.4 and 0.5ppm) constituted the main plot and different times of immersion (0, 12, 24, 36 and 72hrs) constituted the sub-plot treatment. Eight hundred (800)

B. aegyptiaca seeds were extracted from their fruits. The seeds were washed and air dried. The initial moisture content of the samples of the seeds was determined by weighing the seeds on Mettler Top Loading Weighing Balance (Model-Mettler PM 11-K) before and after drying to constant weight. Forty seeds were soaked in different concetrations of potassium nitrate (0.2, 0.3, 0.4 and 0.5ppm) and at different times (0, 12, 24, 36 and 72hrs). Stirring or bubbling was done to ensure uniform treatment and aeration. After each duration, the seeds were removed, washed and air dried for 30 minutes and treated with fungicide (vinclozolin). The seeds were sun dried back to the initial moisture content of the seeds. Treated seeds were sown in 4cm depth of the sterilized sand and 200ml of water was applied at two days interval (Adelani *et al.,* 2014b). Seeds that were not soaked in the potassium nitrate served as control. A seed was considered germinated if the radicle was able to break open the seed coat and plumule emerged. Final germination count was taken when no further germination took place for several days.

Data Analysis

The data was collected on the potentials of two solutions on the germination of *B. aegyptiaca* seeds. The data were subjected to one-way analysis of variance (ANOVA) using SAS (2003) software. Comparisons of significant means were accomplished using Fischer's Least Significant Difference (LSD) at 5% level of significance. Concentration and treatment time made two fact ors with four and five levels respectively. All percent germination data were arcsine-square root transformation prior to analyses because it is appropriate for data on proportions, data obtained from a count, and data expressed as decimal fractions or percentages (Gomez and Gomez, 2010; Sananse and Maidapwad, 2014) covering a wide range (Akindele, 2004). Transformation of data makes the analysis to be *valid* and the conclusions and P values correct. This transformation will spread the values at both ends of the distribution compared with the central part. Germination count was converted to germination percentage, which was obtained as the number of seeds germinated divided by the total number of seeds planted and multiplied by 100.

RESULTS AND DISCUSSION

Effect of hydro-priming hours on the germination of the *B.aegyptiaca* seeds

The result of the effect of hydro-priming hours on the germination of *B. aegyptiaca* seeds is presented in Table 1. A significant increase in percentage germination was recorded with the increasing numbers of hours the seeds were subjected to hydro-priming. The percentage germination was raised from 18.75% to 57.50% for the control (0) and 14hours hydro-priming treatments. This is an indication that *B. aegyptiaca* seeds had longest time to absorb the solution of water that helps in breaking dormancy. Akinola *et al.* (2000) reported that higher duration of exposure to seed treatment resulted in higher cumulative germination in wild sunflower. Positive effect of seed priming on seed invigoration depends on priming duration (Ashraf and Foolad, 2005). Kaya *et al.* (2006) working on germination of sunflower under drought and salt stress reported that hydro-priming improved both rate of germination and mean germination time both under salt and drought stress conditions.

Mean germination time of seeds treated for experimental periods ranged from 21.94 to 23.81 days. Except the seeds that were hydro-primed for 12 hours, increase in the hydro-primed hour increase the mean germination time. The least mean germination time of 21.94 days was recorded for *B.aegyptiaca* seeds not treated. This could be partly adduced to the fact that control did not undergo chemical reaction with water that could affect seed germination. This report is contrary to the observation of the Demir and Mavi. (2004) who stated that prime seeds of water melon emerged 4 days earlier more than those of unprimed ones.

Table 1: Effect of seed hydropriming hours on the percentage germination and mean germination time of the *B. aegyptiaca* seeds
*Means on the same column having different superscript are significantly different (P<0.05) vertically

Seed hydropriming (hours)	Percentage germination (%)	Mean germination time (days)
0	18.75^{c}	21.94^{b}
6	45.00^{b}	23.55^{a}
8	44.45^{b}	23.78^{a}
12	48.85^{ab}	23.73^{a}
14	57.50^{a}	23.81^{a}
SE±	3.23	0.24

The results of effects of potassium nitrate concentrations and treatment times on the germination of *B.aegyptiaca* seeds at the end of experiment are represented in Table 2. Irrespective of treatment time, germination percentage values of 30.07%, 27.01%, 23.30% and 20.10% were recorded for 0.2, 0.3, 0.4 and 0.5 ppm concentration of potassium nitrate. Highest value of 30.07% was recorded for *B.aegyptiaca* seeds treated in 0.2 ppm concentration of potassium nitrate. This shows that germination percentage of *B. aegyptiaca* seeds decreases with increase in concentration of potassium nitrate. This could be attributed to the fact that higher concentration eventually leads to excessiveness or toxicity. Several investigators had reported the efficiency of priming with salts (Hossain *et al*., 2006; Kalpana *et al.,* 2015), on the other hand, earlier reports on argan (Reda Tazi *et al*., 2001) and cereals (Ben Naceur *et al* .2001); chickpea (Al-mutawa, 2003); barley (Kadri *et al*., 2009) and spinach (Keshavarzi *et al.* 2011) reported the unfavourable impact of salinity in priming. The phenomenon is reffered to as ''Over-priming'' (Ely and Heydecker, 1981). At higher NaCl levels and longer duration, germination of *Coriandrum sa,tivum* decreased significantly (Mariem *et al.,* 2013).

Table 2: Effect of potassium nitrate concentrations and treatment times on germination percentage of *B. aegyptiaca* seeds

Treatment concentration of KNO_3 (ppm)	Percentage germination %	Mean germination time (days)	Treatment time (hour)	Percentage germination (%)	Mean germination time (days)
_	__	_	0	10.00^{d}	20.69^{c}
0.2	30.07^{a}	21.85^{a}	12	41.06^{a}	22.84^{a}
0.3	27.01^{a}	22.38^{a}	24	29.96^{b}	23.13^{a}
0.4	23.30^{ab}	22.07^{a}	36	25.95^{b}	22.22^{a}
0.5	20.10^{b}	22.39^{a}	72	18.61^{c}	21.98^{b}
SE±	1.36	0.31	SE±	1.52	0.35

*Means on the same column having different superscript are significantly different P (<0.05) vertically.

B. aegyptiaca seeds soaked in all concentrations of potassium nitrate for this experiment had mean germination times ranged from 21.85 to 22.39 days. Least value of 21.85 days was recorded for 0.2 ppm concentration of potassium nitrate. It can be deduced that the *B. aegyptiaca* seeds germination earlier when treated in low increase concentration of potassium nitrate. This can be attributed to the facts that excessiveness or toxicity of higher concentration potassium nitrate leads to damages of the embryo which eventually affected the time of germination of seeds. Increasing NaCl level led to the reductions in germination percentage for all cultivars of *C.sativum* (i.e Syrian cultivar, Tumision cultivar, Egyptian cultivar, Algerian cultivar) study (Mariem, *et al;* 2013). This germination reduction can be attributed to prevention of water uptake created by the salinity condition. This can be also due to the toxic effects of ions of the salt such as K^{+} and NO_3^{-}. Similar result was obtained by Khajeh Hosseini *et al*. (2013) on the effect of NaCl on the germination of soybean. Contrary to these results are the findings on many crops like melon (Sivritepe, *et al*; 2003), canola (Farhondi *et al;* 2007), pepper (Khan *et al,* 2009), tall fescue (Tilaki *et al.,* 2010), sun flower (Bajehbaj, 2010) and pot Marigold (Sedghi *et al*., 2010). Also, Lqbal *et al*. (2006) on wheat and Shahi *et al.* (2009) on squash suggested that seed priming by higher concentrations of NaCl is an effective method in alleviating the adverse effect of salt stress.

Irrespective of concentration of potassium nitrate, B. *aegyptiaca* seeds treated for 0, 12, 24, 36, 72 hours had germination percentage values of 10%, 41.06%, 29.96%, 25.95% and 18.61% respectively. Highest value of 41.06% was recorded for seeds of *B. aegyptiaca* that was soaked for 12 hours. It can be inferred that *B. aegyptiaca* seeds need 12 hours of immersion in potassium nitrate to enhance its germination. Germination percentage value of *B. aegyptiaca* decreases as the time of immersion in potassium nitrate increases. Time of immersion is an important factor to consider in presowing treatments (Schmidt, 2000). *B. aegyptiaca* seeds treated for 0, 12, 24, 36 and 72 hours had mean germination time values of 20.69 22.84, 23.13, 22.22 and 21.98 days. Least value of 20.69 days was recorded for *B. aegyptiaca* seeds treated for 0hrs. The reason could partly be that untreated seeds need no time to undergo chemical reaction that took place in potassium nitrate treated seeds when watered after planting. The pre-sowing treatment with germination stimulants requires time to undergo chemical reactions. Priming induces a range of biochemical changes in the seed that required initiating the germination process through breaking of dormancy, hydrolysis or metabolism of inhibitors, inhibitions and enzymes activation (Ajouri *et al*., 2004).

The results of interactive effects of concentration of potassium nitrate and treatment time are presented in Table 3. Germination percentage values of *B. aegyptiaca* seeds treated in 0.2, 0.3, 0.4 and 0.5ppm concentration of potassium nitrate for 0, 12, 24, 36 and 72hours had germination percentage values ranged from 10 to 59.83%, 10 to 41.83%, 10 to 36.65% and 10 to 25.95%. Highest germination percentage value of 59.83% was recorded for *B. aegyptiaca* seeds soaked in 0.2ppm concentration of potassium nitrate for 12 hours. From the result of interactive effect of concentration of potassium nitrate and treatment time on the germination percentage of *B. aegyptiaca* seeds, it can be inferred that *B.aegyptiaca* seeds treated for 12 hours had the best result. This is an indication that 12hrs is the moderate time for soaking of *B. aegyptiaca* seeds in the concentrations of potassium nitrate.

Table 3: Interactive effect of concentration of potassium nitrate and treatment time on the germination percentage of *B. aegyptiaca* seeds

	Treatment Time (hours)				
Concentrate of $KN0_3$ (ppm)	0	12	24	36	72
0.2	10.00^a	59.83^a	33.98^b	25.95^c	20.60^c
0.3	10.00^b	41.83^a	31.30^a	31.30^a	20.60^c
0.4	10.00^c	36.65^a	28.63^{ab}	23.28^b	17.95^{bc}
0.5	10.00^b	25.95^a	25.95^a	23.28^a	15.30^b
SE±	3.03	3.03	3.03	3.03	3.03

*Means on the same rows having different superscript are significantly different (P<0.05) horizontally

The result of mean germination time of interactive effects of potassium nitrate and treatment time on the germination of *B. aegyptiaca* seeds are represented in Table 4. Mean germination time values ranged from 19.25 to 23.62days was recorded for seeds soaked in all concentrations and treatment times of potassium nitrate in the second experiment. The least values of 19.00days were recorded for *B. aegyptiaca* seeds treated in 0.4ppm concentration of potassium nitrate for 0hour (control). *B. aegyptiaca* seeds that were not treated (control) germinated within shortest time. It can be inferred from this result that control did not spend time to undergo chemical reaction which affected the germination of the treated ones. This is contrary to the report of Hossain *et al*. (2006) who stated that both priming with NaCl and water resulted in lower time taken value for emergence and mean emergence time and higher value for final emergence, energy of emergence, plant population, achene yield and yield contributing factors as well as achene proteins of *Triticum aestivum*. In the same vein, seed priming contributed to increase speed and synchrony of seed germination (Basra *et al*; 2004; Farooq *et al;* 2006; Pirasteth-Anosheh *et al*; 2011). Ghassemi-Golezani *et al.* (2010) concluded that salt priming; particularly potassium nitrate priming decreases mean germination time and increase seedling size compared with non-primed seeds. Various investigators such as Ghassemi-Golezani *et al*. (2008) (*Cicer arietinum*), Ghassemi-Golezani and Esmaeilpour (2008) (*Cucumis sativus*); Moosavi *et al.* (2009) (Amaranth cv) and Nour-Ed-Din *et al*. (2010) (*Balanites aegyptiaca*) reported that priming decreases mean germination time and increase seedling size.

Table 4: Mean germination time of interactive effect of potassium nitrate and treatment time on the germination of *B. aegyptiaca* seeds

	Treatment time (hours)				
Concentration of $KN0_3$ (ppm)	0	12	24	36	72
0.2	19.60c	22.08b	23.62a	22.40b	21.93b
0.3	21.18a	22.96a	23.03a	23.34a	21.39a
0.4	19.25b	23.37a	22.80a	22.92a	22.01a
0.5	22.75a	22.94a	23.09a	20.59a	22.59a
SE±	0.05	0.05	0.05	0.05	0.05

*Means on the same rows having different superscript are significantly different (P<0.05) horizontally

CONCLUSION

This study has presented the potentials of two solutions in breaking seed dormancy for improved germination rate of B. *aegyptiaca*. We found the highest germination value (57.50%) recorded for *B. aegyptiaca* seeds hydro-primed for 14 hours. The least value of (21.94 days) was recorded for *B. aegyptiaca* seeds hydro-primed for 0 hours. *B. aegyptiaca* seeds treated in 0.2 ppm concentration of potassium nitrate for 12 hours had the highest germination percentage of 59.83%. The least mean germination time of 19.25 days was recorded for seeds treated in 0.4 ppm of potassium nitrate for overall treatment time.

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