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Pre-germination Treatments on Seeds of *Balanites aegyptiaca* (L) Delile and Influence of Potting Mixtures on the Early Growth

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ABSTRACT

Balanites aegyptiaca falls in the category of endangered plants species which has necessitated conservation of this wild growing tree and hence action plan for its maintenance and sustainable development has become indispensable. Therefore, effect of pre-sowing techniques and fertilizer application are inevitable for proper propagation and conservation. This study was conduct to investigate effect of seed pre-treatments and potting mixtures on early growth performance of *Balanites aegyptiaca*. Seeds were subjected to four (4) main pre-sowing seed treatment methods: control in which seeds were sown without pre-treatment (T_1), soak in cold water for 48 hours (T_2), soaked in hot water for 10 minutes (T_3) and soaked in 100 % concentrated Tetraoxosulphate (VI) acid (H_2SO_4) for 10 minutes (T_4). The seedlings were subjected to two different inorganic fertilizers at different levels: Urea (10g), Urea (5g), NPK (10g) and NPK (5g) and the control seedlings planted with soil alone. Growth parameters considered for assessment were shoot height, leaf area, stem diameter and leaf count. Completely Randomized Design was used for experiment. The results of pre-sowing treatments showed that there is significant different (P=0.05) across the four treatments applied on seeds at one month of sowing. However, at the end of 4 week, T_2 maintained the highest germination percentage 85.50% followed by T_3, T_1 and T_4 respectively 81.10%, 78.43% and 77.23%. The results of growth performance showed that there was significant difference (P=0.05) across all fertilizers applied. The shoot height showed that the seedlings performed better in NPK (10g) followed by NPK (5g), control (soil application), Urea (5g) and Urea (10g). Leaf area also showed that there was significant difference (P=0.05) across all fertilizers applied. We reported from our findings that both single application of Urea and combined application of NPK caused enhanced performance on the growth parameters measured.

Keywords: Seed treatment, *Balanites aegyptiaca*, Inorganic fertilizer, Germination

Introduction

Balanites aegyptiaca is a species of classified as a member of the *Balanitaceae*. This tree is native to much of Africa and parts of the Middle East. There are many common names for this plant; in English the fruit is called "Desert date", soap berry tree, Thron tree, while in Egypt the tree is called Egyptian myrobalan and Egyptian balsam. The fruit is called lalob, hidjihi, inteishit, and heglig (hijlij) (Salami and Lawal, 2018a) while in Hausa it is called aduwa, in Tuareg language it is called taboraq, in Swahili mchunju while it is called bedena in Amharic (Delile, 2016). The specific name aegyptiaca was applied by Carl Linnaeus as the species was initially described from specimens collected in Egypt. The species is found in the Sahel-Savannah

region across Africa. It can be found in many kind of habitats, tolerating a wide variety of soil types, from sand to heavy clay, and climatic moisture levels, from arid to sub-humid. It is relatively tolerant of flooding, livestock activity, and wildfire. The *Balanites aegyptiaca* tree reaches 10m (33f) in height with a generally narrow form. The branches have long, straight green spines arranged in spirals. The dark green compound leaves grow out of the base of the spines and are made up of two leaflets which are variable in size and shape. The fluted trunk has grayish-brown, ragged bark with yellow-green patches where it is shed (Delile, 2016). Effective propagation and seedling establishment are the basic requirements for sustainable management of rare species. However, initial stages of plant life cycle, including seed germination and seedling establishment are most susceptible to environmental disturbances, and hence associated with high mortality rates (Moles and Westby, 2004). Seed germination in tree species is sometimes difficult due to hard seed coat and dormant embryos (Jaiswal and Chaudhary, 2005) and the seeds often fail to germinate even under favorable moisture, oxygen and soil conditions. According to (Hartmann *et al.*, 2007), seed germination is influenced by the type of substrate used and environmental factors such as oxygen, water, temperature and for some species may be affected by light. Previous years ago, combination of soil types with different ratios of nutrients have been tested for their suitability towards seed germination (Selivanovskaya and Latypova, 2006), because of its excessive exploitation for a variety of purposes, this species falls in the category of endangered plants species (Elfeel, 2012) which has necessitated conservation of this wild growing tree, and hence action plan for its maintenance and sustainable development has become indispensable. Effective propagation and seedling establishment are the basic requirements for sustainable management rare species. However , initial stage of plant life cycle , including seed germination and seedling establishment are most susceptible to environmental disturbances and hence associated with high mortality rates (Moles and Westby 2004; Salami and Lawal, 2018a) . Seed germination in tree species is sometimes difficult due to hard seed coat and dormant embryos (Jaiswal and Chaudhary, 2005, Salami and Lawal, 2018a) and the seeds often fail to germinate even under favorable moisture, oxygen and soil condition. To overcome this problem, several methods including mechanical scarification, soaking in water and acids (Salami and Lawal, 2018a) and irradiation (Jan and Mahmooduzzafar, 2012 and Aref *et al.*, 2016) are used for treating seeds prior to sowing. This study attempted to investigate the effects of various seed pre-treatment and different potting mixture at different levels on seed germination and seedlings growth of this species in the nursery for plantation establishment in Dutse, Jigawa State of Nigeria.

Materials and Methods

Description of the study area

The study was carried out at Forestry Nursery of the Department of Forestry and Wildlife Management, Federal University Dutse, Jigawa State. It lies between latitude 11.00°N to 13.00°N and longitudes 8.00°E to 10.15°E. The study area covered by Sudan area and also characterized by hot wet summer and cool dry winter with average raining season of 3-5 (644 m) (Jigawa State Ministry of Agriculture and Natural Resources, 2016). Sunshine hours indicate that the town enjoys

10-11 hours of sunshine depending on the season. The topography is characterized by high land area which is almost 750m. Soil tends to be fertile ranging from sandy-loam (Salami and Lawal, 2018b).

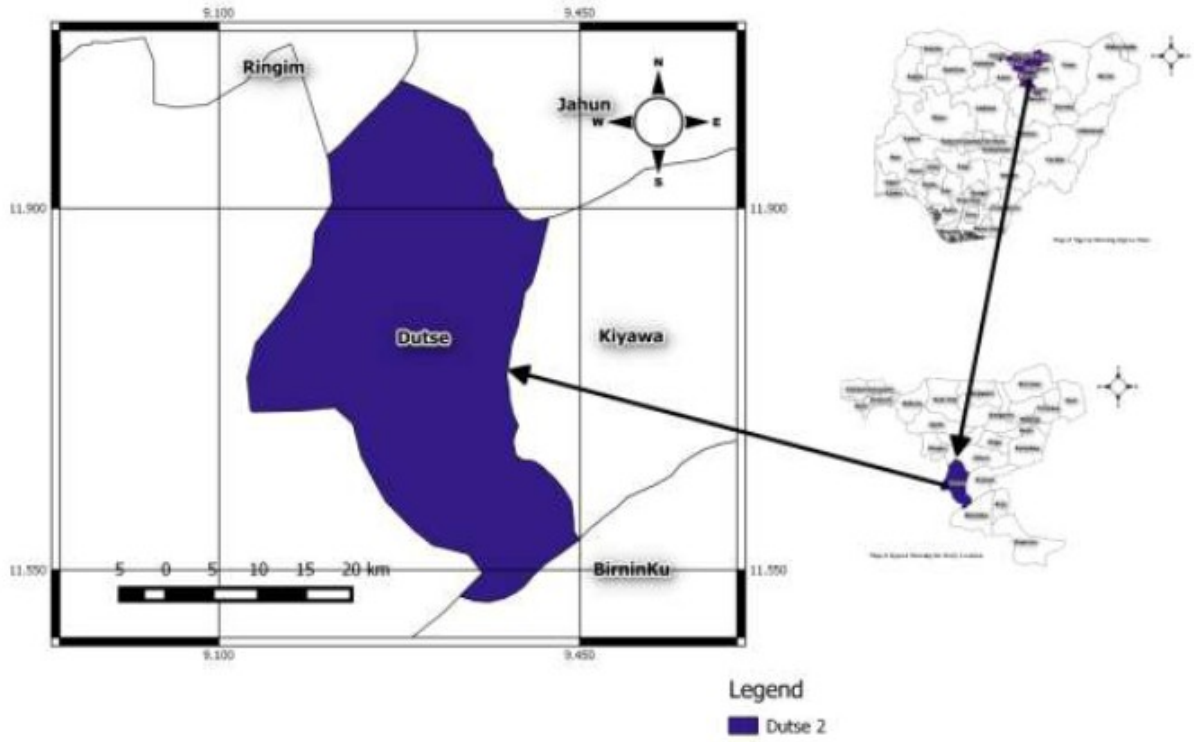

FIG 1: Map of Jigawa showing the study area

Collection of seeds, soil and other materials

Mature seeds of *Balanites aegyptiaca* were collected directly from tree stands in the mother tree in the Federal University Dutse Campus. Seed viability test was carried out by putting the seeds in water and separating those that float. Fresh mature fruits of *Balanites aegyptiaca* were collected and air-dried under normal atmospheric condition. The top soil also collected in the Forestry Nursery Unit of the Federal University Dutse, Jigawa State. Black polythene pots were purchased from University's nursery. Extracted seeds thereby sown in the plastic germination boxes.

Pre-germination Experiment

Materials and Methods

Four pre-treatments methods were used for the scarification of seed lots which enhanced the potential of germination. For each pre-treatment, a total of 105 seeds were subjected to pre-treatment. The untreated seeds were used as a control (T_1) while in another set; seeds were soaked in cold water for 48 hrs (T_2). Seeds were soaked in boil distilled water at 100°C for 10 min in a 100 ml flask (T_3). Finally, seeds were soaked in 100% concentrated H_2SO_4 for 10 minutes and then rinsed thoroughly in cold water finally air-dried (T_4) (Adeniji *et al.*, 2017). Three (3)

germination boxes were used for this experiment; each treatment contained thirty five (35) seeds making one hundred and five (105) seeds for all treatments. Germination is monitored and recorded weekly. For the determination of the effect of different pre-treatments on seeds germination and seedlings growth, for the hot water treatment, water were boiled to approximately 100°C and is then poured into the flask containing 105 seeds and then left for 10 minutes after which the seeds is then planted. Seeds were put into separated flask and Tetraoxosulphate (VI) acid (H_2SO_4) was added to the flask containing 105 seeds and left to soaked for 10 minutes. After immersion, the solution was drained off and seeds were repeatedly rinsed in running tap water until it was safe to handle. Then the seeds were planted and sown. Treated and non-treated seeds were sown directly in plastic germination boxes. Watering was done accordingly for keeping the containers with adequate moisture. The treatments were arranged in a Complete Randomized Design with three replications.

T_1: Seeds treated without treatment (Control)
T_2: Seeds treated with Cold Water
T_3: Seeds treated with Hot Water
T_4: Seeds treated in 100% concentrated chemical (H_2S0_4)

Germination assessment

Data on germination were recorded on weekly basis for a period of eight (8) weeks. Weekly germination percentages were recorded for each treatment. Data collected were subjected to one way analysis of variance (ANOVA) and all means were separated using Duncan multiple range test. Descriptive Statistics was also used to analyze the results collected

Germination percentage and Mean Germination Time

Germination percentage was computed using the formula:

$$\text{Germination percntage} = \frac{\text{Total seed germination}}{\text{Total seed sown}}$$

Mean germination time was calculated using the formula:

$$Mgt = \frac{\sum(fx)}{\sum x}$$

Where x is the number of newly germination seed on each week and f is the number of days, after seeds have set to germinate. x is the total number of seeds that germinated at the end of the experiment. Germination percentage and mean germination time was recorded at one (1) week interval for 8 weeks.

Assessment of growth parameters

The parameters assessed were shoot height, stem diameters, leaf count, and leaf area. Shoot height of seedlings were measured from collar to the tip of the terminal bud and this was done with the help of a metric ruler. The collar diameter of each seedling was measured with the aid of a veneer calliper. The total number of leaves on each seedling was counted, recorded and leaf area (using the Grid method) was assessed.

Preparation of potting mixture

The top soil was air dried and sieved in order to remove all the coarse particles. The top soil and mineral fertilizers varying ratios were applied as treatment. Black polythene pots of (21cm x 14cm) size were filled with 2kg of top soil and there were five treatments replicated ten times making fifty experimental units all together.

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Study two 2: Early growth study
Data analysis

Data collection subjected to one ways Analysis of Variance (ANOVA) with descriptive statistics. Means were compared with the use Duncan Multiple Range Test (DMRT).

Results and Discussion

Results

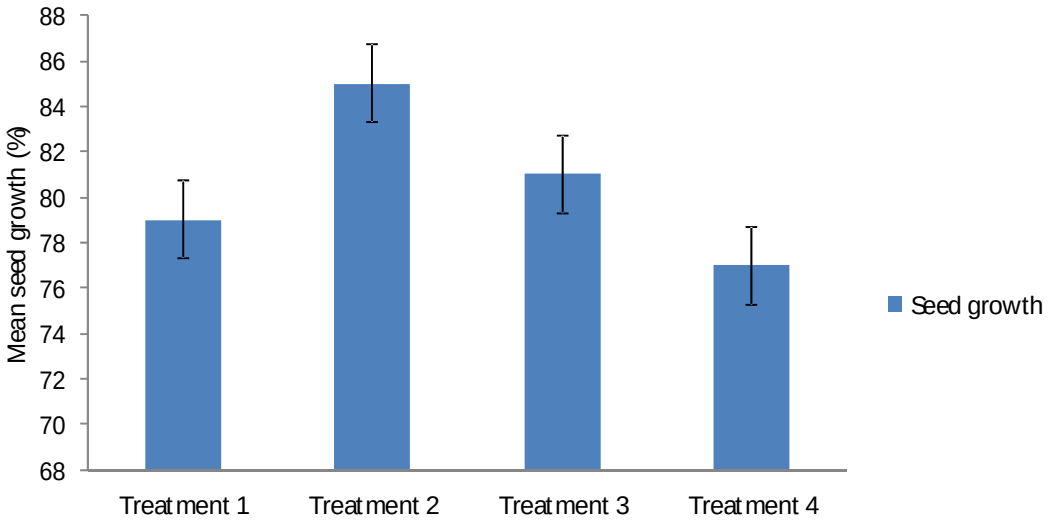

Fig 2: Mean germination percentage of *Balanities eagyptiaca* based on seed treatment

Table 1. Mean Seed germination (%) of *Balanites aegyptiaca* on weekly basis

	wk 1	wk 2	wk 3	wk 4
T1	0.00±0.00	14.53±0.36c	73.06±0.7c	78.43±0.51c
T2	0.00±0.00	22.13±0.23a	82.13±0.43a	85.50±0.55a
T3	0.00±0.00	20.13±0.21b	78.43±0.41b	81.10±0.52b
T4	0.00±0.00	20.13±0.22b	76.46±039b	77.23±0.49c

Significant at p=0.05

Table 2. Mean Shoot height (cm) of *Balanites aegyptiaca* based on fertilizer applications

	Urea 10g	Urea 5g	NPK 10g	NPK 5g	Control
T1	25.93±0.52a	28.20±0.50b	41.03±0.54b	27.03±0.26c	29.56±0.31b
T2	26.24±025a	33.40±0.54a	37.13±0.52c	42.88±0.31a	34.08±0.35a
T3	21.77±022c	32.27±0.51a	33.37±0.51d	32.53±0.36b	31.02±0.41ab
T4	23.93±0.22b	27.44±0.31b	49.03±0.56a	34.27±0.30b	32.67±0.37ab

Significant at P=0.05

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Table 3. Mean Stem diameter of *Balanites aegyptiaca* based on fertilizer applications

	Urea 10g	Urea 5g	NPK 10g	NPK 5g	Control
T1	0.36±0.01a	0.32±0.02a	0.36±0.02a	0.37±0.02a	0.31±0.01a
T2	0.31±0.00a	0.35±0.02a	0.35±0.01a	`0.34±0.02a	0.33±0.00a
T3	0.29±0.01a	0.31±0.01a	0.36±0.02a	0.36±0.01a	0.31±0.02a
T4	0.32±0.02a	0.31±0.01a	0.38±0.02a	0.32±0.01a	0.34±0.02a

Significant at P=0.05

Table 4. Leaf area (cm^2) of *Balanites aegyptiaca* based on fertilizer applications

	Urea 10g	Urea 5g	NPK 10g	NPK 5g	Control
T1	3.53±0.30a	2.93±0.02b	2.77±0.03c	2.60±0.03b	2.73±0.03a
T2	1.93±0.02c	3.23±0.04a	3.20±0.02ab	3.16±0.05a	2.60±0.04a
T3	2.61±0.01b	2.75±0.02bc	3.05±0.04b	3.08±0.05ab	2.30±0.03ab
T4	2.68±0.02b	2.86±0.03b	3.60±0.05a	3.21±0.03a	2.97±0.04a

Significant at p=0.05

Germination

The study established effects of seed treatments and potting mixture on early seedlings growth of *Balanites aegyptiaca*. Figure 2 showed the seed germination of *B. aegyptiaca* based on different treatment on weekly basis. The result showed that there is significant different (P=0.05) across the four treatment applied on seed. However, no growth was recorded on wk 1 across the four treatments. For week 2: T_2 had the highest germination (22.13±0.23%) followed by T_3, T_4 and T_1 with germination values of 20.13±0.21, 20.13±0.22% and 14.0.36% respectively. Looking at wk 3 and wk 4 T_2 still maintained the highest germination percentage followed by T_3, T_4 and T_1 respectively. The growth trend was also represented in Fig 2. This result is an indication that seed treated with cold water performed better than seeds treated with hot water, chemical and control. The highest germination of 85.50±0.55% is in accordance with the findings of Njehoya *et al.*, (2014) in Sudano Sahelian zone of Cameroon where they obtained germination percentage of 69.6% on *Moringa oleifera*. The result of the analysis of variance showed significant different (P=0.05) across the treatments.

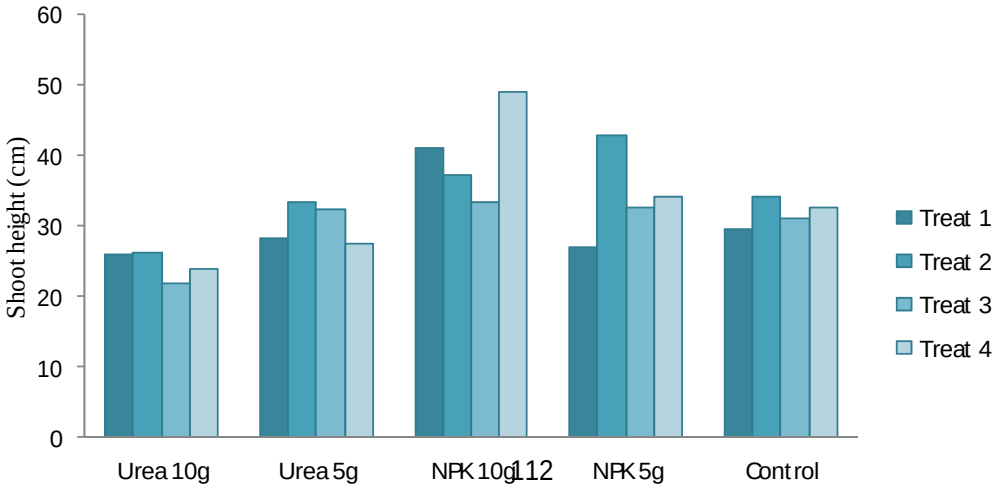

Fig 3: The mean shoot height (cm) of *Balanities eagyptiaca* based on fertilizer application

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Shoot height

The highest mean shoot height was recorded in T_4 under NPK (10g) fertilizer followed by T_2 under NPK (5g), T_1 NPK (10g), T_2 NPK (5g) and T_4 NPK (5g) with values of 49.03±0.56, 42.88±0.31, 41.03±0.54, 37.13±0.52 and 34.27±0.30 respectively. The overall shoot height obtained showed that the plant performed better in NPK (10g) followed by NPK (5g) and soil only, Urea (5g) and Urea (10g) respectively. Fig 3 also expressed the flow trends of the shoot height of *B. aegyptiaca* based on fertilizer applications. This is an indication that the growth in heights of seedlings showed differences with nutrient applications (P=0.05). This is conformity with results obtained for seedlings of *Cedrela liloi* and *Shorea platyclados* treated with different nutrients (Hector, 2000; Ichie *et al.*, 2001 and Ong *et al.*, 2003).

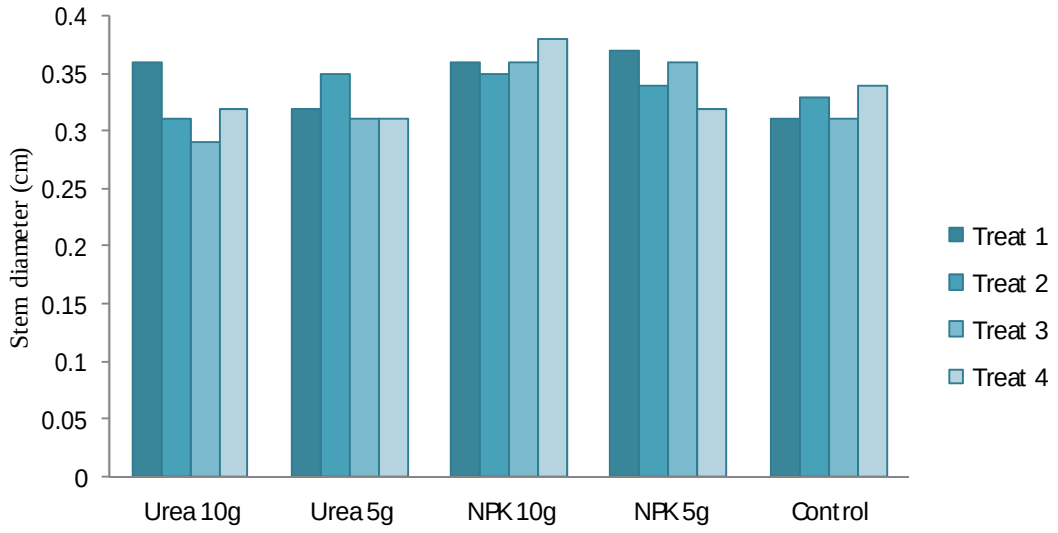

Fig 4: The mean stem diameter (cm) of *Balanities eagyptiaca* based on fertilizer applications

Stem diameter (cm)

There were no significant differences in stem diameter produced by seedlings during the experiment. However, the best treatment combination recorded was T_4 NPK10g. This indicated that nitrogen and phosphorus played important roles in stem diameter formation. Pinkard *et al.*, (2007), working with reported for *Eucalyptus globules*, reported that seedling collar diameter increment did not show significant differences (P>0.05) with nutrient application. Seedling collar diameter increments for fertilized seedlings were generally better than those of the control experiment. Burslem *et al.*, 1995 and Gbadamosi, 2006 also reported similar results for tree seedlings

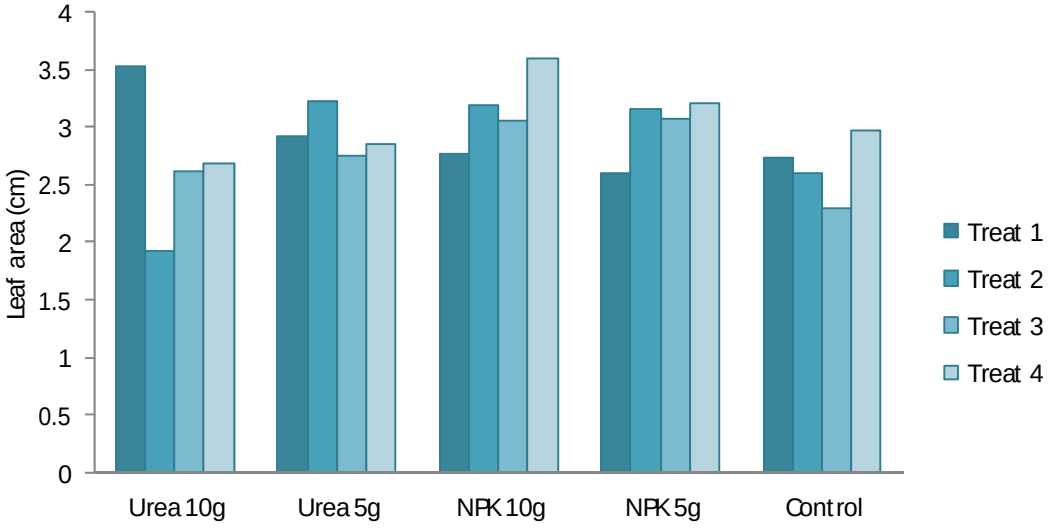

Fig 5: The mean Leaf area (cm) of *Balanities eagyptiaca* based on fertilizer applications

Leaf area (cm^2)

The result of the leaf area (cm^2) obtained showed significant different (P=0.05) as shown in table 4, the trends exhibited by leaf area was presented in Fig 5. From the result it was cleared that T_1 treated with urea (10g) had the highest performance followed by T_4 in NPK (10g). However, the overall leaf areas were found to be relatively good. This is an indication that urea and NPK (10g) aids the leaf area formation of the plant. This is probably by means of stimulating protein formation, rapid cell division and differentiation resulting in collar diameter increment (Bungard *et al.*, 2000; Ong *et al.*, 2003; Hoque *et al.*, 2004). Higher doses of N and P may interfere with metabolic processes and hinder growth (Burslem *et al.*, 1995; Gbadamosi, 2006; Raaimakers and Lambers, 1996). Application of phosphorus and nitrogen resulted in significant increments in leaf area of the species. Fertilized seedlings had better leaf areas than unfertilized ones. This finding is in line with that of Guantilleke (1997) working with *Shorea* species. Pooter (1990) has reported that proper combinations of nitrogen and phosphorus, and other organic compounds may result in increases in leaf area.

A combination of the different nutrients stimulates cell division and promotes stomata opening for cell expansion (Pooter, 1990).

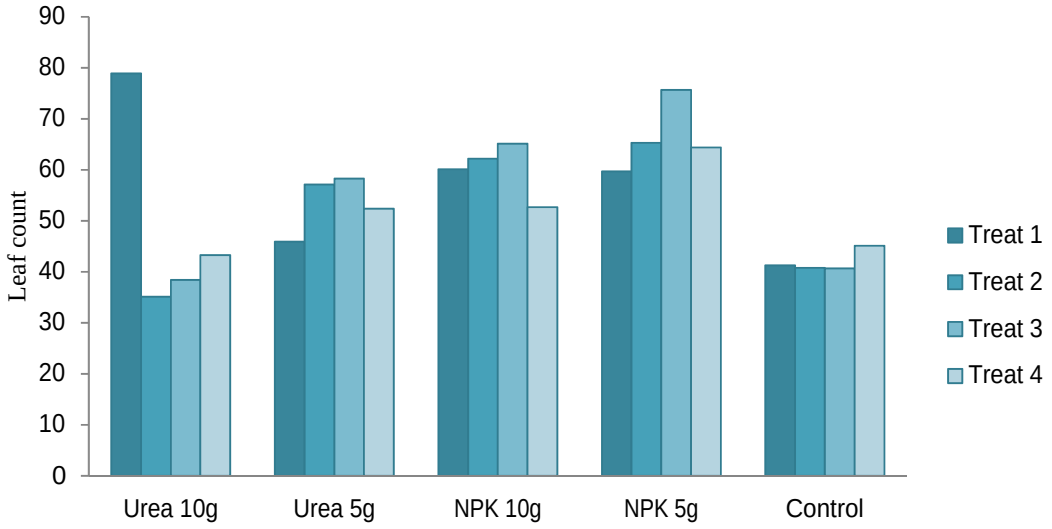

Fig 6: The mean Leaf count of *Balanities eagyptiaca* based on fertilizer applications

Leaf count

Consequently, leaf counts on the other hand also showed different ($P<0.05$) across different fertilizer application (fig 6). The highest leaf areas were found in T_1 Urea (10g), followed by T_3 NPK (5g) while the least was T_2 Urea (10g). This result revealed that fertilizer over time result in significant increase of leaf counts. Similar findings were observed by Bungard *et al*, (2000) in their findings they reported N + P fertilization lead to an increase in plant biomass. Physical characteristics are the visually determinable attributes of tree seedlings in nursery. The major morphological criteria often used to describe timber seedling potentials are shoot height, leaf number, seedling collar diameter, leaf area, root mass and plant root ratio. These are some of the bases to qualify good seedlings for nursery establishment. The findings of this fertilizer experiment show that when there is adequate fertilizer application to seedlings the yield is better contrary to the values of the control. This is also in conformity with reports on plantation trees like *Michelia chapaca* L, *Tectonia grandis* (Teak), and *Entandrophragma cylindricum* Sprague (Hall *et al.*, 2003; Rafiqul *et al.*, 2004). Similar positive results have been reported on growth performance of timber seedlings (Bungard *et al.*, 2000 and Ong *et al.*, 2003).

Conclusion

From the result obtained from the experiment, among the pre-sowing treatments used. Treatment with cold water scarification seems to perform better than others. However, due to the risk involved in the use of concentrated acid and its availability, the use of acid if need arises should be handled by professionals. NPK 15:15:15 and Urea played pertinent roles in stem diameter formation. Fertilization over time resulted in significant increase in leaf counts. The growth performances exhibited across different fertilizer applications are equally noteworthy. The growth variables obtained across different fertilizers were positively correlated.

In conclusion, soaking the seed of *Balanites aegytiaca* in cold water is recommended due to its cost efficiency and also the application of Urea (10g) and NPK (10g) are necessary to raise *Balanites eagytiaca* in the nursery for early growth stage and support development of stem diameter, leaf area and leaf count in preparation for plantation establishment.

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