

## Effects of sowing depths on the germination and early seedling growth of different seed sizes of *Annona muricata* L

Chima, U.D., Etuk, E.C. and Fredrick, C.\*

Department of Forestry and Wildlife Management, Faculty of Agriculture, University of Port Harcourt, Nigeria

\*Corresponding author

Emails: uzoma.chima@uniport.edu.ng, charity.fredrick@uniport.edu.ng\*, etukenobongprety1@yahoo.com

### ABSTRACT

The importance of seed size and sowing depth on germination and early growth of plants cannot be over emphasized. However, information on the right sowing depth for different seed size classes of *Annona muricata* to enhance optimum seed germination and seedling growth is lacking. Viable seeds of *A. muricata* obtained from a single mother tree were sorted and graded into three size classes – small (diameter range = 4.12 to 4.38 mm), medium (diameter range = 5.20 to 5.48 mm) and large (diameter range = 5.50 to 5.66 mm). The mean ( $n = 30$ ) diameter varied significantly ( $p < 0.05$ ) among the seed size classes and was 4.25, 5.36 and 5.58 mm for the small, medium and large seed size classes, respectively. Three depths – 2, 4 and 6 cm, were used to ascertain the effect of sowing depth on germination and early seedling growth of the seed size classes. Seedling height, collar diameter, number of leaves produced and seedling biomass were highest at the 2 cm sowing depth for all the seed size classes and decreased with an increase in sowing depth. The evaluated seedling growth attributes, in most cases, did not vary significantly ( $p > 0.05$ ) between the 2 cm and 4 cm sowing depths on one hand and the 4cm and 6cm sowing depths on the other hand; while they varied significantly ( $p < 0.05$ ) between the 2 cm and the 6 cm depths. Germination rate and growth performance, were also affected by seed size with the large seed size class performing best, followed by the medium seed size class. Seeds in the large seed size class with seed diameter range of 5.50 – 5.66 mm, sown at 2 cm depth, are recommended for optimum germination and early seedling growth performance in *A. muricata*.

**Keywords:** *Annona muricata*, seed sizes, sowing depths, germination, early seedling growth

### INTRODUCTION

Increasing demand for exotic fruits for human consumption has become evident in many countries, both in tropical and temperate regions of the world. The indigenous and often underutilized fruits in many developing countries can provide vitamins, minerals, salt and fiber in human diets (Pinto, *et al.*, 2005).

*Annona muricata*, commonly known as soursop, is a member of the Annonaceae family and is a fruit with long history of traditional use. It is an evergreen plant that is mostly distributed in tropical and sub-tropical regions of the world (Moghadamtousi, *et al.*, 2015). The fruit of *Annona muricata* is extensively used to prepare syrups, candies, beverages, and ice-creams. The edible fruits of the tree are large, heart-shaped and green in color, and

the diameter varies between 15 and 20cm (Moghadamtousi, *et al.*, 2015). *Annona muricata* is a species that is widely studied due to its therapeutic potential. The medicinal uses of the Annonaceae family were reported long time ago and since then, this has attracted attention due to its bioactivity and toxicity (Coria-Tellez, *et al.*, 2016).

Seed size is one of the most important characteristics of seeds that can affect seed development and has a special role in crop production (Ali and Idris, 2015). It is an important focus of selection on the life histories of plants (Saeed and Shaukat, 2000). Zareian *et al.* (2013) reported that seed size is an important physical indicator of seed quality which affects vegetative growth and is related to plant yield. Sowing depth is an important factor in both nursery and plantation establishment depending on the type and size of seed sown (Adeogun and Usman, 2012). Depending on the plant species, some seeds need a deep planting depth for roots to grow deeply for firm anchorage while other plant seeds need to lay on top of the soil without any coverage for germination. An important objective of improved seedling performance is putting seeds at the appropriate depths. Every seed size has a unique planting depth in order to overcome dormancy. Studies on the relationship between seed size and early growth have been reported since early this century (Willenborg *et al.*, 2005). Umeoka and Ogbonnaya, (2016) noted that the effects of sowing depth and seed size on plant growth and development at various stages have been reported by many researchers; however, the effect of sowing depths on germination and the early growth of *Annona muricata* in particular, has not been documented. This study therefore, investigated the effects of sowing depths on germination/emergence and early growth of different seed sizes of *Annona muricata*, for optimal growth and yield.

## MATERIALS AND METHODS

### Study Area

The study was carried at the Department of Forestry and Wildlife Management Arboretum, University of Port Harcourt, Rivers State, Nigeria. The University of Port Harcourt is located at Latitude 4.90794 and 4.90809 N and Longitude 6.92413 and 6.92432 E on a land area of about 400 hectares in Obio/Akpor Local Government Area of Rivers State. The area is characterized with two seasons, the dry and wet seasons, with a nearly all year round rainfall distribution (Aiyeloja *et al.*, 2014). The arboretum is located at the Abuja Campus of the University and it covers a total land area of about 4226.2581m<sup>2</sup> containing several tree species which include *Gmelina arborea*, *Tectona grandis*, *Nauclea diderrichii*, *Khaya grandifoliola* and *Irvingia gabonensis*. These tree species are of great importance in wood production, paper making, food production, among several other uses (Chima *et al.*, 2016).

### Seed Collection, Processing, and Viability Test

The fruit of *Annona muricata* was obtained from a healthy mother tree with good vigour and growth characteristics within the study area. The seed was processed manually by depulping. Seed viability test was done using the floatation method in which the floated seeds were discarded thereafter.

### Experimental Design

The experimental design used was Completely Randomized Design (CRD) for each seed size class. The diameter of the seeds were used to classify seeds into three size classes namely; small (4.12 to 4.38 mm), medium (5.20 to 5.48 mm) and large (5.50 to 5.66 mm). A vernier caliper was used to measure the diameter of the

seeds. A total number of 90 seeds were used for the experiment at 30 seeds per seed size (i.e. 10 seeds per sowing depth). Seeds were directly sown into polypots filled with sterilized forest topsoil at three depths – 2, 4 and 6 cm, for each seed size class. Topsoil was collected from 0 – 20 cm depth at the University of Port Harcourt Arboretum. Leaf litters on the forest floor were removed before the collection of the topsoil. The collected topsoil was properly bulked to enhance its homogeneity. Each polypot was taken as a replicate of its own.

### Data collection

Data were collected on the following:

1. Germination percentage
2. Seedling height, collar diameter and leaf production (number of leaves)
3. Wet weight and dry weight of the root and shoot of the seedlings.

The data on seedling growth attributes of the different seed size classes at different sowing depths were collected on weekly basis for a period of three months (twelve weeks).

### Data Analysis

Germination percentage was computed using the formula below after Bybordi (2010):

$$GP = \frac{n}{N} \times 100$$

Where: n = number of germinated seeds;  
N = number of seeds sown

A one-way analysis of variance (ANOVA) was used to test for significant difference in the measured seedling growth attributes among the three depths for each seed size class. The analysis of variance was performed using Statistical Package for Social Sciences (SPSS) as described by Oloye (2001). The Least Significant

**Table 1: Diameter of different seed sizes**

Seed Class	Seed Diameter (mm)	
	Range	Mean (n = 30)
Small Size	4.12 – 4.38	4.25 ± 0.02 <sup>a</sup>
Medium Size	5.20 – 5.48	5.36 ± 0.02 <sup>b</sup>
Large Size	5.50 – 5.66	5.58 ± 0.01 <sup>c</sup>

Means with different alphabet on the same column are significantly different (p < 0.05)

Difference (LSD) test was used for mean separation.

## RESULTS

### Variation in Seed Sizes

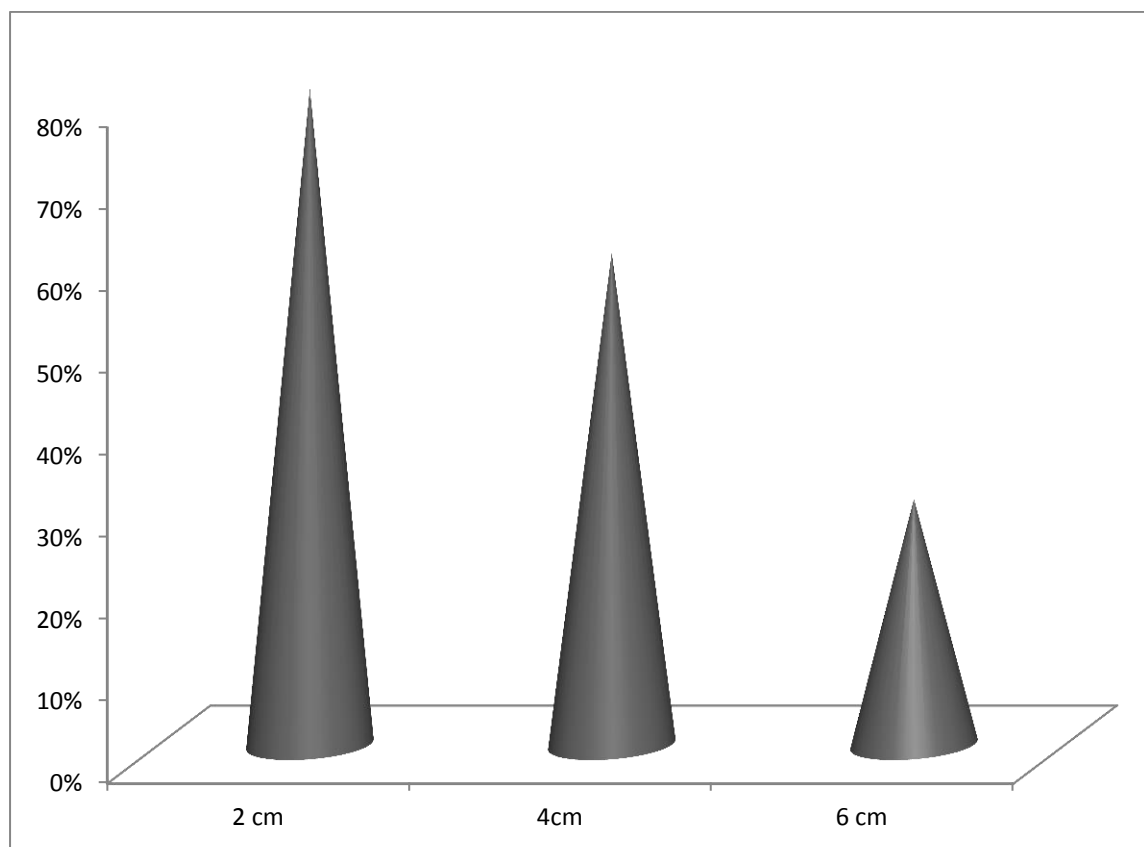
The diameter range and mean of seeds in the different seed size classes is presented in Table 1. The means were significantly different (P < 0.05).

### Effect of Sowing Depth on the Germination of Small-size Seeds of *Annona muricata* and Early Growth of their Seedlings

The germination rate of the small size seed class at different sowing depths is presented in Figure 1. The germination rate was highest in 2 cm depth and decreased with an increasing sowing depth.

Seedling height, collar diameter, and the number of leaves were highest and lowest at the 2 cm and the 6 cm sowing depths, respectively (Table 2). However, seedling height and the number of leaves did not vary significantly between the 2 cm and 4 cm sowing depths while there was no significant difference in collar diameter at the three sowing depths.

Seedling biomass of the small size seed class at the three sowing depths is presented in Table 3. Dry shoot weight and fresh shoot weight were highest at the 2 cm depth and decreased with an increase in sowing depth. Both dry root weight and fresh root weight were also highest at 2 cm sowing depth and decreased with an increase in sowing depth. However, there were significant differences in fresh shoot weight and fresh root weight between the 2 cm and the 6 cm sowing depths only.



**Figure 1: Germination rate of small-size seeds of *Annona muricata* at different sowing depths**

**Table 2: Early growth performance of *Annona muricata* seedlings from small-size seeds sown at different depths**

Growth Attribute	Sowing Depth		
	2 cm	4 cm	6 cm
Total Height (cm)	20.18 ± 0.55 <sup>a</sup>	19.04 ± 0.67 <sup>ab</sup>	16.72 ± 0.87 <sup>bc</sup>
Collar Diameter (mm)	2.49 ± 0.11 <sup>a</sup>	2.34 ± 0.23 <sup>a</sup>	2.16 ± 0.09 <sup>a</sup>
No. of Leaves	8.25 ± 0.37 <sup>a</sup>	8.17 ± 0.31 <sup>ab</sup>	6.67 ± 0.67 <sup>c</sup>

Means with the same alphabet on the same row are not significantly different ( $p > 0.05$ )

**Table 3: Biomass of *Annona muricata* seedlings from small-size seeds sown at different depths**

Biomass	Sowing Depth		
	2 cm	4 cm	6 cm
FSW (g)	3.65 ± 0.23 <sup>a</sup>	3.13 ± 0.30 <sup>ab</sup>	1.70 ± 0.64 <sup>c</sup>
DSW (g)	0.56 ± 0.06 <sup>a</sup>	0.48 ± 0.05 <sup>a</sup>	0.39 ± 0.04 <sup>a</sup>
FRW (g)	0.33 ± 0.03 <sup>a</sup>	0.27 ± 0.04 <sup>ab</sup>	0.17 ± 0.03 <sup>bc</sup>
DRW (g)	0.09 ± 0.01 <sup>a</sup>	0.07 ± 0.01 <sup>a</sup>	0.06 ± 0.01 <sup>a</sup>

Means with the same alphabet on the same row are not significantly different ( $p > 0.05$ )

FSW = Fresh Shoot Weight; DSW = Dry Shoot Weight; FRW = Fresh Root Weight; DRW = Dry Root Weight

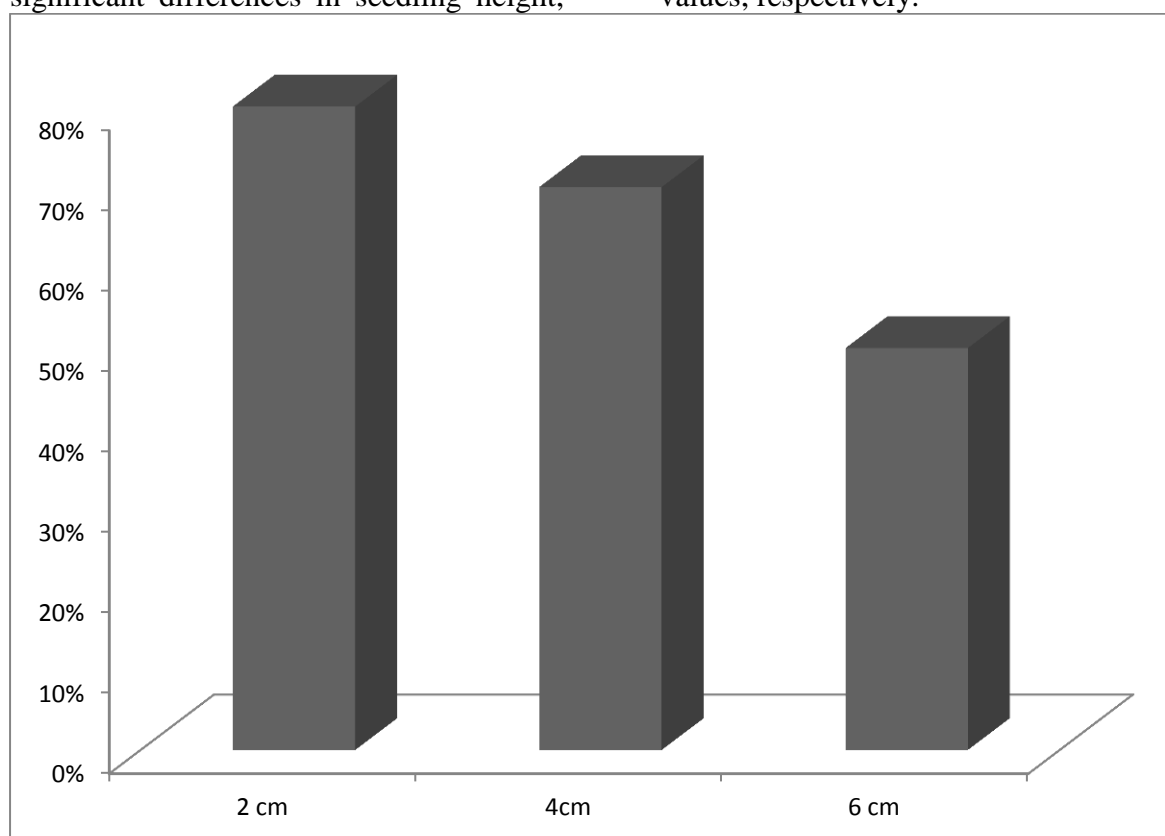
### Effect of Sowing Depth on the Germination of Medium-size Seeds of *Annona Muricata* and early Growth of their Seedlings

The germination rate of the medium size seed class at different sowing depths is shown in Figure 2. The germination rate was highest in the 2 cm sowing depth and decreased with an increase in the sowing depth. However, the gap in the germination rate between the 2 cm sowing depth and the other sowing depths narrowed down when compared with small size seed class.

The growth performance of the medium size seed class is presented in Table 4. Seedling height, collar diameter and number of leaves were also highest at the 2 cm sowing depth and decreased with an increase in sowing depth. There were significant differences in seedling height,

collar diameter and number of leaves between the 2 cm and the 6 cm sowing depths while there were no significant differences in the three growth parameters between the 2 cm and the 4 cm sowing depths on one hand, and the 4 cm and 6 cm sowing depths on the other hand.

Seedling biomass of the medium size seed class at the three sowing depths is shown in Table 5. Dry shoot weight and fresh root weight did not vary significantly at the three sowing depths, although, the highest and lowest mean values were recorded at the 2cm and 6 cm sowing depths, respectively. Fresh shoot weight varied significantly between the 2 cm and the 6 cm depths and also between the 4 cm and the 6 cm sowing depths. Dry root weight also varied significantly between sowing depths with the 2 cm and the 6 cm depths having the highest and the lowest mean values, respectively.



**Figure 2: Germination rate of medium-size seeds of *Annona muricata* at different sowing depths**

**Table 4: Early growth performance of *Annona muricata* seedlings from medium-size seeds sown at different depths**

Growth Attribute	Sowing Depth		
	2 cm	4 cm	6 cm
Total Height (cm)	21.42 ± 0.60 <sup>a</sup>	21.18 ± 0.81 <sup>ab</sup>	18.05 ± 0.85 <sup>c</sup>
Collar Diameter (mm)	2.67 ± 0.12 <sup>a</sup>	2.62 ± 0.67 <sup>ab</sup>	2.28 ± 0.97 <sup>c</sup>
No. of Leaves	9.00 ± 0.27 <sup>a</sup>	8.71 ± 0.42 <sup>ab</sup>	5.20 ± 0.37 <sup>c</sup>

Means with the same alphabet on the same row are not significantly different ( $p > 0.05$ )

**Table 5: Biomass of *Annona muricata* seedlings from medium-size seeds sown at different depths**

Biomass	Sowing Depth		
	2 cm	4 cm	6 cm
FSW (g)	3.95 ± 0.37 <sup>a</sup>	3.64 ± 0.24 <sup>ab</sup>	2.34 ± 0.34 <sup>c</sup>
DSW (g)	0.62 ± 0.07 <sup>a</sup>	0.54 ± 0.06 <sup>a</sup>	0.42 ± 0.05 <sup>a</sup>
FRW (g)	0.39 ± 0.05 <sup>a</sup>	0.33 ± 0.04 <sup>a</sup>	0.26 ± 0.05 <sup>a</sup>
DRW (g)	0.13 ± 0.01 <sup>a</sup>	0.09 ± 0.01 <sup>b</sup>	0.05 ± 0.01 <sup>c</sup>

Means with the same alphabet on the same row are not significantly different ( $p > 0.05$ )

FSW = Fresh Shoot Weight; DSW = Dry Shoot Weight; FRW = Fresh Root Weight; DRW = Dry Root Weight

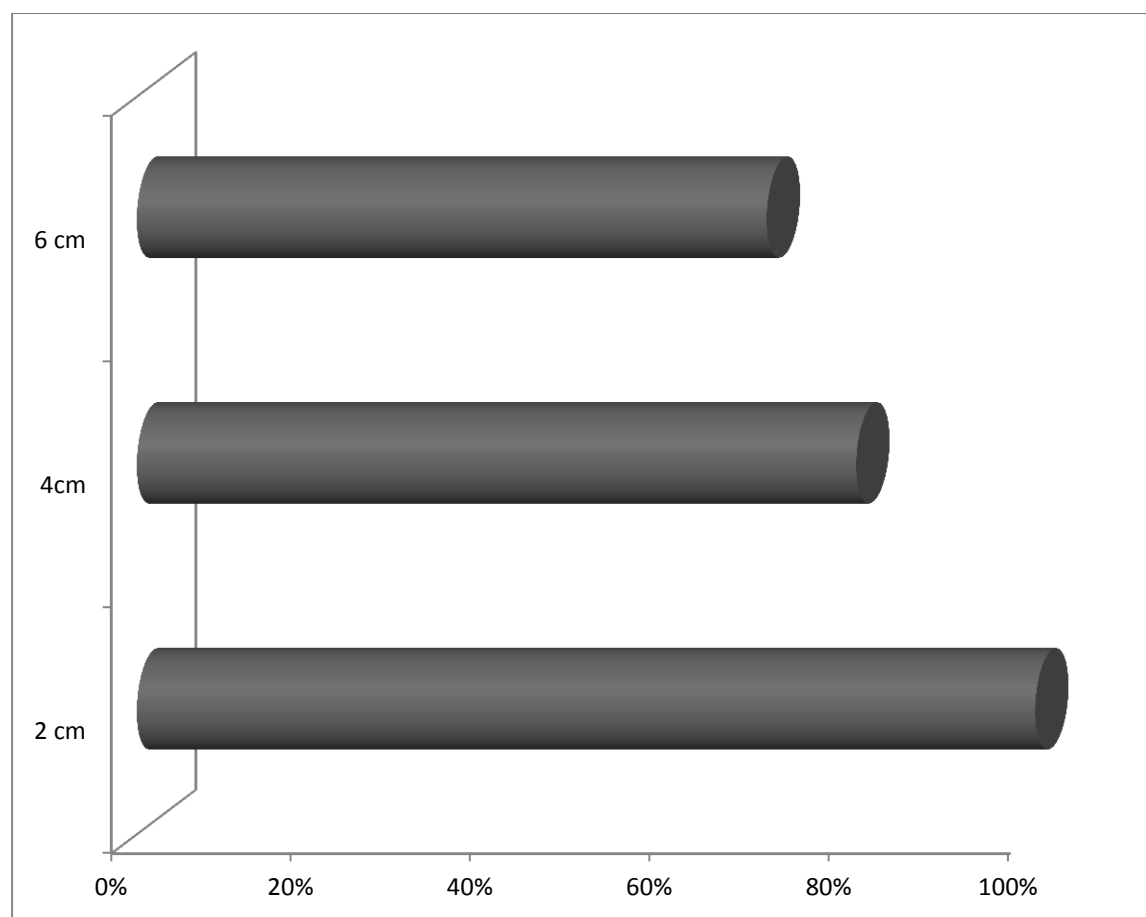
#### **Effect of Sowing Depth on the Germination of Large-size Seeds of *Annona muricata* and early Growth of their Seedlings**

The germination rate of the large size seed class is shown in Figure 3. The germination rate further improved with the large size seed class, with 100 % germination recorded at the 2 cm sowing depth. However, germination also decreased with an increase in the sowing depth.

The growth performance of seedlings of the large size seed class is shown in Table 6. Higher mean values were recorded in seedling height, collar diameter and number of leaves than in both the small and the medium size seed classes. However, the trend with respect to sowing depth remained the same with these growth attributes, as the highest and the lowest mean values were recorded at the 2 cm and the 6 cm sowing depths,

respectively. There were significant differences in seedling height, collar diameter and number of leaves between the 2 cm and the 6 cm sowing depths while no significant differences were observed in seedling height, collar diameter and number of leaves between the 2 cm and the 4 cm sowing depths, although their mean values were slightly higher at the 2 cm depth.

The seedling biomass for the large size seed class is shown in Table 7. The highest and the lowest mean values were recorded at the 2 cm and the 6 cm sowing depths, respectively. Fresh shoot weight varied significantly among the three sowing depths. However, no significant differences were observed in dry shoot weight, fresh root weight and dry root weight between the 2 cm and the 4 cm sowing depths. Seedling biomass of the large size seed was also higher than that of small and medium size classes.



**Figure 3: Germination rate of large-size seeds of *Annona muricata* at different sowing depths**

**Table 6: Early growth performance of *Annona muricata* seedlings from large-size seeds sown at different depths**

Growth Attribute	Sowing Depth		
	2 cm	4 cm	6 cm
Total Height (cm)	22.15 ± 0.67 <sup>a</sup>	21.68 ± 0.77 <sup>ab</sup>	18.62 ± 0.66 <sup>c</sup>
Collar Diameter (mm)	3.02 ± 0.10 <sup>a</sup>	2.81 ± 0.12 <sup>ab</sup>	2.33 ± 0.10 <sup>c</sup>
No. of Leaves	9.40 ± 0.22 <sup>a</sup>	9.25 ± 0.25 <sup>ab</sup>	6.00 ± 0.72 <sup>c</sup>

Means with the same alphabet on the same row are not significantly different ( $p > 0.05$ )

**Table 7: Biomass of *Annona muricata* seedlings from large-size seeds sown at different depths**

Biomass	Sowing Depth		
	2 cm	4 cm	6 cm
FSW (g)	4.96 ± 0.22 <sup>a</sup>	4.08 ± 0.22 <sup>b</sup>	3.13 ± 0.30 <sup>c</sup>
DSW (g)	0.76 ± 0.08 <sup>a</sup>	0.71 ± 0.09 <sup>ab</sup>	0.49 ± 0.05 <sup>b<sup>c</sup></sup>
FRW (g)	0.56 ± 0.05 <sup>a</sup>	0.46 ± 0.07 <sup>ab</sup>	0.27 ± 0.04 <sup>c</sup>
DRW (g)	0.15 ± 0.01 <sup>a</sup>	0.14 ± 0.01 <sup>ab</sup>	0.09 ± 0.01 <sup>c</sup>

Means with the same alphabet on the same row are not significantly different ( $p > 0.05$ )

FSW = Fresh Shoot Weight; DSW = Dry Shoot Weight; FRW = Fresh Root Weight; DRW = Dry Root Weight

## DISCUSSION

Each species has specific sowing depth requirement based on the type of seed and environmental condition (Agboola, 1996; McWilliams, *et al.*, 1998; Aldrete and Mexal 2005). The germination rate of the three seed size classes was affected by increasing sowing depths with the 2 cm depth having higher mean values for the different growth attributes evaluated. This result agrees with Nabi, *et al.* (2011) and Siddig, *et al.* (2015) who in their studies on cotton (*Gossypium* spp) and faba beans (*Vicia faba* L.) respectively, reported that germination rate reduced significantly with increased sowing depth. Apart from the observed effect of increasing depths on germination rate of *A. muricata* as observed in this study, Kumar and Srivastava (2010) had also reported that seed depth affected germination percentage in *Ricinus communis. muricata*. The seed size was also observed to have affected the germination rate of *A. muricata* as the large seed size class had 100% germination at the 2 cm sowing depth as against 80% for both the medium and the small seed size classes. Large seed have been reported to have positive effect on germination and seedling growth of plants (Seyed, 2011). Amico *et al.* (1994) noted that higher vigor observed in larger seeds could be due to larger food reserves in these seeds. Mtambalika *et al.* (2014) also reported about 95% germination of large seeds of *Afzelia quanzensis*. However, Umeoka and Ogbonnaya (2016), observed the opposite in *Telfairia occidentalis* where small seeds germinated faster and were more established than the medium and large seeds. These differences may be due to the physiological pathway of the germinating seeds.

Seedling height was significantly affected by both the seed size and the sowing depth. For instance, seeds in the same size class performed better at the 2cm depth while the large seed size class performed better than the medium and small seed size

classes at each depth. Mtambaliki *et al.* (2014) reported that large seeds produced crops with higher heights than small seeds at the same planting depth in *Afzelia quanzensis*. The success of larger seeds in plant vigor was explained by Ali and Idris (2015), who interpreted this phenomenon better using the seeds anatomical and physiological characters – larger seeds have more endosperm to supply adequate potential for increased growth and enhanced development. This contradicts the findings of Umeoka and Ogbonnaya (2016) who revealed that increasing sowing depths significantly reduced cumulative height growth over time and that small seeds attained the highest plant height irrespective of the sowing depth.

There was no significant difference in the collar diameter of all small size seed class at all the sowing depths; however, for medium sized seeds and large seeds, collar diameter varied significantly at 2 cm and 6 cm sowing depths, with large seeds having the highest collar diameter. This is similar to the finding of the study of Mtambaliki *et al.* (2014) on *Afzelia quanzensis* where the collar diameter was found to be largest in plants from larger seeds but disagrees with that of Umeoka and Ogbonnaya (2016), where small seeds had the highest radial growth from the beginning to the end of the growth period, followed by the medium sized seeds. However, in another study by Gholami *et al.* (2007) on *Pistacia atlantica*, collar diameter was not significant with changes in sowing depth.

The number of leaves produced by seedlings from the three seed size classes was significantly different between the 2 cm planting depth and 6cm planting depths, but did not vary significantly between 2 cm and 4 cm planting depth. In this study, number of leaves was highest in large seeds sown at a planting depth of 2 cm. Similar results were observed by Ali and Idris (2015), where the number of leaves reduced with increasing sowing



depth but increased with seed size in the cultivation of faba beans (*Vicia faba* L); and also by Sime, *et al.* (2016), where maize cultivated at a shallow sowing depth of 2cm produced more leaves significantly different from maize cultivated at 6 cm. However, contrary results were reported by Kumar (2007) in his paper on Neem seeds where small seeds had larger number of leaves compared to larger seeds.

The variations in seedling biomass almost followed the same trend with the variations in the evaluated seedling attributes especially growth in height and can equally be explained by the factors responsible for the variations in the latter.

## CONCLUSION

The three seed size classes (small, medium and large) of *A. muricata* performed best at the 2cm sowing depth followed by the 4cm and 6 cm depths, respectively. However, seedling growth performance based on the evaluated attributes did not vary significantly between the 2cm and 4cm sowing depths on one hand and the 4 cm and 6 cm sowing depths on the other hand; while they varied significantly between the 2 cm and the 6 cm depths. Both germination rate and growth performance, were also affected by seed size with the large seed size class performing best, followed by the medium seed size class. Seeds in the large seed size class with seed diameter range of 5.50 – 5.66 mm, sown at 2 cm depth are recommended for optimum germination and seedling performance in *A. muricata*.

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