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Stem cutting size influence on sprouting and survival of stem cuttings of African teak (*Milicia excelsa* (Welw.) in Kenya

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African Teak (Milicia excelsa (Welw.) belongs to the Moraceae family. Vegetative propagation has been proposed to overcome some of the reproductive biology challenges in the species. The study was to determine the influence of stem size on sprouting and survival of stem cuttings of African teak. The experiment was conducted at Egerton University, Njoro, Kenya. The propagation materials were collected from healthy mother trees from the Coast and Western regions of Kenya. A Randomized Complete Block Design was used. The cutting lengths were 3, 6 and 9 cm. The planting medium was composed of river sand. Data were collected monthly on the number and height of sprouts and the cutting survival at the end of 150 days. Data were then subjected to analysis of variance (ANOVA) using GENSTAT 15th Edition. Separation of means was performed using the SED. The results showed that the cutting length had a significant effect on the number of sprouts, F=43.45, df=2, p= 0.001 and height of sprouts produced, F=48.25, df=2, p=0.001. None of the cuttings obtained 50% of survival though the highest survival percentage obtained was 48.1% from cuttings with 9 cm length. The survival of cuttings obtained from 6 cm length reached 41.27% while 3 cm length attained 31.75%. It was concluded that cutting length positively influences the sprouting of cuttings; hence longer cutting should be selected for propagation. Further studies should take into account the optimum cutting length and the factors which influence the shoot formations and cutting survival including diseases prevention and age of cuttings.

Key words: Milicia excelsa, stem cuttings, vegetative propagation.

INTRODUCTION

Milicia excelsa (formerly known as *Chlorophora excelsa* (Welw.), is an economically significant timber species of West, Central and East Africa (Dossou-Yovo et al.,

2020). Most of the timber for this species comes from natural forests and currently, only a few commercial plantations have been established (Tabuti, 2012). Gall-

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Author(s) agree that this article remain permanently open access under the terms of the <u>Creative Commons Attribution</u> <u>License 4.0 International License</u> forming insects in the genus Phytolyma (Homoptera: Psyllidae), which includes; *P. lata, P. fusca,* and *P. tuberculata*, attacks Milicia species in plantations, leading to slow and general failure to grow the species (Elisabeth et al., 2016). Due to its vulnerability to the gall-forming insect Phytolyma which deforms the shoot apex and inhibits growth, its cultivation is mostly constrained.

Vegetative propagation is the process of using a fragment of a parent plant and inducing it to develop into a new plant. The offspring shares the same genetic makeup as the parent plant. Asexual reproduction uses a plant's vegetative components such as; its stems, roots, or leaves (Olaniyi et al., 2022). Asexual reproduction has a number of benefits, such as being simpler and quicker than in woody perennials like Milicia and being the only method to duplicate particular cultivars while avoiding the juvenile features of some species (Zamora et al., 2022). In order to encourage the establishment of trees in agrosystems, a number of propagation techniques have been developed, including the propagation of seeds and vegetative propagation techniques such as layering and cuttings (Meunier et al., 2016).

Rooting has been found to be positively correlated with cutting length in a number of tree propagation experiments (Lustosa et al., 2022). This is attributed to the presence of carbohydrates and mineral elements in longer cuttings, which are necessary for root formations. Other studies have shown that in many plant species, cuttings taken from different positions within a shoot differ in their rooting ability potential (Duarte et al., 2019).

Earlier studies from seedling screening experiments suggest that pest-resistant genotypes may exist in wild populations, according to research done in 1996 at the Forestry Research Institute of Ghana (FORIG), which aimed to produce pest-resistant planting stock through a genetic improvement program (Ugwu and Omoloye, 2015). Vegetative propagation techniques are necessary to replicate genotypes that exhibit pest resistance. *Milicia excelsa* can be propagated through seedlings and wildings, although their seeds are susceptible to pest attacks. On the other hand, suckers are also produced which can be used in its propagation (Ugwu and Omoloye, 2015).

The African teak is dioecious with male and female trees slightly different in appearance. Male trees exhibit longer and slender trunk and crown and forking being more common in male compared to female trees (Djagbletey et al., 2011). On the other hand, male flowers are white and closely clustered in a slender, pendulous catkin up to 20 cm long whilst females are greenish in a hairy spike that is shorter and wider. Raising nursery seedlings for planting is a problem due dioecism nature of the species.

Seed germination is slow and might take longer in the seedbed; hence the need for strategies to improve seed germination (Trivedi et al., 2020). To address the sluggish and unpredictable seed germination as well as the dioecy

phenomena in the species reproductive biology, various research findings suggested that vegetative propagation could be employed to solve the problem (Marunda et al., 2020). This paper describes experiments which were undertaken with stem cuttings of *M. excelsa* to determine the influence of stem size on sprouting and survival.

MATERIALS AND METHODS

The study was conducted at the Agroforestry Tree Nursery, Egerton University, Njoro, Kenya. The University is located in Njoro, a small community located 25 kilometers southwest of Nakuru City. The latter is located approximately 182 kilometers northwest of Nairobi. The study site lies on a latitude 0°22'11.0"S, Longitude 35°55'58.0"E and an altitude of 2,238 m above sea level. The area falls in agro ecological zone; Lower Highland 3. The experimental site receives mean annual rainfall of 1200 mm with a bimodal rainy season. Long rains are received from April to August while short rains are from October to December each year. The temperatures lie between 10.2 and 22.0°C while the soils are mollic andosols (Ngetich et al., 2014).

Experimental procedure

The cuttings were collected from two regions which were; Taita Taveta County in Coastal province and Busia County in Western region, Kenya in January 2022 from trees scattered in farmlands. The rooting medium used in the whole experiment was composed of sand (approximately 2-3mm diameter) which was collected from a riverbed. The nursery containers were 9*16-inch length and width respectively and these were placed inside the polyethene sheet tunnel (Figure 1) to maintain humidity and positioned under a shade screen of trees growing within the nursery.

Watering was done three times per week until the end of experiment. To obtain cuttings, a cut in the base, or heading to the tip of the branches depending on the nature of each mother plant was done using sterile pruning scissors in the early morning. The cuttings were misted with water and stored in the cooler boxes with ice inside for one day from the site of collection to the planting site prior to propagation. The shoots were severed into cuttings and the foliage was removed.

The stem bases of the cuttings were dipped into a rooting powder ('Seradix No.2'with 0.6% IBA, Amiran Itd, Nairobi, Kenya) prior to insertion in the nursery containers filled with a sand rooting medium. All plant materials were of local origin. The lengths of the cuttings were chosen based on nodes arrangement and also to obtain maximum number of cuttings per cutting position on the shoot, and the cutting lengths were; 3, 6 and 9 cm.

A total of 270 cuttings from each region (90 cuttings per replicate) were taken, and inserted in two blocks with 10 cuttings per treatment per block.

The total cuttings from the two regions were 540 cuttings. Cuttings were assessed monthly for the presence of new shoots, number and length of new sprouts and mortality. Seven cuttings per treatment were randomly sampled and the cuttings were allowed to grow for five months before the final data was collected. Data collected was subjected to analysis of variance (ANOVA) using General Linear Model for factorial design to obtain the p value of the effect of each treatment using GENSTAT 15th edition (Adams et al., 2016). The treatments which were found to be significant were separated using Standard Error of Difference (SED) at $p \le 0.05$ level of significance. Cuttings were defined as dead when either shrivelled or severely rotted.



Figure 1. Polyethene sheet tunnel for propagation of *M. excelsa* cuttings. Source: Author

RESULTS

Survival of cuttings

The survival of sprouted stem cuttings (Figure 2) was considered at the end of experiment after counting all dead cuttings. None of the cuttings obtained 50% of survival; hence the highest survival percentage obtained was 48.1% from cuttings with 9cm length. Cutting length was significant (p<0.05) on percentage of survival of stem cuttings at the end of experiment, df=2, F=26.58, P-value=0.001. The survival of cuttings obtained from 6cm length reached 41.27% while 3cm length attained 31.75% which was the least performed (Table 1).

Effects of cutting length on sprouting of *Milicia* excelsa

The three length of stem cuttings of *M. excelsa* responded differently after propagation. It took 28 days for the cuttings to start showing a sign of sprouts where the cuttings of 9 cm length showed high sprouting rate compared with the other cutting lengths. The effect of cutting length had a significant effect on the number of sprouts of cuttings produced among different levels of length throughout the experiment, F=43.45, df=2, p-value= 0.001 (Table 2). The cuttings with 9 cm length and 6 cm length had high mean values of sprouts from 30 days to 120 days while the cuttings of 3 cm length increased only from 30 days to 90 days as follows; 0.30, 0.57 and 0.79 respectively. The cuttings of 6 and 9 cm

length had reduced in the mean values of sprouts at 150 days which was at the end of experiment, and the 3 cm length cutting reduced from 90 to 150 days.

The effect of cutting length was significant on height of sprouts produced by the cuttings throughout the experiment, df=2, F=48.25, p-value=0.001 (Table 3 and Figure 3).

The mean height of sprout of the cuttings in all three length levels increased from 30 days up to 120 days, as follows; 0.59, 1.37, 2.27 and 2.80 respectively and the mean height of the cuttings reduced (2.6746) at the last month of the experiment.

DISCUSSION

The different sizes of cutting lengths had a significant effect on the sprouting of *M. excelsa* in this study. In propagation studies with a range of tree species, rooting and sprouting has been found to be positively correlated with cutting lengths (Ambassa, 2021). This can be explained by the fact that longer cuttings have larger reserves of carbohydrates and mineral nutrients which are required for root and shoot development. The longer cuttings attained longer height of sprouts as well as a greater number of sprouts. This might also be attributed to the fact that the shorter cuttings may have used all the nutrient reserves available resulting to severe competition compromising growth of sprouts (Meunier et al., 2016). The poor performance of shoot production in this study

may have resulted due to the older cuttings because the tree age was not specifically known during the collection

Cutting length (cm)	Cutting survival (%)		
3	31.75c		
6	41.27b		
9	48.41a		
CV	17		
SE	6.88		
P Value	<.001		

Table 1. Effect of cutting length on survival of cuttings.

Source: Author



Figure 2. Sprouted cuttings of *M. excels.* Source: Author

of cuttings from the two sites. Some earlier researchers have revealed that the ease of adventitious root formation in stem cuttings declines with the age of ortet (Hartmann et al., 1990; Ambassa, 2021).

This is an important problem because desirable phenotypic characteristics are generally not expressed until a plant has reached a considerable size or maturity. This also implies that by the time individuals who are genetically resistant to Phytolyma lata gall attack can be identified, they may have lost their rooting ability (Djagbletey et al., 2011).

Contributory factors may be a reduction in the supply of endogenous auxin, carbohydrate or nitrogenous

substances, or a decline in meristematic activity with increasing tree age, increasing sclerification as the tree ages may also be influential (Apetorgbor et al., 2004). When there is a high potential for rooting, a rooting hormone like IBA encourages rooting, but when there is a low possibility for rooting, it may have less effect or even become mildly inhibitive (Kesari et al., 2009).

The physiological state of the stock plant has an impact on its capacity to shoot as well as cultural factors and maturation (Ozel et al., 2005). Rooting hormones boost the proportion of cuttings that take root, speed up the process of root formation, encourage more cutting roots and improve the uniformity of cutting roots. However,

		N	lumber of sprou	its		
Cutting length (cm)	Interval of data collection					
	30 days	60 days	90 days	120 days	150 days	
3	0.30 ^c	0.57 ^c	0.79 ^c	0.74 ^b	0.43 ^c	
6	0.56 ^b	0.77 ^b	1.07 ^b	1.15 ^a	0.65 ^b	
9	0.65 ^a	0.98 ^a	1.34 ^a	1.39 ^a	0.88 ^a	
CV	5.70	16.90	13.00	44.10	20.50	
SE	0.056	0.130	0.14	0.48	0.13	
P Value	<0.001	<0.001	<0.0001	0.001	<0.001	

Table 2. Effect of cutting length on the number of sprouts.

Means on the same column having different superscripts are significantly different at P<0.001. Source: Author



Figure 3. *Milicia excelsa* cuttings height levels. Source: Author

higher concentrations of IBA can lead to inhibition of shoot production and rotting on mature cuttings (Ozel et al., 2005). When compared to greenhouses or mist propagation chambers, the use of polyethylene sheet tunnels for Milicia cuttings propagation is a successful, less expensive method (Akakpo et al., 2014). The bacterial and fungal infection may potentially be the cause of poor performance of shoot production which showed some symptoms of darkening of the lower portions of the cuttings, rotting and shedding of new sprouts. This may have affected the performance of cuttings towards the end of experiment at 150 days after propagation. This observation corroborates with previous findings by De Almeida et al. (2017) who stated that bacterial and fungal diseases are frequent during the multiplication of rooted cuttings and can reduce the cuttings' survival and ability to root if not properly controlled.In the current study, the cuttings used were lignified at different degrees of maturity since the exact age of cuttings were not known with the less mature ones developing molds leading to death while the strongly lignified ones dried up.

Indeed, the quality of the cuttings is quite dependent on the stage of their development at the time of their

collection (Daïnou et al., 2012a). Lignified cuttings seem to contain few nutrients reserves to ensure their resprouting, which could compromise the formation and development of roots and shoots, and reduce the viability of budding buds. However, the rooting environment is one of the most important factors affecting the rooting success of cuttings (Daïnou et al., 2012b). This is in accordance with previous works by Agbogan et al. (2014), who specified that temperature and water are external and important factors for germination in a medium. The amount of water for irrigation regrettably seems to have been insufficient to induce the good growth of the cuttings in this study. On the other hand, the plants resulting from the cuttings have genetic program as their mother stock and return relatively to production according to the cropping calendar of the stock of origin (Nguema et al., 2013) thus, the result obtained on the cuttings of *M. excelsa* would probably be of genetic origin. Several earlier studies have demonstrated that the ability to root or to shoot is affected by the genotype (Ouinsavi et al., 2010; Bizoux et al., 2009). Previous research on the vegetative propagation of tropical tree species using stem cuttings has identified varying factors that can be optimized to increase rooting

	Height of sprouts (cm) Interval of data collection					
Cutting length						
	30 days	60 days	90 days	120 days	150 days	
3	0.24 ^c	0.75 ^c	1.43 ^c	1.52 [°]	1.37 ^c	
6	0.40 ^b	1.19 ^b	1.97 ^b	2.38 ^b	2.20 ^b	
9	0.59 ^a	1.37 ^a	2.27 ^a	2.80 ^a	2.67 ^a	
CV	5.70	5.90	12.80	15.00	12.90	
SE	0.05	0.07	0.24	0.33	0.27	
P Value	<.001	<.001	<.001	<.001	<.001	

Table 3. Effect of cutting length on the height of sprouts.

Means on the same column having different superscripts are significantly different at (P<0.001). Source: Author

rate in tropical trees, including cutting length, cutting positions, auxin concentration and sowing media (Husen and Pal, 2006). The capacity of cuttings to root may also be impacted by cutting length, which will influence the depth of insertion into the rooting medium. The factor that affects a tropical trees ability to take root successfully is the location of the cutting within the shoot (Agbo and Obi, 2007).

The cuttings were taken towards the tip of the branches in this study. The variation is a function of concentration of some hormones that run from base to the apex of stem tissue in relation to the chronological age such as; leaf water potential, leaf carbon balance, stem lignification, carbohydrate content and respiration (Agbo and Obi, 2007). Rooting of stem cuttings with reproductively matured ortet or stockplant typically has the benefit of shortening the period of a plant to reach maturity in addition to providing genetic benefits (Olaniyi et al., 2021). Cutting length is an exogenous factor that affects sprouting activity in this study, and can be attributed to more food (carbohydrate) reserves in the longer cuttings compared to shorter cuttings. Similar findings were reported in Khaya ivorensis (Tchoundjeu and Leakey, 1996) and Picea abies (OuYang et al., 2015) where long cuttings rooted significantly better than short cuttings. These findings were also in agreement with Naidu and Jones (2009) who attributed large storage reserve and photosynthetic area of longer cuttings to rooting success of Eucalyptus hybrid clones.

Conclusion

The study concluded that the different length sizes had a significant effect on the number and height of sprouts obtained on the cuttings of *Milicia excelsa*. This therefore means that before planting *M. excelsa* cuttings, there is consideration of length of cuttings suitable for planting as in this study showed that the 9 cm length performed better compared to other shorter lengths in the experiment and also showed higher percentage of cutting

survival. Further studies can be conducted on factors influencing shoot and root production of cuttings of *M. excelsa*, such as; developing vegetative propagation techniques which would enable pest-resistant genotypes to be multiplied, providing suitable materials for reforestation, and future studies on the reduction of diseases and pests which hinder the good performance of cuttings.

CONFLICT OF INTERESTS

The author has not declared any conflicts of interests.

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