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Propagation of *Pentaclethra macrophylla* Benth (Fabaceae) through seed and rooting of leafy stem cuttings

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Key words: Auxin, hormone, physiology, scarification, seed position.

Abstract

We assessed a number of factors controlling seed germination and rooting success of leafy stem cuttings of a high priority fruit (food and medicine) tree species from West and Central Africa, i.e. *Pentaclethra macrophylla* Benth (Fabaceae). In the case of seed germination, we tested three substrates (soil, sand, and a mixture of soil + sand (2:1)), mechanical seed scarification and seed position in the pod (proximal, median and distal), while for leafy stem cuttings, the two factors tested were substrate (sawdust, river sand and the mixture of sawdust + river sand (1:1)), seradix-2 (an auxin used for rooting stimulation, compared to a control) and type of auxin (naphthalene acetic acid (NAA), indol butyric acid (IBA) and indol acetic acid (IAA)). The study was conducted in Yaoundé, Cameroon in 2011. Data analysis shows that there was no significant difference between germination substrates ($P = 0.445$). Meanwhile, mechanical scarification significantly increased germination percentage ($P = 0.04$), whereas seed position induced significantly higher germination rates with the median position (100%, $P > 0.001$). Concerning leafy stem cuttings, the study showed sawdust to be the best rooting medium followed by river sand and mixture. Rooting percentages were at 60.26 ± 6.22%, 32.22 ± 6.04% and 25.27 ± 5.67%, respectively ($P < 0.001$). Seradix-2 and type of auxin had significant effects on rooting ability of leafy stem cuttings ($P = 0.05$ and $P = 0.002$, respectively). NAA and IBA were the best hormones. Our results show *P. macrophylla* is moderately amenable to rooting of leafy stem cuttings. Choosing seeds from median position in a pod can optimize germination percentage.

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Introduction

Pentaclethra macrophylla Benth (Fabaceae); commonly called "Ebaye" in Cameroon is a tree species of the humid forest zone of Africa. It has hermaphrodite flowers, producing long, suspended woody and dehiscent fruits. In general, mature *P. macrophylla* trees reach 30 m in height, with the main trunk at 80 cm in diameter (Eyog Matig *et al.*, 2006). The species occurs naturally from Senegal to Democratic Republic of Congo passing through southeastern Sudan and Sao Tomé et Principe Islands (Ladipo and Boland, 1995; Vivien and Faure, 1996; Oboh, 2007).

P. macrophylla has gained interest in West and Central Africa over the last years because it is the source of products that have important nutritional and medicinal values. Oboh (2007) reported that in Africa humid forest zone, seeds are boiled or roasted and consumed in several household. In Southern part of Nigeria, seeds are fermented to obtain a spice with the taste closely to that of meat called *Ugba*. Also from seeds, vegetable cooking oil can be extracted. This justifies why the species is also called "oil bean tree" (Gill, 1992). The seed oil contains up to 30 – 36 % of protein, and is suitable for soap and candle making (Ehiagbonare and Onyibe, 2008). Mature fruits of *P. macrophylla* are used for the treatment of both human and animal diseases (Oboh, 2007). Extractions of leaves, barks, seeds, and seed pulp have anti-inflammatory and analgesic properties, and are used to treat gonorrhea and convulsions. Roots are also used as laxative and for treatment of dysentery. In Cameroon, a decoction from the bark is used to interrupt pregnancy. *P. macrophylla* is also used to fend off evil spirits (Koné *et al.*, 2008). In Cameroon, the creation of a market information system for non-timber forest products (NTFPs) has greatly promoted the exploitation and commercialization of *P. macrophylla* as a species with high commercial, nutritional and medicinal potential (Ngueko, 2008). Recently, a new market has been developed for the species in Nigeria with about fifteen millions potential consumers (Mbong, 2009).

P. macrophylla is victim of unsustainable exploitation characterized by the burning and destruction of seedlings during agricultural practices, predation of seeds by wild animals, and overexploitation of bark by farmers and traditional medical practitioners. Then, if no precautions are taken to preserve the species, serious genetic erosion will occur with severe negative consequences for both stock and price of derived products. Mandeng (2009), in collaboration with farmers, identified the species' domestication as a crucial alternative to address this issue which is in line with Leakey (2001) regarding the importance of domestication in the conservation of neglected indigenous species.

Related to the domestication of this species, tree-to-tree phenotypic variation has been assessed (Tsobeng *et al.*, 2012) to identify superior trees to be used as parents for producing improved materials. As far as propagation is concerned, research on factors controlling vegetative or generative propagation in order to improve production and planting material is still under way. Since the seeds of this species as well as those of many Fabaceae are characterized by seed coat dormancy, they need pretreatment to allow and initiate germination. Ehiagbonare and Onyibe (2008) worked on the effect of temperature on seed germination and seedling radicle length, and on the effects of soaking seeds in tap water, in sulfuric acid and in coconut milk. With these methods, the authors were able to the seed germination of this species with the percentage of 86%. Previous studies demonstrated the existing of several pretreatment methods to release dormancy (Hartmann *et al.*, 2002). Farmers or others stakeholders involved in nursery production will not be then able to always practice such methods regarding the differences capable to exist between sites in terms of context.

So, diversifying pretreatment methods mainly by testing some which are most common to farmers would be very useful. Amongst the latter, we think of: (i) modulating the germination substrate as the latter has been shown to have an effect on germination of several species such as *Prunus africana* (Avana, 2006); (ii) mechanical seed scarification as demonstrated by Ibiang *et al.* (2012) as factor influencing *Tetrapleura tetraptera* seed germination; and (iii) seed position in pod as demonstrated by Nkongmeneck *et al.* (1996) and Ibn Tattou (1981) as factor controlling germination of *Tetrapleura tetraptera* and *Medicago* spp. repectively.

The availability of seeds for germination is still a big constraint because they are highly collected by traders or consumed by wild animals. To address this issue, vegetative propagation, specifically using leafy stem cuttings, has been suggested by farmers, since in addition to transferring characters from mother trees to descendants (Hartmann and Kester, 1983),

facilitates the mass production of selected materials (Tchoundjeu, 1989).

Hartmann and Kester (1983) and Leakey (2004) stated that the success rate of propagation by leafy stem cuttings depends on some factors such as rooting medium and use of hormones as they can influence propagation success of these cuttings. Their impact for the propagation by leafy stem cuttings has been successfully tested on several species such as *Khaya ivorensis* and *Lovoa trichilioides* (Tchoundjeu, 1989); *Ricinodendron heudelotii* (Schiembo *et al.*, 1997); *Dacryodes edulis* (Mialoundama *et al.*, 2002); and *Allanblackia floribunda* (Atangana *et al.*, 2006). To our knowledge, this technique has not yet been tested on *P. macrophylla*. Hence, the aim of this study was to assess the effects of (i) three rooting media (decomposed sawdust, river sand, mixture of sand and sawdust) (ii) Seradix-2 (compared to control) and (iii) three types of hormone (Indol Butyric Acid (IBA), Indol Acetic Acid (IAA) and Naphtalen Acetic Acid (NAA)) on rooting ability of *P. macrophylla*.

Materials and methods

Site, plant material and treatment

This study was conducted in 2010 at ICRAF research nursery in Nkolbisson (3°52'N, 11°26'E), near Yaoundé, Cameroon. The site has an equatorial type climate, with an average temperature of about 25 °C and a bi-modal rainfall regime ranging from 1500 - 2000 mm per year. The experiment was carried in a shade house which had a mixture of translucent and corrugated iron roofing sheets bordered with shade cloth allowing about 542 µmol M^2 S^{-1} of irradiance.

Seed germination

Healthy seeds were collected around/on mature stems of *P. macrophylla* from the wild. They were transported to the nursery shade house, dried during three days before sowing in seed germination bed. Seed beds were buckets containing about fifteen liters of substrate, arrayed under the shade house. Each bucket was sprayed every day in the morning with about 500 ml of water (based on the field capacity of the soil) to improve the seed germination conditions (Hartmann *et al.*, 2002). The particular nursery conditions and experimental design used for each experiment can be defined as follow:

Effect of substrate on untreated seed germination ability: untreated seeds were sowed following a completely randomized design. Treatments tested were river sand, sawdust, ground and mixture ground + river sand (2:1). The number of seeds per treatment was eighty with a seed as experimental unit.

Effect of pretreatment on seed germination ability: the tested treatment was a mechanical scarification (that consists of enlarging the micropyle of the seeds to about 4 mm diameter using sandpaper). This treatment facilitates the infiltration of oxygen and water, main drivers of the growth of embryo. The experimental design was the completely randomized. The control consisted of unscarified seeds. The number of seeds used was forty per treatment. The substrate used was the mixture ground + river sand (2:1).

Effect of seed position in the pod on seed germination ability: for this experiment, scarified seeds collected from different position in the pod (collected from the healthy mature trees) were sowed using the substrate composed of mixture ground + river sand (2:1). The different modalities were proximal, median and distal (Fig. 1). The experiment was conducted following a completely randomized block design with a seed as experimental unit. Three blocks were considered with thirty seeds per treatment and per block. Total number of seeds per treatment was ninety.

Leafy stem cuttings

The experiments were carried out in a non-mist propagator, developed by Leakey *et al.* (1990), installed under the shade house. Cuttings of *P. macrophylla* were collected in the nursery on nine months old seedlings. Before collecting the cuttings, young plants were watered early in the morning. All cutting tools (pruners, knives) were disinfected before use. Semi-lignified parts were cut and conserved in a disinfected humid polyethylene bag. Leaves were cut back to 50 cm^2 and stems to 3-5 cm in length, with a circular base to guarantee homogenous distribution of rooting and a slant-wise upper part to facilitate runoff of water during spraying (Tchoundjeu, 1989). For the rooting medium trial, Seradix-2 which is an indole-3-butyric acid was applied to the base of each cutting. For Seradix-2 (1µg/cutting) assessment, the rooting hormone was applied to the base of each treated cutting (except control). For type of hormone experiment, auxins were prepared by dissolving 0.05 g of pure auxin powder in 10 ml of 90% industrial alcohol. Auxins were applied to the base of the leafy stem cuttings using a micrometer syringe following

method described by Tchoundjeu (1989). All three auxins were applied as 10 µg drop to the cutting base. Treated cuttings were at once set randomly on sawdust (1-1) that had previously (seven days before) been disinfected with insecticide (cypermethrine, 50 ml diluted in 16 liters of water) and fungicide (dimethoate, with dilution of 50 g in 16 liters of water). In order to keep cuttings cool, a light spray of water was applied on the foliage using a knapsack sprayer whenever the propagator was opened.

To achieve the objectives, three simple completely randomized block structures were used in three different trials. The three experimental treatments consisted of successive levels of the three factors namely: rooting media (decomposed sawdust, river sand, mixture of sand and sawdust), Seradix-2 (treated and control) and type of hormone (Indole – 3-butyric acid (IBA), Indole-3-acetic acid (IAA) and 1-naphtalen acetic acid (NAA)).

Three blocks were used per experiment. In each block, twenty, thirty and eighteen cuttings were set for different rooting media, Seradix-2 and type of hormone, respectively. This yielded overall totals of one hundred and eighty, one hundred and eighty and one hundred and sixty-two leafy stem cuttings for the whole set of trials, respectively.

Every morning before 10.00 am, the propagator was opened and fallen leaves were removed to avoid infection development. The propagator's water level was checked and adjusted accordingly, while the transparent plastic was cleaned to facilitate sunlight access inside the propagator and cuttings sprayed with water. After two weeks, weekly assessments of rooting were carried out. Data collection ended after ten weeks for the first two trials and sixteen weeks for the third trial (type of hormone). The cutting was said to have rooted when it had one or more roots exceeding 1 cm length. Rooted cuttings were removed from the experiment; their roots were counted before putting each of them into a polyethylene bag containing a 2:1 mixture of agricultural-field top soil and river sand.

Data collection and statistical analysis

Seed germination: every week, each bucket was visited, the response of each seed was coded for germinated (1) or not (0) and registered in view of assessing percentage of germinated seeds, latency time (time in days that elapsed between sowing and germination of the first seed) and germination speed (time necessary to reach 50% of percentage of germination) (Bonner, 1998).

Leafy stem cutting: the response of each stem cutting was coded for rooted or dead [rooted (1) or not (0), dead (1) or not (0)]. The number of roots of rooted cuttings was counted. Dead cuttings were removed from the propagator while live ones were re-set for further observation the following week. At the end of the trial, percentage of rooted cuttings, percentage of dead cuttings and the mean number of roots per rooted cuttings were estimated for each treatment.

To determine the effect of experimental factors on seed germination or rooting ability of cuttings, percentages of seeds germinated, of rooted cuttings and of dead cuttings, and mean number of roots developed per cutting collected were subjected to an analysis of variance using Linear Regression Model procedures of Genstat V.13 software. Specifically, percentages of seeds germinated, of cuttings rooted and of dead cuttings were assessed using Logistic Regression while a Log Linear Regression model was fitted to the root number data. Factors having a significant effect on germination percentage, rooting percentage, mortality percentage and average number of roots per rooted cutting were compared among treatment levels using the Least Significant Difference (LSD) procedure, considering a confidence interval of 0.05.

Results

Seed germination

a1) Effect of substrate on seed germination ability

Results obtained from this experiment showed germination started after one week and continued for fifteen weeks. None significant difference between substrate was observed at the end of the observations ($P = 0.445$). The highest percentage of germination was registered in the mixture of ground + river sand ($47.79 \pm 5.27\%$) followed by river sand, ground and sawdust ($45.56 \pm 5.25\%$, $38.89 \pm 5.14\%$ and $37.78 \pm 5.10\%$, respectively).

a2) Effect of pretreatment on seed germination ability

At the end of this experiment, a significant difference was observed between scarified and control seeds ($P = 0.041$). Germination started after one week for treated seeds versus three weeks for the control. After fifteen weeks, scarified seeds had germinated better than control with values at $46.33 \pm 3.72\%$ and $38.33 \pm 2.62\%$, respectively (Fig. 2).

a3) Effect of seed position in the pod on seed germination ability

Overall germination started five weeks after sowing and continued for twelve weeks (Fig. 3). At the end of the observations, the analysis of variance showed highly significant effect of seed position on percentage of germination ($P < 0.001$). The median position had greatest percentage of germination (100%). Related to this parameter, proximal and distal positions were not significantly different from each other (84.44 ± 6.19% and 74.44 ± 5.09%, respectively).

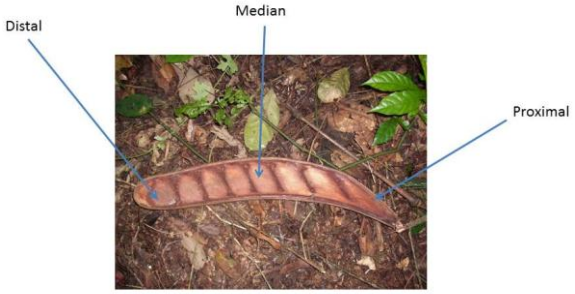

Fig. 1. Position of seeds in a pod.

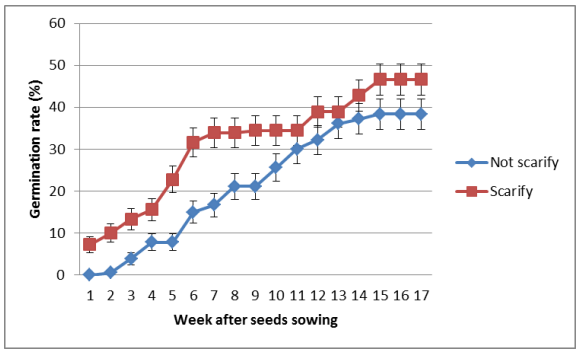

Fig. 2. Effect of scarification on germination ability of *P. macrophylla* seeds.

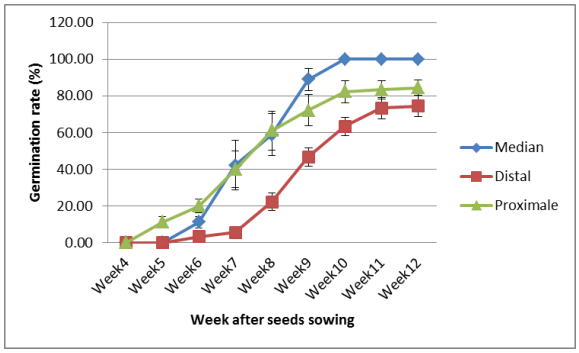

Fig. 3. Effect of seed position on germination ability of *P. macrophylla* seeds

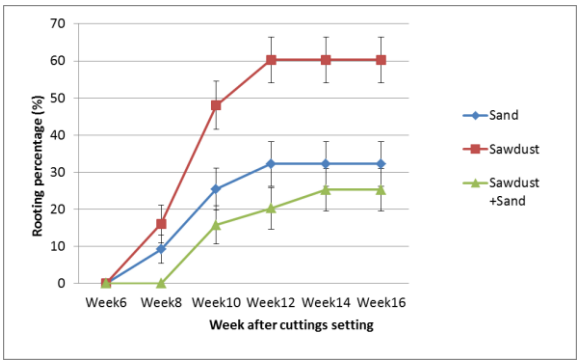

Fig. 4. Effect of rooting medium on percentage of rooted cuttings of *P. macrophylla*.

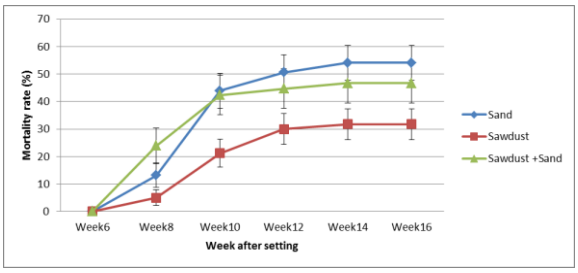

Fig. 5. Effect of rooting medium on the percentage of died cuttings of *P. macrophylla*.

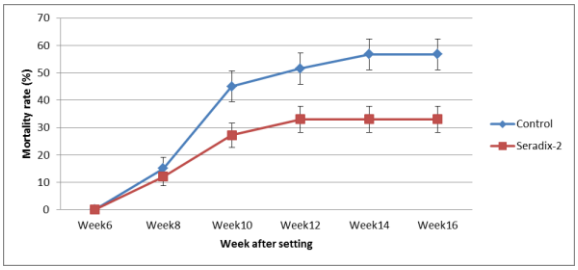

Fig. 6. Effect of seradix-2 on the percentage of rooted cuttings of *P. macrophylla*.

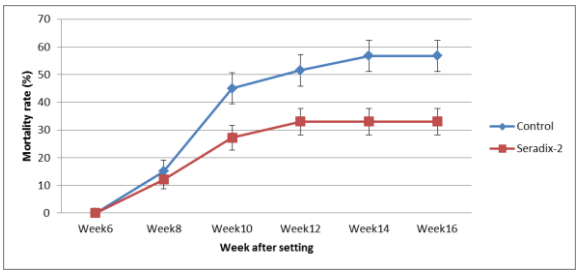

Fig. 7. Effect of seradix-2 on percentage of died cuttings of *P. macrophylla*.

Leafy stem cuttings

b1) Effect of rooting medium on rooting ability of leafy stem cuttings of *P. macrophylla*

Results obtained on percentage of cuttings rooted show that differences between various rooting medium were highly significant ($P < 0.001\%$) at sixteen weeks after cutting setting. Rooting started in week six and continued until week fifteen, after which no rooting was recorded. The best rooting medium was sawdust followed by river sand, and mixture of

river sand and sawdust (1:1). There was no significant difference between river sand and sawdust (Fig. 4). The percentages of rooted cuttings in the three rooting media were 60.26 ± 6.22%, 32.22 ± 6.04% and 25.27 ± 5.67%, respectively.

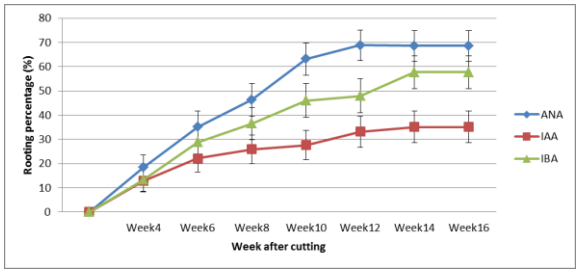

Fig. 8. Effect of type of hormone on the percentage of rooted cuttings of *P. macrophylla*.

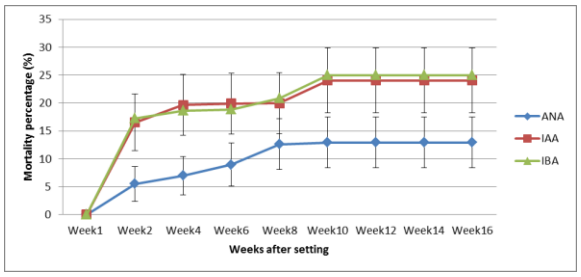

Fig. 9. Effect of type of hormone on the percentage of died cuttings of *P. macrophylla* leafy stem cutting.

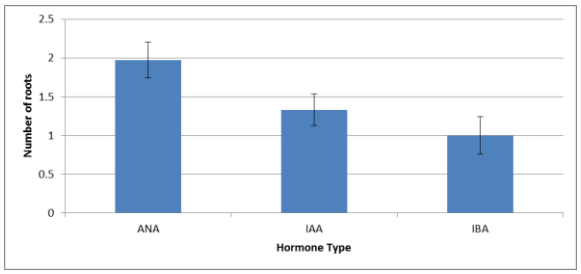

Fig. 10. Effect of type of hormone on the mean number of roots per rooted cutting.

For the percentage of died cuttings, the difference between river sand, sawdust and mixture was significant (P = 5.00%). Contrary to the percentage of died cuttings, both sand and mixture presented the highest mortality percentages (54.07 ± 6.35% and 46.74 ± 7.23%). Sawdust presented the significant lowest mortality rate (31.76 ± 5.60%) recorded at week fifteen after cutting setting (Fig. 5).

For the mean number of roots per rooted cutting, the difference between treatments was not significant (P = 76.10 %). The average numbers of roots per rooted cuttings set in sawdust, sand and mixture were respectively 1.10 ± 0.24, 1.00 ± 0.22 and 1.22 ± 0.22.

b2) Effect of Seradix-2 on rooting ability of *P. macrophylla*

Rooting started after eight weeks and continued until week twelve. At the end of the rooting process, the analysis of variance showed significant differences between treated and untreated cuttings (P = 5.00%). Percentage of rooted cuttings was higher in treated cuttings compared to control (44.65 ± 4.79% and 31.5 ± 5.02%, respectively) (Fig. 6).

The percentage of died cuttings was significantly affected by Seradix-2 (P = 0.2%). The highest percentage of dead cuttings was observed in control (56.74 ± 5.6%) compared to treated with Seradix-2 (33.01 ± 4.81%). This trend was observed from week ten until week fourteen (Fig. 7).

As observed in the effect of rooting medium, analysis of variance indicated that average number of root was not significantly influenced by the use of Seradix-2 (P = 81.7) at the end of this experiment (week twelve). The mean number roots per rooted cuttings remained low (1.17 ± 0.22 and 1.13 ± 0.19, respectively for treated and untreated cuttings).

b3) Effect of type of hormone on rooting ability of *P. macrophylla*

When rooting commenced at week four, difference between NAA, IBA and IAA was no perceptible until week eight. From week ten to week fourteen, this difference became significant (Fig. 8) with the probability of 0.20% at week fourteen with the greatest rooting percentage observed with NAA and IBA (68.63 ± 6.30% and 57.68 ± 6.85%, respectively) while the lowest rate was observed in IAA (35.14 ± 6.50%).

Mortality started at week two with the highest percentages observed in IBA and IAA. This difference remained significant until week ten (P = 0.021). The treatments with the highest percentage of died cuttings were IBA and IAA (25.00 ± 5.99% and 24.50 ± 5.8%, respectively) compared to NAA that presented the lowest percentage of died cuttings (12.94 ± 4.57%) (Fig. 9).

The ANOVA of the mean number of roots per rooted leafy stem cutting showed the significant effect of type of hormone (P = 0.012) fourteen weeks after cuttings setting. The highest mean number of rooted per rooted cutting was observed on those treated with NAA (1.97 ± 0.23) followed by IAA (1.33 ± 0.2) and IBA (1.00 ± 0.24) (Fig. 10).

Discussion

Seed germination

This study was not the first conducted on seed germination on this species. However this was a pioneer study testing the effect of sand, sawdust, soil and a mixture of soil + river sand (2:1), mechanical scarification and seed position in the pod on germination ability of *P. macrophylla*.

To germinate, a seed's embryo needs water and oxygen. After development of the first leaves, light is necessary in starting/initiating the photosynthesis process (Meyer, 1998). Factors influencing embryo germination are classified into intrinsic and extrinsic. The main extrinsic or environmental factors are humidity, temperature, oxygen and light which are also controlled by the nature of the germination medium (Hartmann *et al.*, 2002). Its richness in pores improves the circulation of water and oxygen, while its abundance in organic matter improves its aptitude to conserve water or humidity. For that purpose, the study of the 'best' germination medium is of great concern for each studied species due to the fact that each species' seeds need particular conditions to allow the embryo to germinate (Dun *et al.*, 2006). This is for example the case of *Garcinia mangostana, Hevea brasiliensis, Mangifera indica, Persea americana, Theobroma cacao* (Chin *et al.*, 1984) and *Prunus africana* (Avana, 2006) with highest germinating rate observed in the river sand. *Allanblackia floribunda* with a very good germination rate in a substrate composed of mixture ground + river sand (2:1) (World Agroforestry Centre, 2006). These results are not in line with those obtained for *P. macrophylla* which showed no significant difference between substrates. This could be due to the comparable influence of the targeted substrates on the germination ability of this species.

Baskin and Baskin (2004) described five types of dormancy: physical, physiological, morphological, morpho-physiological and combined dormancy. Given the characteristics of each type, ebaye seeds seem to be in the category of physical dormancy because of the presence of a solid pericarp (Ehiagbonare and Onyibe (2008), consequently, need a pretreatment to accelerate germination since it facilitates the infiltration of oxygen and water in the kernel to trigger the germination process. Related to physical dormancy, several pretreatments can be applied such as scarification (Hartmann *et al.*, 2002). This result is in line with those reported by Karaguzel *et al.* (2004) and Armin *et al.* (2010) for *Lupinus varius* and *Citrullus lanatus* respectively. It could also explain why the scarified seeds germinated more rapidly and had the best percentage of germinated seeds in the present study. However, scarification is not the only way to proceed. In fact, Ehiagbonare and Onyibe (2008) cited a temperature treatment as another way to pretreat seeds of ebaye. With higher temperature germination rate was very high (about 90 – 100%) (Ehiagbonare and Onyibe, 2008). The positive effect of pretreatment of seeds has been registered on several tropical tree species such as *Prunus africana* on which partial remove of pericarp reduced the latency period from thirty to twenty-five days whereas it increased germination rate from 28 to 39%. It was also documented for *Tetrapteura tetratplera* on which the mechanical or chemical scarification increased percentage of germinated seeds from 18.3 to about 85% (Ibiang *et al.*, 2012).

Regarding the germination ability of seeds in terms of its position in the pod, the present study showed the greatest germination of seeds occupying the median position followed by proximal and distal seeds. A similar trend was described by Ibn Tattou (1981) on *Medicago* spp. and by Nkongmeneck *et al.* (1996) on *Tetrapleura tetraptera*. According to these authors, the differences should be explained by way seeds are placed in a pod. Nkonmeneck *et al.* (1996) have demonstrated the heaviest seeds to occur in the median zone compared to proximal and distal seeds, and allocated this best germination to the weight of the seeds that is supposedly highest in this position. As it is, germination is starting with a respiration phase during which reserves (carbohydrates) are transformed into various components such as energy to be used by embryo to start the active germination phase (Raven *et al.*, 1992). The higher this reserve, the more respiration products are generated, consequently the seed will germinate more easily (Upadhaya *et al.*, 2007). This suggests that we should quantify carbohydrate content of seeds in different positions to better understand the relationship between position, carbohydrates reserve and germination rate (Batin, 2011).

Leafy stem cutting

With respect to rooting medium, *P. macrophylla* showed similar responses as other tropical tree species such as *Ricinodendron heudelotii* (Schiembo et al.,1997), *Gnetum africanum* (Schiembo *et al.*, 1996a), *Irvingia gabonensis* (Schiembo *et al.*,1996b)

and *Milicia excelsa* (Ofori, 1994), all of which displayed highest percentage of rooted cuttings and lowest percentage of died cuttings in sawdust. Our results disagree with the findings of Atangana *et al.* (2006) on *A. floribunda* as they demonstrated the best performance with river sand. Tsobeng *et al.* (2011) working with on *Dyospyros crassiflora* were unable to find any significant difference between sawdust and mixture of river sand and decomposed sawdust. These controversial results confirm the affirmation made by Leakey *et al.* (1990) who highlighted the difference between performances of different rooting medium. He attributed this difference to some internal factors controlling rooting ability such as oxygen, pH, water and porosity which are generally different from one substrate to another. In fact, as described by Loach (1988) and Leakey and Newton (1994), these factors may affect tissue respiration and cell dedifferentiation at the base of the cutting from where roots appear. The difference pointed between different species can enable to conclude that the interaction between substrate and physiology or genetic characteristics should play a considerable role. This could explain the dissimilarity of the results of different species in the same substrate.

The results of the experiment with Seradix–2 show that hormonal stimulation increased the rooting percentage of *P. macrophylla* leafy stem cutting. These findings are in line with the statement of Leakey (2004), who after a literature review of the contribution of exogenous auxins to modification of plant physiology, concluded that typically, cuttings treated with auxins root better than untreated one. The auxins' effects on the rooting percentage of *I. gabonensis* (Schiembo *et al.*, 1996b), *Dacryodes edulis* (Mialoundama *et al.*, 2002) and *Allanblackia floribunda* cuttings (Atangana *et al.*, 2006) were previously described. Seradix-2 is known as an exogenous IBA, which is an adventitious root-stimulating hormone. Naturally, plants during photosynthetic periods synthesize endogenous auxin, which is transported to the base of the cutting and promotes adventitious root formation (Davis, 1988; Hilt and Bessis, 2003). So, the positive effect of hormone might be due to an increase of plant auxin by addition of Seradix – 2 (containing 0.3% of IBA). This effect is not always positive due to the fact that, referred to the experiment conducted on many species such as *R. heudelotii* (Schiembo *et al.,* 1997) and *Triplochiton scleroxylon* (Leakey *et al.*, 1982), the effect of exogenous hormones can greatly be influenced by the physiological state of the cuttings during stimulation. If the stimulation comes with a surplus of auxin, this can be toxic for the cuttings. This explains why a trial should be set up in order to assess the effect of different doses of IBA as described by Schiembo *et al.* (1997) on *R. heudelotii* and Mialoundama *et al.* (2002) on *Dacryodes edulis*.

As far as hormone concentration is concerned, type of hormone can influence rooting percentage and number of roots of leafy stem cuttings. This is the case for *P. macrophylla* where NAA was found as best hormone compared to IBA and IAA. Leakey (2004) already indicated that IBA is effective in promoting root formation (at different concentrations). The author added that occasionally NAA can perform better, as observed in *Parkia biglobosa* (Teklehaimanot *et al.*, 1996). Also, a highest performance of NAA was described previously by Atangana *et al.* (2006) on *Allanblackia floribunda*. These results are different from those obtained by (Mialoundama *et al.*, 2002) on *Dacryodes edulis;* they found significant performance of IBA (compared to NAA and IAA). These differences between species can be explained in several ways: auxins are known as growth regulatory substances in plant tissues; amongst many other functions, they promote cell elongation and induce root growth. Their effect on the plants cell can be influenced by many factors such as type, genes and physiological state of the plant tissues (Harman *et al.*, 2002; Atangana and Khasa, 2008) which are different within different species (Woodhard and Bartel, 2005). The fact that the various types of hormone have different implications can be explained by their molecule structures which have dissimilar chemical reactions. A clear identification of distinctive effects and relative contribution of exogenous auxins on rooting ability of cuttings is needed, as some genes have been found to be induced in response to exogenous auxins in rooting ability of forest species (Sanchez *et al.*, 2007).

Previous studies highlighted that the increase in percentage of rooted cuttings is not always proportional to the increase in hormone concentration such as observed in *Triplochiton schleroxylon* (Leakey *et al.*, 1982), *Lovoa trichilioides* (Tchoundjeu, 2009) and *Prunus africana* (Tchoundjeu *et al.*, 2002). Consequently, as recommended with IBA, the quantification of ANA should be a next point to follow to optimize the rooting percentage of *P. macrophylla*.

Conclusion

The results obtained from the present study indicate that *P. macrophylla* can be successfully propagated by seed and leafy stem cuttings using a non-mist propagator. The fact that maximum rooting percentages of over 75 % were achieved suggests that the species could be multiplied by small-scale nurseries using leafy stem cutting technique when seeds are not available or during the selection process. The variation observed between the rooting percentages in the various experiments indicates the need to develop appropriate post-severance protocols such as NAA quantification and combination of factors to optimize rooting percentage.

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References

Armin M, Asgharipour M, Razavi-Omrani M. 2010. The effect of seeds priming on germination and seedling growth of water melon (*Citrullus lanatus*). Advance in Environmental Biology **4**(3), 501 – 505.

Atangana AR, Khasa DP. 2008. Preliminary survey of clonal variation in rooting of *Allanblackia floribunda* leafy stems cuttings. Canadian Journal of Research **38**(1), 10–15.

Atangana AR, Tchoundjeu Z, Asaah EK, Simons AJ, Khasa DP. 2006. Domestication of*Allanblackia floribunda*: amenability to vegetative propagation. Forest Ecology and Management **237,** 246–251.

Avana ML. 2006. Domestication de *Prunus africana* (Hook F.)Kalkam : Etude de la germination et du bouturage. Doctoral Thesis. University of Yaounde I, Cameroon, 165 p.

Baskin CC, Baskin JM. 1998. Seeds: Ecology, biogeography and evolution of dormancy and germination. New York Academic Press, 666 p.

Batin CB. 2011. Seed germination and seedling performance of *Jatropha curcas* L. fruit based on color at two different seasons in Northern Philippines. International Conference on Environment and Bioscience **21**, 94-100.

Bonner FT. 1998. Testing tree seed for vigour: A review. Seed Technology **20**(1), 5-16.

Chin HF, Hor YL, Mohl Lassim MB. 1984. Identification of recalcitrant seeds. Seed Science and Technology **12**, 429 – 436.

Davis TD. 1988. Photosynthesis during adventitious rooting. In Davis TD, Haissig BE, Sankhla N (eds): Adventitious root formation in cuttings. Advances in Plant Sciences Series, Vol 2., Dioscorides Press, 79 - 87.

Dun EA, Ferguson BJ, Beveridge CA. 2006. Apical dominance and shoot branching. Divergent opinions or divergent mechanisms? Plant Physiology **142**, 812 - 819.

Ehiagbonare JE, Onyibe HI. 2008. Regeneration studies on *Pentaclethra macrophylla* Beth. Scientific Research and Essay **3**(11), 531 – 536.

Eyog Matig O, Ndoye O, Kengue J, Awono A. 2006. Les fruitiers forestiers comestibles du Cameroun. IPGRI, 220p.

Gill LS. 1992. Ethno medicinal uses of plants in Nigeria. University of Benin, Benin City, Edo State, Nigeria, 184-185.

Harman TC, Taylor PJ, Walsh MP, La Forge BE. 2002. Quantum dot super lattice thermoelectric materials and devices. Science **297**, 2229-2232.

Hartmann HT, Kester DE. 1983. Plant propagation : Principles and Practices. Fourth edition, Prentice-Hall, Upper Saddle, New Jersey, USA.

Hartmann HT, Kester DE, Davies FT, Geneve RL. 2002. Plant propagation: Principles and practices. Seventh edition, Prentice Hall, Upper Saddle, River, New Jersey, USA.

Hilt C, Bessis R. 2003. Abscission of grapevine fruitlets in relation to ethylene biosynthesis. Vitis **42**, 1-3.

Ibiang YB, Ita EE, Ekanem BE, Edu NE. 2012. Effect of different pretreatment protocols on seed germination of *Tetrapleura tetraptera* (Schum and Thonn). Journal Of Environmental Science,

Toxicology And Food Technology Volume **2**(3), 25-29.

Ibn Tattou 1981. Biologie comparée de quelques *Medicago* annuelles. Etudes d'échantillons récoltés au Maroc, région de Taza et de cultivars australiens. Thèse de 3ième Cycle, Université des Sciences et Techniques du Languedoc, Montpellier.

Karaguzel O, Cakmackci S, Ortacesme V, Aydinoglu B. 2004. Influence of seed coat treatments on germination and early seedling growth of *Lupinus varius* L. Pakistan Journal of Botany **36**(1), 65-74.

Koné I, Lambert JE, Refisch J, Bakayoko A. 2008. Primate seed dispersal and its potential role in maintaining useful tree species in the Taï region, Côte-d'Ivoire: implications for the conservation of forest fragments. Tropical Conservation Science **1**(3), 293-306.

Ladipo DO, Boland DJ. 1995. *Pentaclethra macrophylla*: a multipurpose tree from Africa with potential for agroforestry in the tropics. NFT Highlights, NFTA 95-05, September 1995. Winlock Int., Morrilton AR, United States, p 4.

Leakey RRB. 2001. Win-Win land use strategies for Africa: 2. Capturing economic and environment benefits with multistrata agroforests. International Forestry Review **3**, 331-340.

Leakey RRB. 2004. Physiology of vegetative reproduction. In: Encyclopaedia of Forest Sciences (Eds. J. Burley, J. Evans and J.A. Youngquist). Academic Press, London, UK, 1655 – 1668.

Leakey RRB, Chapman VR, Longman KA. 1982. Physiological studies for tropical tree improvement and conservation – some factors affecting root initiation in cuttings of *Triplochiton scleroxylon* K. Schum. Forest Ecology and Management **4,** 53 – 66.

Leakey RRB, Mésen JF, Tchoundjeu Z, Longman KA, Dick JMcP, Newton A, Martin A, Grace J, Munro RC, Muthoka PN. 1990. Low technology techniques for the vegetative propagation of tropical trees. Commonwealth Forestry Review **69**(3), 247 – 257.

Leakey RRB, Newton AC. 1994. Domestication of "Cinderella" species as the start of a "woody plant revolution". In Leakey RRB and Newton AC (Eds); Tropical Trees: The Potential for Domestication and the Rebuilding of Forest Resources. HMSO, London, 3-6.

Loach K. 1988. Controling environmental conditions to improve adventitious rooting. In Davis TD, Haissig BE, Sankhla N (eds): Adventitious root formation in cuttings. Advances in Plant Sciences Series, Vol 2. Dioscorides press, Portland, Oregon, 248 – 272.

Mandeng NMO. 2009. Evaluation des potentialités de domestication de l'ebaye (*Pentaclethra macrophylla* Benth.) Par les pygmées baka de l'arrondissement de Djoum, région du Sud Cameroun. Université de Dschang, Cameroun, Niveau 5, 82 P.

Mbong ESE. 2009. Analyse socio-économique et prioritisation des espèces exotiques et locales à haute valeur socio-économique et culturelle pour les baka du grand Djoum. Rapport d'études, Partenariat ICRAF-Projet AGEFO, 45 p.

Meyer GA. 1998. Pattern of defoliation and its effect on photosynthesis on growth of Goldenrod. Functional Ecology **12**, 270 – 279.

Mialoundama F, Avana ML, Youmbi E, Mampouya PC, Tchoundjeu Z, Mbeuyo M, Galamo GR, Bell JM, Kopguep F, Tsobeng AC, Abega J. 2002. Vegetative propagation of *Dacryodes edulis* (G. Don) H. Lam by marcotting, cutting and micropropagation. Forests, Trees and Livelihoods **12** (1), 85-96.

Ngueko R. 2008. Evaluation de la première phase du système d'information des marchés des PFNL. Projet GCP/RAF/408/EC « Mobilisation et renforcement des capacités des petites et moyennes entreprises impliquées dans les filières des produits forestiers non ligneux en Afrique Centrale ». FAO-CIFOR-SNV-ICRAF-COMIFAC, 16 p.

Nkongmeneck BA, Nwaga D, Ndemmeze A, Helle F. 1996. Caractéristiques et capacitée de germination des graines de *Tetrapleura tetraptera* (Schum &Thonn.) Taub. (Mimosaceae) d'après leur situation dans la Gousse. Revue d'Ecologie (la Terre et la Vie), vol. 51, 113 – 120.

Oboh G. 2007. *Pentaclethra macrophylla*: Record from PROTA 4U. Van der Vossen, H.A.M. & Mkamilo,

G.S. (Editors). PROTA (Plant Resources of Tropical Africa / Ressources végétales de l'Afrique tropicale), Wageningen,Netherlands.

Ofori DA. 1994. Vegetative propagation of *Milicia excelsa* by leafy stem and root cuttings. PhD Thesis, University of Edinburgh, UK.

Raven PH, Evert RF, Eichhorn SE. 1992. Biologie Végétale. De Boeck 6ième edition, p 565.

Sánchez C, Vielba JM, Ferro E, Covelo G, Solé A, Abarca D, De Mier B, Díaz-Sala C. 2007. Two *SCARECROW-LIKE* genes are induced in response to exogenous auxin in rooting-competent cuttings of distantly related forest species. Tree Physiology **27**, 1459–1470.

Shiembo PN, Newton AC, Leakey RRB. 1996a. Vegetative propagation of *Gnetum africanum* Welw.: a leafy vegetable from West Africa. Journal of Horticultural Science **71**(1), 149-155.

Schiembo PN, Newton AC, Leakey RRB. 1996b. Vegetative propagation of *Irvingia gabonensis*, a West African fruit tree. Forest Ecology and Management **87**, 185-192.

Schiembo PN, Newton AC, Leakey RRB. 1997. Vegetative propagation of *Ricinodendron heudelotii*, a West African fruit tree. Journal of Tropical Forest Science **9**(4), 514 – 525.

Tchoundjeu Z. 1989. Vegetative propagation of the Tropical Hardwoods of *Khaya ivorensis* (A. Chef) and*Lovoa trichilioides* (Harm). Thesis Submitted to the University of Edinburgh for the Degree of Doctor of Philosophy, 261 p.

Tchoundjeu Z, Avana ML, Leakey RRB, Simons AJ, Asaah E, Duguma B, Bell JM. (2002) Vegetative propagation of *Prunus africana:* Effect of rooting medium, auxin concentrations and leaf area. Agroforestry Systems **54,** 183 – 192.

Teklehaimanot Z, Tomlinson H, Lemma T, Reeves K. 1996. Vegetative propagation of *Parkia biglobosa* (Jacq.) Benth., an undomesticated fruit tree from West Africa. Journal of Horticultural Science **71**(2), 205–215.

Tsobeng A, Asaah E, Tchoundjeu Z, Degrande A, Nkeumoe F, Takoutsing B, Tanly L, Ngaunkam P. 2012. Domestication of *Pentaclethra macrophylla* Benth: Phenotypic variation in tree, pod and seed traits in the humid lowlands of Cameroon. Book of abstract, AFTP international symposium hold in Yaounde, 26 – 28 November 2012.

Tsobeng A, Tchoundjeu Z, Kouodiekong L, Asaah E. 2011. Effectiveness of propagating *Diospyros crassiflora* (Hiern) using twig cuttings. Internal Journal of Biosciences **1**(4), 109 – 117.

Upadhaya K, Pandey HN, Law PS. 2007. The effect of seed mass on germination, seedling survival and growth in *Prunus jenkinsii* Hook.f. & Thoms. *Turkey* Journal of Botany **31**, 31-36.

Vivien J, Faure JJ. 1996. Fruitier sauvage d'Afrique: espèces du Cameroun. CTA et Ministère Français de la Coopération, 416 p.

Woodward AW, Bartel B. 2005. Auxin: Regulation, action, and interaction. *Ann. Bot. (Lond.)* **95**, 707–735.

World Agroforestry Centre 2006. *Allanblackia* seed germination: lecture note for the *Allanblackia* domestication workshop, 23 – 27th October 2006, p. 15.