Effect of Pawpaw Seed Extracts on the Rooting of Leafy Stem Cuttings of *Irvingia wombolu* (Vermoesen).

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ABSTRACT

The effects of three pawpaw seed ethanol extracts, namely immature, semi-mature and mature seeds on the rooting of leafy stem cuttings of *Irvingia wombolu*, obtained from coppiced shoots and inserted in a low technology non mist propagation system was investigated in the nursery of the Department of Forestry and Wildlife at the Delta State University, Asaba Campus, Nigeria. The control was dipped in ethanol only and arranged in a randomized complete block design and replicated four times. The results showed a significant effect of pawpaw seed extracts on rooting percentage (P = 0.02), with values ranging from 14.8 % to 26.7 % in the control and mature pawpaw seed extracts.. Arising from the findings of this study, it is concluded that *I. wombolu* has the potentials for regeneration by vegetative means using cheap and available pawpaw seed extracts as rooting hormones to induce rooting in a non mist propagation system.

Key words: *Irvingia wombolu*, pawpaw seed extracts, softwood cuttings, propagator

Introduction

*Irvingia wombolu* is a highly sought after multipurpose fruit tree ranked as the most important species for its food and commercial value in West and Central Africa. In spite of its importance in providing economic and livelihood benefits to subsistence farmers, it is still not widely cultivated as most of the edible fruits and kernels are still got from the wild hence and they are becoming increasingly difficult to collect due to deforestation and old tree age [32,16]. Constraints to the production of this highly sought multi-purpose tree species include long gestation period of seed sown trees, [30,17], poor germination capacity due to the recalcitrant nature of the seeds [31], variability of fruits, kernel characteristics and tree size [17,33] and limited knowledge base [35]. There is therefore the need for improvement of the species for domestication. Vegetative propagation has been shown to facilitate the process of domestication of *Irvingia* and other fruit trees by enabling the rapid multiplication of selected genotypes and the production of superior planting stock [21,22,29], and shortening fruiting time for farmers [30].

Rooting success of many species by leafy stem cuttings in terms of rooting percentage and the quality of roots produced are directly influenced by many interacting factors, both pre-severance and post-severance [29]. It has been confirmed that auxin is required for the initiation of adventitious roots on stems and indeed that the division of the first root initial cells are depended upon either applied or endogenous auxin [34,8,19,27]. According to Hartmann *et al*. [13], the formation of root premodium cells depends on the endogenous auxins in the cuttings and on a synergic compound such as diphenol. The application of auxins and some growth retardants have been used to improve the rooting capacity of cuttings in some species [37,14] identified indole-3-acetic acid (IAA) as a natural
occurring compound having considerable auxin activity. Growth regulators have marked influence on rooting several trials involving the various plant growth regulators notably IAA, IBA and NAA have been undertaken. IBA treated cuttings rooted better than those treated with NAA and IAA [36, 23, 26]. IBA originally classified as a synthetic auxin is indeed an endogenous plant compound [25, 4].

The present study aims at developing a protocol for replacing expensive synthetic rooting hormones with pawpaw seed extracts for mass clonal propagation of the species vegetative means in a non-mist propagation system.

Materials and methods

The study was conducted over a period of three years from February 2006 to March 2009 in the nursery of the Department of Forestry and Wildlife at the Delta State University, Asaba Campus, Asaba (06°14´N and 06°49´E) in Oshimili South Local Government Area of Delta State, Nigeria between.

Delta State lies in the tropical rainforest zone, an area characterized by about seven months of rainfall. Fruits of I. wombolu were procured from collectors in Ossissa, Delta State. The fruits were depulped and sun dried for three days and sown afterwards in 0.20 litter polythene pots filled with top soil. Four weeks after germination the seedlings were sown directly in the field with a spacing of 50cm x 50cm. when the seedlings attained the height of approximately one meter, four cuttings were taken 50cm. when the seedlings attained the height of approximately one meter, four cuttings were taken from the single mainstem for the experiment. The seedlings were sprayed with systemic fungicides and insecticides (Imidacloprid 10%+Metalaxyl 10%+Carbenzadim 10% WS) prior to severance.

A propagation unit was established at the Teaching and Research Farm, Delta State University, Asaba Campus for the experiment. The propagator consisted of a metal frame measuring 3.05m x 6.10m x 2.14m and enclosed in a clear polyethylene with a water-tight block work base. The base of the propagator is a modified design described by Leakey et al. [20]. was used for the study. The propagation unit was sited in a shade-house.

Pawpaw fruits at different stages of maturity namely immature, semi-mature and mature fruits were harvested from fruit trees on the campus. The flowers from the selected trees tagged and the developing fruits were harvested at intervals of 52 days (immature), 102-112 days (semi-mature) and between 160-170 days, when the fruits were ripe (mature).

The seeds were extracted and washed thoroughly in clean water and sun dried for two weeks after which they were ground separately in a sterile mortar to obtain 100g each of the dry seed powder. Ethanol extracts of the seeds were obtained by adding 100g each of the seed powder to 100ml of ethanol (wt/v) in a beaker to obtain the extracts, and left to settle for five hours at room temperature before the extracts were filtered using cheesecloth. Sterilization of the extracts was done by applying 125mg of streptopenicillín (a mixture of 62.5mg of streptomycin & 62.5mg of penicillin [12].

Three hundred and twenty single node softwood cuttings, four from each shoot were harvested from the seedlings described earlier. Eighty cuttings each were randomly assigned to the four treatments namely immature, semi mature and mature seed extracts and a control (50% alcohol). The treatments were applied by dipping the base of the cuttings in the solution for five seconds with the control dipped in alcohol only. Thereafter, the alcohol was evaporated in a gentle stream of cold air prior to insertion of the cuttings in composted sawdust inside the propagator in a randomized complete block design and replicated four times. The rooting media, consisting of composted sawdust was sprayed with a systemic fungicide (Carbendazim 12%+Mancozeb 63%W.P.) prior to insertion of cuttings. Cuttings were assessed weekly for the presence and number of roots per rooted cuttings (-2mm in length), rooting percentage, mean root length, leaf abscission, cutting mortality and shoot formation. The number of shoots formed, leaf abscission and cutting mortality was determined by direct count, while two cuttings each were uprooted weekly for the determination of rooting percentage, number of roots per rooted cuttings and mean root length.

Data collected were subjected to analysis of variance (ANOVA) and significant means were separated by Fisher’s Least Significant Difference (LSD) at 5% level of probability, using Genstat 3 Discovery edition [10]. Prior to ANOVA, all percentage data were arcsine transformed, root length data were log transformed while number of roots, leaf abscission, cutting mortality and shoot formation data were square root transformed, 11, 28.

Results and Discussion

Rooting was observed at Week 5 with no significant effect of pawpaw seed on rooting percentage (P>0.05). Rooting percentage ranges from 9.1% in both semi-mature seed extract and the control to 22% in immature seed extract. At Week 6 mature seed extract recorded significantly (P = 0.02) higher rooting percentage than the other treatments which were not different from each other. This trend continued thereafter such that by Week 7, there was significant (P<0.05) difference between the
treatments, with mature seed extract displaying higher rooting percentage than the control. Mature seed extract was however not different from immature seed extract and semi-mature seed extract which were not different from the control, (Figure 1).

There was no significant treatment effect on number of roots (P>0.05) with mature seed extract recording the highest number of roots. Semi-mature seed extract and the control were yet to root at this time. At Week 6, mature seed extract recorded significantly (P<0.05) higher number of roots than immature seed extract and the control, which are not different from semi-mature seed extract. However mature and semi-mature were not different from each other. At Week 7, there was no significant treatment effect (P>0.05) on number of roots with values ranging from 0.80 to 1.27 in the control and mature seed extract respectively, (Figure 2).

Root length was unaffected by treatment (P>0.05). A higher value in terms of root length was recorded in mature, while the least was recorded in semi-mature in Week 5 and Week 6. At Week 7 WAS, root length range between 0.06cm to 0.42cm in the control and mature respectively, (Figure 3).

There was no treatment effect (P>0.05) on leaf abscission in Week 1 to Week 3. At Week 4 the control recorded a significantly (P<0.05) higher leaf abscission than the rest treatments, between which there were no significant differences. Treatment effect tended to diminish with time, such that by Week 7 there was no significant effect of treatment (P>0.05) with the proportion of leaf loss ranging from 1.78% in both semi-mature seed extract and immature seed extract to 7.15% in the control.

Pawpaw seed extract had no significant effect on leaf abscission in Week 1 to Week 3. At Week 4 the control recorded a significantly (P<0.05) higher leaf abscission than the rest treatments, between which there were no significant differences. Treatment effect tended to diminish with time, such that by Week 7 there was no significant effect of treatment (P>0.05) with the proportion of leaf loss ranging from 1.78% in both semi-mature seed extract and immature seed extract to 7.15% in the control.

Discussion

The results from this experiment clearly established the potentials of pawpaw seed extracts in enhancing the rooting of leafy stem cuttings of I. wombo. The higher rooting percentage obtained in the cuttings treated with pawpaw seed extracts compared to the control is not unusual. A similar result of enhanced rooting was obtained by Koyejo and Omokhua [15] with coconut milk and Dolor et al., [9] with different seed extracts. Ljung et al., [24] confirmed the seedlings can synthesize IAA in leaves, cotyledons and roots. According to Ashanmugavalu, [3], extracts of cashew seeds when chemically assayed, contains natural auxin IAA. While Ljung et al., [24] and Aloni et al., [1,2] identified young shoots as the sites of auxin production. Zolman et al., [38] pointed out that these endogenous auxins, mainly IAA are stored in developing seeds as IAA conjugates and IBA for use later during germination. The pronounced effect of mature seed extract on the rooting percentage compared to immature and semi-mature seed extract may be due to elevated levels of stored auxins in the former. Chudasama and Thaker [6], in a study on auxin level in two varieties of pigeon pea observe that free IAA increased gradually, while the levels of conjugate IAA increased approximately twice with seed age in both species. This was earlier confirmed by Cohen and Bandurski [7], Bialek et al. [5] and Ljung et al. [24]. These authors asserted that IAA conjugates have been found in different plant parts but more abundant in mature seeds. Hartmann et al. [13], identified three physiological stages of seed development with varying levels of free and conjugate IAA, namely histo-differentiation (stage I), cell expansion (stage II) and maturation drying (stage III). They stated further that free IAA is high during stage I and II while conjugate forms are abundant in mature seeds and during germination.

Although no significant effect of pawpaw seed extract on mean number of roots and the length of roots produced was recorded, the treatments means recorded mimicked the trend obtained for rooting percentage with the pawpaw seed extract treatments recording higher values than the control. The reason for lack of treatment effect is not clear. The results obtained tended to suggest intuitively that increasing the concentrations of the pawpaw seed extract may have a pronounced effect.

Although no pronounced effect of pawpaw extracts on leaf abscission, cutting mortality and shoot formation was obtained, there is an indication from the trend in the results obtained that the seed extracts may have the potentials to lower leaf abscission and cutting mortality, while enhancing the number of cuttings forming new shoots.

Conclusion

The present study has made available some domestication techniques of the species, through an experiment to determine the effects of pawpaw seed extracts on the rooting of leafy stem cuttings of I. wombo.
The results suggest that cutting treatment with pawpaw seed extracts enhanced the rooting potentials of *I. wombolu* significantly. Pawpaw seed extracts could therefore replace the expensive synthetic IBA in enhancing the rooting of the stem cuttings of the species in a non-mist propagation system designed for use in rural areas, with no requirement for electricity or piped water for vegetative propagation.

**Fig. 1:** Effect of different Pawpaw seed extracts on rooting percentage of leafy stem cuttings of *Irvingia wombolu*

**Fig. 2:** Effect of different Pawpaw seed extracts number of roots of leafy stem cuttings of *Irvingia wombolu*

**Fig. 3:** Effect of different Pawpaw seed extracts on root length of leafy stem cuttings of *Irvingia wombolu*
Fig. 4: Effect of different Pawpaw seed extracts on shoot formation of leafy stem cuttings of Irvingia wombulu. Error bars indicate lsd

References


