Vegetative propagation of *Cinnamomum camphora* L. Presl by shoot cuttings: Effect of shoot physiological age

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ABSTRACT

Vegetative propagation techniques such as cuttings have been employed in propagation of plants of the family Lauraceae. Vegetative propagation using cuttings is currently the method used for reproducing camphor tree (*Cinnamomum camphora* L. Presl) at CSIR-Plant Genetic Resources Research Institute. The study was carried out to determine shoot physiological age of the camphor tree that would promote root initiation and cuttings performance. Hundred shoot cuttings of similar diameter (0.9 cm) were selected for each shoot physiological age. The first 10 cm from the apex of the shoot was considered softwood, 10 cm from the base of the softwood cuttings was considered semi-hardwood and 10 cm from the base of the semi-hardwood cuttings was considered hardwood.Shoot cuttings of the physiological ages were treated with 4000 mg l^{-1} of indole-3-butyric acid (IBA), and inserted into a mixture of river-sand and decomposed palm fibre mixed in a ratio of 1:2(v v^{-1}). Data collected after 12 weeks indicated that the physiological age of shoot had an effect on root initiation and cutting performance. Number of survived cuttings, number of rooted cuttings, root length, callus formed and sprouted cuttings differed significantly amongst shoot physiological ages. Root initiation and shoot performance were greatest in softwood cuttings. They, however, decreased with increasing physiological age of shoot.

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Introduction

Vegetative propagation has played an important role in tree improvement programmes over the years. It involves producing new plants from vegetative organs of parent plants to generate offspring with similar parental genotype (Zobel & Talbert, 1984). The method is essential for multiplying tree species of superior genotype as it bars any somatic mutation. It also ensures rapid maturity of desirable tree species for early utilisation; an advantage over seed propagation which generally takes a long time to bear fruits and set seeds, as seen in the lychee plant (*Litchi chinensis* Sonn.), which takes 10 or more years to bear fruits when seed propagated (Davie *et al*., 2011).

Davie *et al*. (2011) reported propagation by grafting, budding, layering and cuttings as some of the most common techniques in vegetative propagation. However, the use of cuttings is simple, rapid, amateur-friendly, and does not require expensive equipment (JICA, 2002).

Cuttings have been used to propagate *Lindera benzoin* L. and *Sassafras albidum* (Nutt.) Nees of the family Lauraceae of which *Cinamomum camphora* L. Presl belongs (Jenna *et al*., 2005). The method, therefore, was a viable option for reproducing *Cinamomum camphora* (Camphor tree) on the premises of CSIR-Plant Genetic Resources and Research Institute (PGRRI).

Camphor tree has been underexploited due to over reliance on artificial sources of fragrances and flavours. However, recent demand for natural sources of fragrances and flavours calls for propagation techniques aimed at increasing the population of natural sources of fragrances and flavours like camphor to meet future demand. Essential oils from *C. camphora* have been identified to be among the most important oils in world trade.

Camphor oils are extracted from the roots, branches, leaves and wood of the tree for pharmaceutical use and flavouring. The fruit is about 40 per cent oil which has industrial and medicinal uses. The wood is good for construction, ship building and cabinet-making (Li & Bai, 1982).

The study was carried out to determine shoot physiological age of camphor tree that would promote root initiation and cutting performance when propagated by shoot cuttings.

Materials and methods

Experimental site

The experiment was carried out at CSIR-PGRRI, Bunso in the Eastern Region of Ghana between September and November, 2011. Bunso lies on latitude 5°46^{I} N and longitude 1°1^{1} W with temperatures from 24 °C to about 28 °C. Average annual rainfall is 1750 mm.

Experimental design

Acompletely randomised design with three treatments and three replications was used. There were 100 shoot cuttings per treatment. The treatments were three shoot physiological ages of the camphor tree, namely softwood, semi-hardwood and hardwood cuttings.

Selection of leafy shoots

Leafy shoots that measured 30 cm in length from the apex and approximately 0.9 cm in diameter were selected from healthy branches of a matured *C. camphora* tree at premises of CSIR-PGRRI, Bunso. Each shoot was marked into three physiological ages; 10 cm from the apex was considered softwood-cuttings, 10 cm from the base of the softwood-cuttings was considered semi-hardwood cuttings and 10 cm from the base of the semi-hardwood cutting was considered hardwood cuttings (Davie *et al*., 2011). The leafy cuttings were collected from 06:00 to 08:00 h and kept cool by storing in cool boxes at 25 °C.

Preparation of rooting hormone

Indole-3-butyric acid (IBA) at a concentration of 4000 mg l^{-1} was used as rooting hormone of the different treatments. IBA concentration of 4000 mg l^{-1} was prepared by weighing 400 mg of powdered stock IBA. The weighed IBA powder was then poured into a volumetric flask containing 100 ml of 50 per cent ethanol and stirred continuously till it completely dissolved.

Preparation of rooting medium

River sand and composited oil palm fibres in proportions of 1:2 (v v^{-1}) were mixed thoroughly on a thick sheet of polythene. Stones and other foreign materials were removed. The mixture was sterilised by autoclaving at 121 °C and 15 psi for 15 min. It was then dished into 40 cm wide, 15 cm deep perforated plastic bowls. A transparent polythene sheet was mounted on the bowl to serve as a humid chamber.

Insertion of cuttings in rooting medium

The bases of shoot cuttings were cut flat with a pair of secateurs to avoid one-sided rooting. Cuttings were immersed in a fungicide solution (Mancozeb-0.75g l^{-1}) for 15 min and air-dried for 30 min. The bases of the cuttings were then dipped in freshly prepared 4000 mg l^{-1} IBA for 10 s (Reintenet, Gertze & Arends, 2002). Cuttings were inserted into the rooting medium which had been allowed to cool after sterilisation to a depth of 2 cm, and watered till the medium was moist and then placed in a plant house of 40 per cent light intensity. Watering was done regularly for 12 weeks.

Data collection and analyses

Data were collected on the shoot cuttings after 12 weeks of insertion into the rooting medium. Data collected were number of survived cuttings, number of rooted cuttings, root length, sprouted cuttings and callused cuttings. Root length was measured with a metric measuring rule. Shoot cuttings that had green leaves and stem at 12 weeks after insertion were considered alive. Cuttings that were alive were counted for the three shoot physiological ages. Shoot cuttings that had developed leaf buds or leaves after the 12 weeks of insertion were considered to have sprouted. Sprouted cuttings were counted for each shoot physiological age.

Data collected were analysed using Microsoft Excel Analysis of Variance (ANOVA). The Duncan's multiple range test was used to determine differences between means at least significant difference (LSD) of 5 per cent.

Results and discussion

Per cent survival

Fifty-four per cent of softwood cuttings set survived after 12 weeks compared to 40 per cent of semi hardwood cuttings and 13 per cent of hardwood cuttings. There were significant difference between softwood cuttings and hardwood cuttings. However, survived softwood cuttings did not differ significantly from semi hardwood cuttings (Table1). Survival and rooting performance of the cuttings may have been inhibited with the cuttings for this research being harvested in late September. Cervenyet, Gibson & Barrett, (2006) working in Australia observed that physiological age of shoot had no effect on survival of cuttings of *Tecoma stans* L. Juss and *Murraya paniculata* L. Jack; rather, the season of collection affected cutting survival and rooting performance, with cuttings harvested in June performing better than cuttings harvested in September.

Per cent sprouting

Softwood cuttings recorded the highest sprouting of 24 per cent. Semi-hardwood cuttings had 10 per cent sprouting. Hardwood cuttings did not sprout. Differences between sprouting of softwood and semi-hardwood cuttings were significant (Table 1). Agbo & Obi (2008) working on *Gongronema latifolia* Benth, however, reported a lower number of shoots for softwood cuttings and a higher number of shoots for hardwood cuttings

TABLE 1

Effect of Shoot Physiological Age on Survival and Sprouting of Cuttings of Camphor Tree

Shoot physiological age	*Per cent survival*	*Per cent sprout*
Softwood	54a	24a
Semi-hardwood	40a	10b
Hardwood	13b	0c

Note: Per cent sprout with the same letter are not significantly different at (P = 0.05) according to Duncan's multiple range test.

after 8 weeks, implying that the hardwood cuttings developed more shoots probably due to their expected higher carbohydrate reserves. Significantly higher rooting percentages and root lengths may have resulted in increased sprouting due to easy translocation of water and mineral nutrients to the above ground parts of the cuttings leading to rapid growth and multiplication of shoots. Vigorous growth within younger materials is a more likely cause. Meristematic activity within softwood cuttings leading to the formation of new shoot may be fast and vigorous, while cells within the tissues of older cuttings (semi-hardwood and hardwood cuttings) may be completely differentiated and, thus, have little meristematic activity as supported by Davie *et al.* (2011).

Per cent callus and root formation

Callus formed in 20 per cent of semi-hardwood cuttings and 10 per cent of softwood cuttings. Hardwood cuttings formed no callus. Callus formation showed significant difference between semi-hardwood and hardwood. Physiological age of cuttings, therefore, had effect on callus formation. Differences in the rooting of cuttings were significant amongst physiological ages. Rooting in softwood cuttings was highest (Table 2).

The higher rooting percentages and root lengths for softwood cuttings could be attributed to higher concentrations of endogenous root promoting substances present in soft cuttings. This agrees with Elnour *et al.* (1990) and Elkhalifa (1990), whose studies on vegetative propagation of tropical trees had softwood cuttings recording the highest rooting percentages. Cerveny *et al.* (2006), however, reported that physiological age of cuttings had no effect on rooting performance of *Tecoma stans* and *Murraya paniculata* cuttings. Auxin levels decrease with increasing distance from the apex of the shoot (Prusinkiewicz, *et al*., 2009). Palanisamy &

TABLE 2

Effect of Shoot Physiological Age on Per cent Callus and Rooting of Cuttings of Camphor Tree

Shoot physiological age	*Per cent callus*	*Per cent rooting*	*Mean no of roots*	*Mean root length (cm)*
Softwood	10ab	20.0a	12.1a	8.4a
Semi-hardwood	20a	3.3b	3.7b	2.2b
Hardwood	0b	3.3b	1.0c	1.0b

Note: Figures with same letter are not significantly different at P = 0.05 according to Duncan's multiple range test.

Kumar (1997) reported that cuttings nearer the apex of *Azadirachta indica* L. Adelb (Neem) branches rooted better than those from the middle or the proximal end, implying that endogenous auxin levels could indeed be greatest at the distal end, decreasing towards the proximal end.

Conclusion

It is concluded from the study that the physiological age of shoot had an effect on root initiation of *Cinnamomum camphora* shoot cuttings. Softwood cuttings recorded greatest rooting performance. Rooting performance drastically decreased with increasing physiological age of shoot.

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