

progenies FCRI 22 (357.48 g), FCRI HC 21 (328.07 g), FCRI HC 10 (325.01 g), FCRI HC 18 (305.43 g), FCRI HC 12 (255 g), FCRI HC 20 (252.26 g) and FCRI HC 27 (250 g) recorded maximum seed yield at 9 months after planting (Figure 4 and Table 5). These hybrid clones also expressed superiority in terms of early flowering and fruiting coupled with early yield, which thus lends scope for further promotion and utilization of *Jatropha* as a successful biofuel crop. The existing local seed sources of *Jatropha* are beset with problems of variation in seed yield, poor seed and oil yield and susceptibility to pest and diseases. The variable hybrid progenies developed so far and the hybrid progeny in the pipeline will help to solve the issue of seed yield and oil content.

All the identified hybrid mother plants exhibited distinct morphological features coupled with higher seed yield (700 g to 1.4 kg/plant) at the third year after establishment and oil content (17.95 to 55.26%). Except few hybrid clones, the others exhibited oil content more than 25%. The fruiting behaviour of some clones was unique, which produced fruits of different size, shape and colour (Figure 3). Five hybrid clones, viz. FCRI HC 2, 11, 21, 32 and 33, exhibited distinct variations in terms of oblong and coloured fruit coats. The hybrid clone 21 expressed oblong character coupled with continuous fruiting type from the base to top of the plant. In each branch, two to three bunches of fruits were seen from the base to top of the plants. In each bunch, a minimum of 15 fruits were observed. Three hybrid clones (FCRI HC 20, 21 and 22) recorded an average yield of 1.4 kg/plant (mother plant) on single plant basis at the third year after establishment. This yield was more than 300% than the local *Jatropha* seed sources yield, which was 200 to 300 g/plant at the same age and hence the hybrid clone proved to be promising. The superiority of the individual hybrid clone is now raised on a plantation scale which also expressed early superiority in terms of yield at 9 months after planting, thus lending scope for the promotion of a *Jatropha*-based biofuel plantation programme.

Systematic testing trials are already established and all the hybrid clones expressed early flowering and fruiting within 3 months after planting. Within 5 months, three hybrid clones, viz. FCRI HC20, 21 and 22 recorded excellent growth, including fruiting characteristics and seed yield. Such yield improvement in *Jatropha* through hybrid development is currently not available for utilization. Hence the present study is an attempt which will provide a scope to all user agencies. Further studies on testing of *Jatropha* hybrid genetic resources at multilocations are underway to screen and promote potential *Jatropha* high-yielders.

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Development of efficient techniques for clonal multiplication of *Jatropha curcas* L., a potential biodiesel plant

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Effect of auxins (IAA, IBA and NAA) and vitamin B₁ (thiamine) on rooting response of branch cuttings and air-layers of *Jatropha curcas* during spring and monsoon seasons was studied. Spring season was found best for clonal multiplication of genetically superior material in *Jatropha*. Cuttings treated with 600 and 800 mg l⁻¹ thiamine showed 100% sprouting during both seasons. The average sprout growth was also found maximum in thiamine treated cuttings. Auxins enhanced rooting of cuttings during spring season, but showed poor performance or even failed to root during monsoon. Interestingly, thiamine triggered highest

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rooting during monsoon and was comparable during spring. Average per cent rooting was also recorded maximum in air-layers treated with thiamine (75, 150 and 300 mg l⁻¹) in comparison to auxins during both seasons. However, the number of roots per layer increased with increasing concentration of NAA and IBA in spring, but decreased in the monsoon season. The cleft grafting was found more promising in terms of success and growth of scion in the spring season. This technique can be practically applied on commercial scale in the areas where *J. gossypifolia* grows as a weed.

Keywords: Air-layering, cutting, grafting, jatropha, vegetative propagation.

OVER hundred plant species are known to produce fatty oils. Among the oil-bearing trees, *Jatropha curcas* L. (family Euphorbiaceae), also known as 'ratanjot' in India, appears to be a potential source of biodiesel, which has evoked worldwide interest in recent years¹. In spite of some doubts regarding this species, it appears to hold promise for future as a potential biodiesel plant. At present, considerably good improved strains of jatropha are available at different centres in our country for large-scale planting. However, the required quantity of genetically pure seed material for commercial plantings is still not available. A lot of genetic variation exists in the available seed material², which obviously necessitates vegetative reproduction for the multiplication of desired strains. Therefore, the present study was undertaken to develop the appropriate protocols for mass production through clonal propagation (stem-cutting, air-layering and budding/grafting) in order to maintain genetic purity, uniformity and gainful exploitation of useful variation, and also to meet the required demand for high-quality planting material at commercial scale.

The present study was carried out at the nursery of the Forestry Department, CCS Haryana Agricultural University, Hisar (20°10'N, 75°46'E, and 215 m), India. Elite progenies of candidate plus plants (CPPs) of *J. curcas* containing more than 40% oil (seed basis) formed the basic material. These were selected after analysis of oil content and fatty-acid profile of 398 lines of *J. curcas* collected from natural and planted populations from diverse eco-geographical regions of India. Three clonal propagation methods, viz. rooting of branch cuttings, air-layering and grafting were applied. Healthy and uniform cuttings (20–25 cm long and 2–3 cm thick) of *J. curcas* were taken from the middle portion of one-yr-old branches of three-yr-old plants during mid-February (spring) and end of July (monsoon) 2006. Cuttings were dipped in 0.1% bavistin fungicide for 2–3 min and subsequently washed in distilled water before giving hormonal treatment. These were divided into 29 groups of 30 cuttings each. Group 1 was treated with distilled water as control. Groups 2–29 were treated with 50, 100, 200, 400, 800,

1000 and 2000 mg l⁻¹ of indole-3-acetic acid (IAA), indole-3-butyric acid (IBA), naphthaleneacetic acid (NAA) and thiamine (vitamin B₁). Cuttings were treated by submersing about 3 cm basal portion in each treatment solution for 16 h. Cuttings were then transferred into polythene bags (9" × 6" size) containing coarse sand (0.2–2.0 mm particle size; pH 7.0) as rooting medium. Only one cutting was planted in each bag and the 30 bags under each treatment were divided into three blocks, each block containing ten cuttings. The bags were then kept in nursery beds and irrigated regularly depending on weather conditions and sand moisture status. Observations on sprouting, rooting and root characters were recorded after 90 days of planting.

For air-layering, one-yr-old healthy branches with 1.5–2.0 cm girth were taken on the selected mother plants. Girdles of 3.0–4.0 cm length were made around using a budding knife at 30 cm from the branch tip, without injuring the underlying wood. The layers were made during spring and monsoon seasons of 2007 and treated with the same growth regulators as used in the first experiment on rooting of branch cutting, but the concentrations of these plant growth regulators were modified in the light of the results obtained in the above experiment. Under each treatment, five girdles were made. The growth regulators (IAA, IBA, NAA and thiamine) with different concentration levels, viz. 75, 150, 300, 600, 900, 1200, 1500 mg l⁻¹ and distilled water (control) were applied to the exposed surface with sterilized cotton. A handful of moistened sphagnum moss was placed around the cut surface and wrapped by a polythene sheet so that the sphagnum moss completely covered the cut surface and it was tied at both ends with a thread in order to avoid moisture loss. The polythene sheet was wrapped by an aluminum foil. Regular observations at an interval of 7 days from the date of treatment, were recorded. The air-layers were detached from the mother plants when roots were visible from the polythene covers and a cut was made just below the lower end of the ringed portion with the help of a sharp secateur. After detachment, the polythene cover was removed gently under tree shade without injuring or disturbing the roots. Observations on per cent rooting, number of roots/layer and root length in all the air-layers were recorded. The rooted air-layers were then transplanted in pots filled with sand, soil and farmyard manure in equal proportion and hardened in the mist chamber for a week followed by gradual hardening under partial shade for 15 days.

Further, different grafting techniques, viz. cleft, side veneer, whip and tongue, and T-budding were attempted both during the spring and monsoon seasons to graft the early and high-yielding genotypes of *J. curcas* on *Jatropha gossypifolia* (hardy and resistant to root rot). For grafting, 5–6-month-old plants (50–85) having of 3–4 cm stem diameter were selected as stock. The scion of the same thickness was collected from the branch apex.

Aftercare of grafted plants was taken using customary practices. Data were analysed for analysis of variance and test of significance was tested employing statistical model³.

Except thiamine, in general, per cent sprouting, sprout growth, rooting and number of roots were found higher in spring season compared to monsoon season (Figure 1). Such seasonal variation has been reported in other tropical forest tree species as well^{4,5}. The cuttings showed poor performance or even failed to root in spite of auxin treatment during monsoon. This could be due to higher meristematic growth and consequently more endogenous auxins level during the monsoon season, and thus applied auxins raising the concentration to supra-optimal levels that were inhibitory. On the other hand, in the spring season exogenously applied auxins enhanced rooting by raising it to an optimal level. In the present study, among different growth regulators, thiamine induced maximum sprouting during both seasons. Cuttings treated with 600 and 800 mg l⁻¹ thiamine showed 100% sprouting in both seasons. The effects of vitamins on different living beings are well known for a long time, but their similar favourable effects on plants have not fully been established. The mechanism of promotion of rooting due to vitamin application is not clear. Nevertheless, vitamins might serve as

a co-factor of auxins in root formation. IAA too was found equally responsive in sprouting of cuttings. However, 800 mg l⁻¹ and higher concentrations of NAA treatment inhibited sprouting during both seasons. The average sprout growth was also found maximum in thiamine-treated cuttings during both seasons. It appears that possibly thiamine acts as a mobilizing factor in one way or other to synthesize rhizocaline complex in light-exposed leaves, transported basipetally to promote adventitious rhizogenesis as auxin synergist⁶. Sprouting alone is not the right and dependable parameter for the final success/rooting of cuttings. Cutting sprouts with the support of reserve food material. Therefore, for the success of cuttings, rooting is all the more necessary. Untreated cuttings rooted fairly well in spring, and the application of auxins and vitamin further triggered to enhance rooting significantly. The better formation of roots due to auxin treatment may be due to accumulation of metabolites at the site of application, synthesis of new protein, callus formation, cell division and cell enlargement⁷. Cuttings treated with 100, 200 and 400 mg l⁻¹ concentrations of IBA, IAA and NAA respectively, showed 100% rooting in spring season, but poor response or even failed to root during monsoon (Figure 1). Interestingly, thiamine triggered the highest rooting during monsoon and was comparable to auxins during spring season (Figure 2a). Also, thiamine is approximately five times cheaper than generally used synthetic auxins (IBA and IAA) in national and international markets. Seasonal variation in rooting behaviour of branch cuttings could be due to differences in the activities of hydrolysing enzymes causing seasonal fluctuations in the availability of sugars, which are the principal source of metabolic energy required for cell division and differentiation during root initiation in cuttings⁸. The maximum number of roots (9.3) in monsoon and 63.6 roots per cutting in spring season were observed in the cuttings treated with 50 and 1000 mg l⁻¹ NAA respectively. Results are indicative of varied stimulus of hormone application with the nature of hormone in different

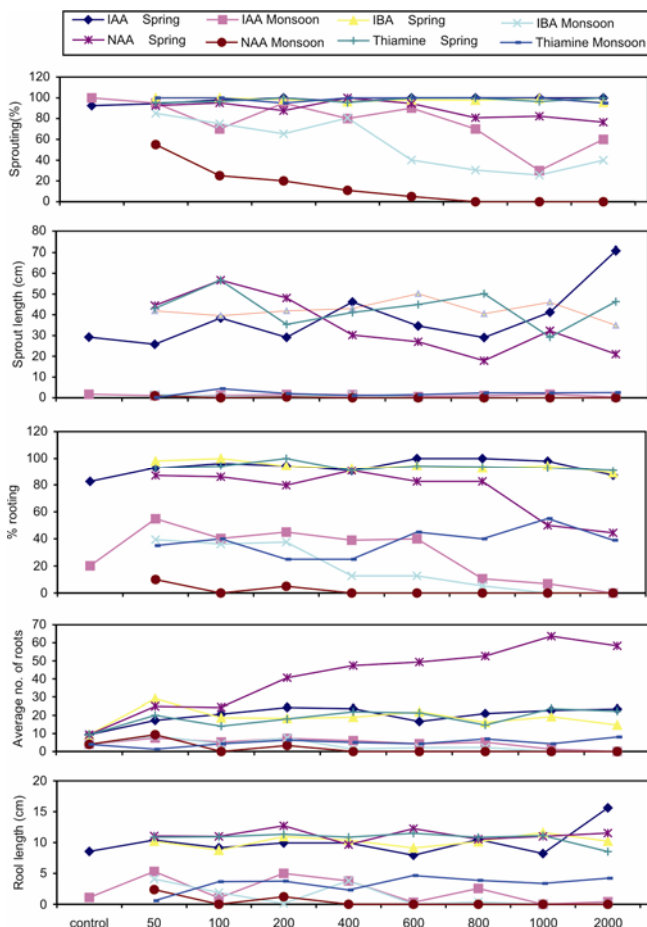


Figure 1. Effect of different growth regulators on sprouting and rooting of branch cuttings in *Jatropha curcas*.



Figure 2. Clonal multiplication techniques in *J. curcas*. a, Rooting of cuttings; b, Air-layering, and c, grafting.

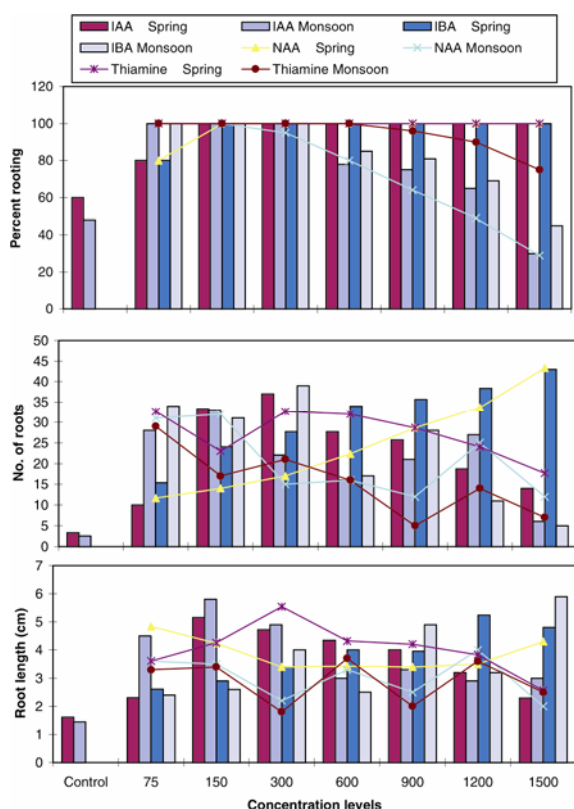


Figure 3. Effect of different growth regulators on rooting of air-layers in *J. curcas*.

seasons. The seasonal variation in the rooting response of stem cuttings has been reported earlier by other workers^{4,5,9}. Such variation could be due to varying levels of endogenous root-forming substances^{7,10} caused by fluctuations in climatic factors such as temperature, light and humidity¹¹.

Regarding rooting of air-layers, except 75 mg l⁻¹ IAA, NAA and 150 mg l⁻¹ NAA treatments, 100% rooting was recorded in all the treatments during spring season (Figure 3). Profuse rooting in air-layered shoots has been reported with higher concentration of auxins in other tree-borne oilseed species¹². During monsoon season, however, per cent rooting decreased with increasing concentration of growth hormones. During both seasons, the average rooting percentage recorded was maximum in air-layers treated with thiamine (Figure 2 b) in comparison to auxins. The number of roots increased with the increasing concentration of NAA and IBA in spring, whereas a reverse trend was observed in monsoon season. The number of roots per layer was found comparatively less during monsoon season and the number of roots decreased with increase in concentration of different growth regulators. Maximum number of roots (43.3) developed in the branch layers treated with 1500 mg l⁻¹ NAA, which was closely followed by similar concentration of IBA (43.0) during spring. However, higher concentrations of IAA

adversely affected the air-layers in promoting root growth. No definite trend was noticed in thiamine. Among different concentrations of thiamine, the highest number of roots (32.6) was recorded in 75 and 300 mg l⁻¹, and lowest (17.7) in 1500 mg l⁻¹ treatments. The maximum number of roots with an average length of 5.54 cm was observed in layers treated with 300 mg l⁻¹ thiamine and minimum (1.6 cm) in untreated ones in spring. Interestingly, the lower concentrations (75 and 150 mg l⁻¹) of NAA, IAA and thiamine were effective in increasing root length. But, IBA at higher concentrations (1200 and 1500 mg l⁻¹) was found more effective. Similar trend was recorded in air-layers during monsoon season.

Cleft method of grafting was found more successful during both seasons for triumphant union of *J. curcas* as scion and *J. gossypifolia* as rootstock in terms of initial sprouting of the scion at two weeks after grafting (Figure 2 c). Such findings were reported in *Madhuca latifolia*, *Azadirachta indica* and *Prosopis* species¹³⁻¹⁵. It was found that cleft grafting was more strong and stable than other grafting methods. Data show that cleft grafting exhibited 83.5 and 89.6% success in sprouting of scion of *J. curcas* on *J. gossypifolia* in the spring and rainy seasons respectively. Side veneer technique was observed second best in spring, which gave 71.2% success. However, in the rainy season whip and tongue method performed second best in successful grafting of both species. T-budding failed totally in the sprouting of scions. The grafted plants, obtained from different techniques in spring, showed moderate to high flowering and fruiting after about one and half months of grafting. The shortening of time lag, easy accessibility of genotypes both in time and space, and early flowering induction are the main character features of the grafting¹⁶. However, the growth of the grafted plants obtained in monsoon was quite slow and failed to produce flowers.

It could be concluded from the present study that for large-scale production of genetically pure and improved planting material of *J. curcas*, vegetative reproduction through stem cuttings treated with 600 and 800 mg l⁻¹ thiamine for 16 h and air-layers treated with 75, 150, 300 mg l⁻¹ thiamine was the most effective and economic method in spring and monsoon seasons, since thiamine is cheaper than the auxins. Secondly, cleft method of grafting was found to be more thriving during both seasons for successful union of *J. curcas* as scion and *J. gossypifolia* as rootstock. The grafted plants obtained from different techniques in spring, showed more vegetative growth and moderate-to-high flowering and fruiting.

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Anemophily, anemochory, seed predation and seedling ecology of *Shorea tumbergaia* Roxb. (Dipterocarpaceae), an endemic and globally endangered red-listed semi-evergreen tree species

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Shorea tumbergaia is an endemic and globally endangered, red-listed, semi-evergreen tree species, restricted to the southern Eastern Ghats in Andhra Pradesh and Tamil Nadu. The flowering is ephemeral and also not an annual event. Massive blooming, drooping inflorescence with pendulous flowers, ample pollen production, gradual pollen release as a function of anther appendage, aerodynamic size of the pollen grains with reticulate exine and muri separated by lumina and strong protogyny – all contribute to anemophily. The plant is self-compatible but there appears to be abortion of fruits from selfed-flowers. The fruits are large, winged and anemochorous. The fruits are attacked by an unidentified bruchid beetle prior to dispersal. In healthy fruits, the seed has no dormancy and it germinates as soon as it falls from the tree. The study reveals that non-annual, massive flowering, short flowering period, partial flowering at tree level, seed predation, short-distance seed dispersal, absence of seed dormancy, low rate of seedling establishment and inability of seedlings to compete with other plants collectively contributed to the occurrence of a small population of *S. tumbergaia* in a restricted area of the Eastern Ghats forests and interplay of all these factors might have led to the ‘endangered’ status of this species.

Keywords: Anemochory, anemophily, protogyny, seed predation and seedling ecology, *Shorea tumbergaia*.

THE genus *Shorea* (family Dipterocarpaceae) is native to Southeast Asia, from northern India to Malaysia, Indonesia and the Philippines^{1,2}. It is a tropical genus with 196 species of mainly rainforest trees, out of which 148 species are currently listed in the IUCN Red List; majority of them are listed as critically endangered. Many species are economically important timber trees. Janardhanan³ documented that *Shorea* species are found on the borderline between the moist fertile evergreen forests and the less moist and dry deciduous forests in India. The *Shorea* species found in India are *S. assamica*, *S. robusta*, *S. roxburghii* and *S. tumbergaia*. The last species is an endemic and globally endangered semi-evergreen tree species,

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