# Propagation Techniques in Coconut

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Coconut (Cocos nucifera L.) is a major crop in tropical areas, providing cash and subsistence to smallholders. As every part of the tree can be made into universally used products, it is popularly known as "the tree of life". Though India was the first country in the world to evolve a commercial hybrid of coconut (Patel, 1937) and the country has since released many high yielding varieties and hybrids, current production of quality planting materials meets only about 25 percent of the annual requirement of planting material needed for area expansion and replanting. Predominantly cross-fertilized nature of coconut results in enormous variability in the seedling progenies, leading to dearth of quality planting materials. This article describes the major advances in propagation techniques of coconut palm, conventionally, by seeds and through plant tissue culture techniques.

## I.a. Seed propagation

Seed propagation is the only viable method of producing planting materials in coconut palms. Coconut is long lived and has a very long juvenile phase. The performance of coconut palm can be judged only after 10-15 years of planting. The long life span and large capital outlay involved in establishing a coconut plantation necessitates the use of quality planting materials as a first step for the successful cultivation of the crop. If poor planting materials are used for planting, the new plantation can prove to be uneconomic, causing considerable loss of time and money to the grower. Coconut being a perennial crop, poor selection continues as a source of loss throughout its life period. Through a series of selections made at different stages, it is possible to obtain quality seed nuts and seedlings. Hence, for production of quality planting material of coconut,

1.Seed propagation -WCT mother palm

2. Seed propagation-Seednut

3. Seed propagation
-seedling selection

4. Hybridization- Dwarf parental palm

6 Hybridization Dx T hybrid nut

5. Hybridization Tall parental palm

7 Hybridization Dx T hybrid

a three tier selection approach is adopted starting from mother palm selection, seed nut selection and seedling selection.

In India, about 30 varieties have been developed and released for commercial cultivation through mass selection by ICAR-CPCRI, State Agricultural Universities and coordinating centres under All India Coordinated Research Project on Palms. These released varieties are propagated by identifying typical mother palms of the respective variety and collecting open pollinated nuts from the selected mother palms. Seedling selection is done based on the morphological characters like vigour and petiole colour confirming to their mother palms.

## Important features of superior mother palms:

• Palms should be regular bearers and should yield 80 or more nuts/year under rainfed conditions and 100-120 nuts/year under irrigated condition.

• Palms should have typical characteristics of the variety with regard to palm traits, crown, leaf and nuts

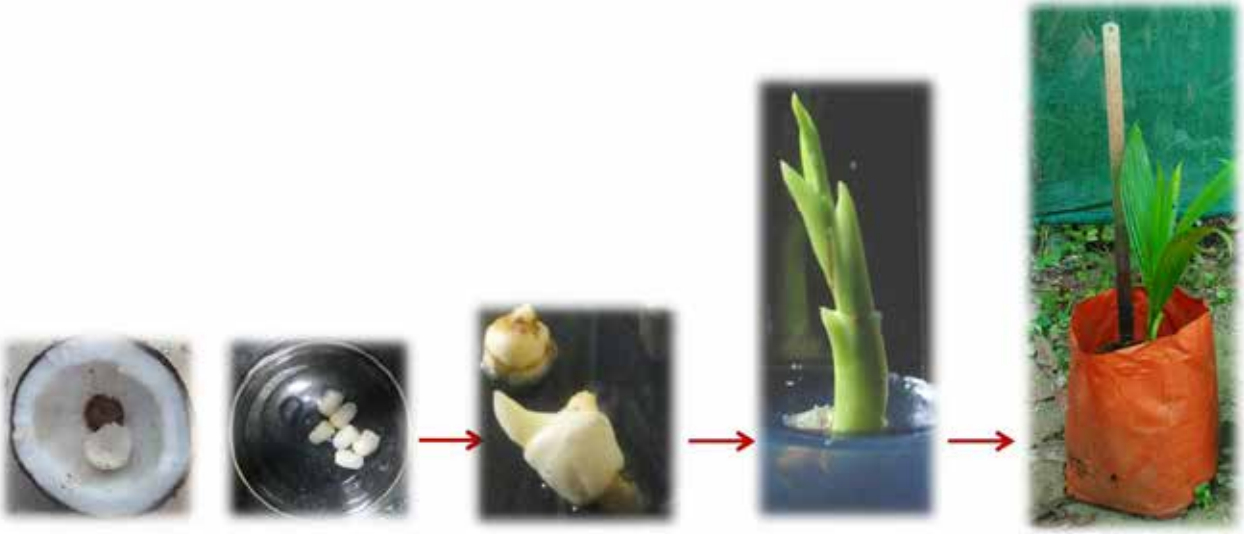
• Palms should have short and stout inflorescence stalk with bunches, preferably resting on the leaf petioles of the lower whorl.

• Palms with more than 30 leaves and 12 inflorescence carried evenly on the crown

• Palms should be more than 22 years of age

• Palms should be free from all diseases and pests.

For producing planting material for the root (wilt) disease prevalent tract, select healthy and high yielding palm located in the midst of root (wilt) affected palms. The disease-free status of mother palms should be confirmed by serological testing. The age of West Coast Tall (WCT) mother palms should be more than 35 years and should be surrounded by palms of which atleast 80% are affected by root (wilt) disease.




## Seed nut selection

Seed nuts can be collected from selected palms when nuts attain full maturity. In Talls it takes usually 11-12 months for maturity, whereas in Dwarfs, they mature in 10-11 months. Nuts should not be damaged while harvesting. Discard nuts having irregular shape and size. Harvested nuts can be stored in shade to prevent drying up of nut water. Harvested seed nuts should be stored in shade to prevent drying of nut water, till their husks become completely dry. Seed nuts of tall variety can be stored upto two months after harvest. The seed nuts of dwarfs should be sown within 15 days of harvest.

## Seedling selection

Selection of seedlings is as important as the selection of parental palms and seed nuts. In the case of WCT, ungerminated nuts, multiple sprouts, thin/etiolated, bent/spindled and albino seedlings can be removed from 150 days from the date of sowing and in dwarfs culling can be taken from 120 days onwards. If rigorous standards of selection are adopted, 60 to 65 % quality seedling can be obtained from the total nuts sown. An ideal one-year-old coconut seedling has the following characters:

1) Seedlings should be healthy, vigorous and robust- looking, with large numbers of leaves, good girth at the base, short, thick leaf stalks and large number of roots.

2) Early germinated nuts give better seedlings than the late germinated ones and are associated with early bearing

3) Early splitting of leaves into leaflets is a good sign of vigour

4) From the one-year-old nursery, select vigorous seedlings having minimum of six leaves and girth of 10 cm at the collar.

Seedlings of dwarf varieties can be easily identified by their early germination, short height, short and sturdy leaves with short and narrow leaflets and early splitting of leaves. Different dwarf varieties are easily recognized by their colour of petiole.

## I. b. Hybridization

Among the several breeding methods, exploitation of heterosis has the maximum impact on improvement of cross-pollinated crops. Since the desired characters such as high yield, precocity in bearing, better quality, high copra and oil content, drought tolerance and disease resistance are distributed among different varieties or different individuals of the same variety, hybridisation is the most useful method to bring together the desirable traits. Harland advocated the exploitation of hybrid vigour to increase the productivity of coconut. A new dimension to coconut improvement was added with the discovery that the hybrids made by Patel (1937) between Tall and Dwarf cultivars showed enormous vigour, enhanced production potential and early bearing tendency. The reciprocal combination of Dwarf X Tall showed an even higher productivity, indicating strong possibility of cytoplasmic influence of the dwarf parent. During 1970-1990 production of T X D hybrids was common throughout India. However, efforts to evaluate D X T hybrids started simultaneously and the first D X T (Chandra Sankara) was released by ICAR-CPCRI during 1985. Nowadays, D X T is more common due to the ease with which it can be produced compared to T X D hybrids.


## Hybridisation technique

### ▶ *a. Emasculation*

The first step in hybridization is the removal of male flowers from the inflorescence of the female parent to avoid self pollination. This is called emasculation. To avoid chances of pollen contamination it is better to do the emasculation in the initial few days after the opening of the inflorescence. This is done either by removing the individual male flowers by hand or by cutting the spikelet (with knife/secature) about 4 to 5 cm away from the upper most female flower and removing the remaining male flowers by hand. Generally, 1 to 2 male flowers are found attached to the base of the female flowers and care should be taken to ensure that these male flowers are also removed at the time of emasculation. In WCT mother palms, emasculation is done by cutting the spikelets 5 cm above the female flowers within 10-14 days of opening of the inflorescence. When dwarf varieties are used as female parent, emasculation should be carried out within 5-6 days of the opening of the inflorescence.

### ▶ *b. Bagging*

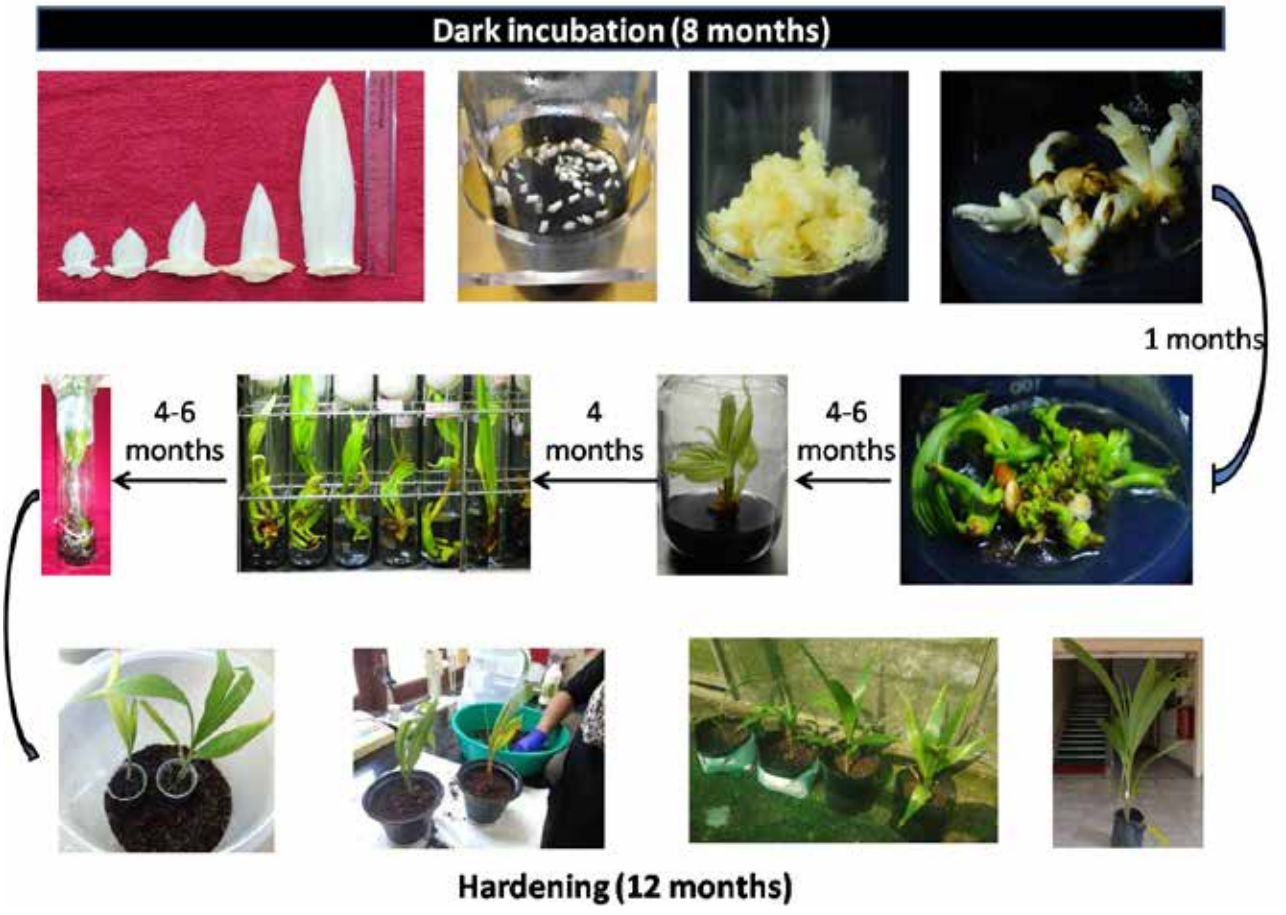
A few days before the female flowers become receptive; the inflorescence is covered and tied firmly with the pollination bag. In WCT, the 'buttons' or female flowers become receptive approximately 19-21 days after opening of the inflorescence and pollination is carried out when female flowers become receptive. Receptivity of a single female flower will last for one to two days. In dwarf varieties, the female flowers become receptive 8-16 days after bunch opening (depending on the variety). In Chowghat Green Dwarf the female flowers become receptive within 6-8 days and continue up to 14-18 days whereas in Malayan Green Dwarf the receptive period is from 16-23 days after bunch opening.

### ▶ *c. Collection of male flowers and processing of pollen*

To carry out artificial pollination in coconut it is necessary to collect and store the pollen to carry out pollination as and when the female flowers become receptive. Maturity of the anthers is indicated by the bluish green tinge at the tip. These mature male flowers should be kept, in between two sheets of thick paper and pressed gently, using wooden ruler and dried by keeping at 38-40$^{o}$C in an incubator, for 24 hours. By sieving the dried male flowers, we get the yellow coloured dust of pollen grains. Pollen may be preserved in polypropylene vials, in desiccators for about 10-12 days without losing its viability.

### ▶ *d. Pollination*

The pollen grains and chalk-powder should be mixed in 1: 9 ratio and filled in the applicator, just before use. The receptivity of the female flowers (buttons) is marked by the honey like exudates from the ivory coloured stigma, which can be seen through the transparent plastic window of the pollination bag. At this stage, a small hole should be made in the plastic sheet. Insert the tube of the applicator through this hole and press the rubber bulb to spray the pollen-chalk mixture inside the bag. Care should be




taken to close the hole after spraying the pollen-chalk mixture, using adhesive tape. All female flowers do not attain receptivity on the same day, so the above process should be continued till all the buttons in the inflorescence become receptive. It should be kept in mind that the pollen from the same parent should be used for individual inflorescence and since the honey dries progressively during the morning hours, 7-11 am is the most preferred time for conducting artificial pollination.

### ▶ *e. Bag removal and labeling*

On completion of fertilization the stigma turns brown and the secretion of honey stops. Three days after the last pollination (on the last female flower to attain receptivity), the pollination bag should be removed and the bunch should be labelled properly, to retrieve the details of the cross later at harvest. As usual, 10 to 12 months after pollination, mature nuts can be harvested.

## Identification of hybrid seedlings

Hybrids usually express hybrid vigour in the nursery for vegetative characters such as height, girth at collar and number of leaves in the seedlings. The colour of the petiole and vigour of the seedlings can be used as a selection criterion for hybrid seedlings in the nursery. Hybrid usually exhibit hybrid vigour at the seedling stage itself and petiole colour of the hybrid seedlings may range from green/ brown/ intermediate shades of the parents.

In India, nearly 20 coconut hybrids have been developed and released by ICAR- Central Plantation Crops Research Institute and State Agricultural Universities.

For production of hybrids, artificial pollination need to be carried out every time using selected mother palms using the pollen collected from concerned male parent. That means the hybrid palm should not be used as mother palm and the progeny obtained from a hybrid palm should never be used for planting. Because of the segregation in subsequent

generation, the chance of obtaining hybrid vigour is very less in such seedlings.

## ▶ *II. APPLICATION OF TISSUE CULTURE TECHNIQUES IN COCONUT PROPAGATION*

Mass multiplication of elite coconut palms, with high yield and resistance to biotic and abiotic stresses, is the need of the hour for obvious reasons. Unfortunately the progress achieved in clonal propagation in coconut has been rather sluggish. The recalcitrant nature of coconut is the main impediment for development of a commercial scale protocol for in vitro multiplication. Selection of explants is the key element for its successful outcome. Numerous tissues viz., leaves, inflorescence, plumular tissues, ovaries, anthers, roots and zygotic embryos have been utilized as explants for coconut tissue culture. Major advances in the application of plant tissue culture techniques in coconut propagation are coconut embryo culture, plumule culture and immature inflorescence culture.

### *II. a. EMBRYO CULTURE*

The success of in vitro germination of coconut zygotic embryos provides an alternative way of transportation of coconut germplasm in the form of embryo cultures. This method also avoids the formalities of quarantine regulations, which include treatments of the nuts with insecticide, fungicide and fumigation. Further, embryo collection also considerably reduces the transportation cost as 500 seed nuts weighing 600-700 kg can be transported in one briefcase containing 500 embryos weighing 5-6 kg (including weight of briefcase). Standardization of embryo culture protocol in coconut can benefit in embryo rescue, collection and exchange of coconut germplasm and in vitro screening for biotic and abiotic stress.

The protocol for embryo culture developed at ICAR-CPCRI (Karun et al., 1999) has successfully been used in germplasm expeditions since early 2000's. Another important application of embryo culture is in the germplasm collection and exchange. Collecting and exchange of coconut germplasm is difficult and not economic because of short dormancy and bulkiness of the seed resulting in seed germination when stored for more time in a germplasm expedition. Moreover phytosanitary restrictions too severely limit the germplasm introduction. Standardization of embryo culture technique provides an easy and safe alternative for the movement of coconut germplasm and is emphasized in the technical guidelines of FAO/IPGRI (Diekmann, 1997).

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### Embryo rescue

Embryo rescue is an effective technique for obtaining plantlets in vitro from embryos which either fail to germinate in nature or exhibit delayed germination. Mohachao Narel is a coconut variant reported from Guhaghar taluk of Ratnagiri District of Maharashtra, which is characterized by sweet and soft kernel and has less fibre content (Samsudeen et al., 2013). The weight of embryo obtained from sweet endosperm nuts was significantly lower than nuts possessing normal endosperm. This may be the reason which hinders its germination under natural conditions. To overcome this problem, embryos from sweet kernelled nuts were 'rescued' via embryo rescue and plantlets could be regenerated successfully through the embryo culture protocol standardized by ICAR-CPCRI.

### *II. b. PLUMULE CULTURE*

Coconut plumular culture protocol (Karun et al., 2008) could be applied for rapid multiplication of dwarf palms and also for in vitro conservation of genotypes. Besides increasing the multiplicative rate of coconut to many folds, plumule culture will be handy while initiating transformation studies as the culturing period is considerably less when compared with other explants. Further when a genotype is cryopreserved in the form of embryos, its regeneration in large numbers could be possible by means of plumule culture.

### Extraction of explants

- Scoop out zygotic embryo with a portion of endosperm using a cork borer from dehusked and split opened 10 months old mature coconuts.
- Extract the embryo from the endosperm with the help of scalpel or blade
- The extracted embryos are surface sterilized with 20 % sodium hypochlorite for 20 minutes followed by 4-5 washes with sterile distilled water

## Inoculation

Inoculate surface sterilized embryos into Y3 medium containing 3% sucrose and 1 g/litre charcoal and agar 5.5 g/litre. The embryos are inoculated in test tube containing 20 ml solidified media and incubated in dark condition. After one month of incubation, slice out the plumular region of embryo with the help of a sharp scalpel. Each plumular region can give about 4-5 slices. Inoculate these explants into same basal medium supplemented with 2,4-D (16.5 mg/L) with TDZ (1mg/L) and incubate in dark for callus induction. Sub culture the explants to same basal media supplemented with 2,4-D , BA and TDZ. Gradually reduce 2,4-D concentration from 8.25 mg/L to 4mg/L, then to 2 mg/L at each monthly and finally media free of 2,4-D.

## Plantlet regeneration

Observe somatic embryogenesis with an incubation period of 16 weeks. Transfer germinated embryos with two leaves and primary leaves to liquid rooting media. Developing plantlets should be subcultured atleast once every 4-5 weeks. Select plantlets with well developed root and root system for hardening.

### ▶ *II. c. IMMATURE INFLORESCENCE CULTURE*

Immature inflorescence contains numerous meristematic points and therefore is considered a potential source of explant to clonally propagate important crop plants. The success depends on the selection of inflorescence of correct maturity stage. The technique can be used to propagate adult coconut palms, whose performance has been established (e.g. productivity or resistance to diseases). The advantage of rachillae explants from immature inflorescence is that it enables multiplication of adult bearing palms of known performance for production of true-to-type planting materials which otherwise is not possible in a cross pollinated crop like coconut. Hence, this method can be used as clonal propagation technique for tall palms. The technique involves inoculation of rachllae bits of 1 mm size in Y3 media supplemented with 2,4-D and incubation in dark for a period of eight months. The white shoot like outgrowths were transferred to ½ MS media containing NAA and BAP and incubated in 16 hours light conditions. The multiple shoots developed were separated and cultured in shoot regeneration medium containing Y3 media fortified with NAA and BAP. The plantlets with 3-4 leaves were transferred to rooting medium and after developing sufficient roots, the plantlets were transferred to potting mixture containing cocopeat: vermiculite for hardening. Even though ICAR-CPCRI has succeeded in plantlet regeneration from immature inflorescence explants, lot of refinement is further required for developing a commercial scale protocol. Standardization of the regeneration protocol from immature inflorescence will lead to mass production of elite, high yielding and disease-resistant planting materials. ■

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**Obituary**

Veteran agriculture expert and former Director of Agriculture, Government of Kerala, R Hali (87) passed away on 13th December 2020. He was a member of the Kerala Agricultural Policy Committee, one of the pioneers of farm journalism in Kerala and was a frequent contributor of articles for Indian Coconut Journal and Indian Nalikera Journal.

CDB places on record deepest condolences on his demise.