



Novel Propagation of Fruit Species Through Mini-Cuttings and Leaves

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Abstract: The main objective of the study was to standardize mini-cutting and leaf propagation in selected four fruit species locally available around College of Forestry, Sirsi. Mini-cuttings and leaves of 4 species namely *Annona muricata*, *Annona squamosa*, *Garcinia morella* and *Morinda citrifolia* were taken and planted in zip lock covers with coir pith as the rooting media inside the polyhouse. Three treatments were control, 1000 ppm of IBA and 1000 ppm of Coumarin were given. The experiment was conducted in two seasons *i.e.*, spring and rainy season. Survival per cent of all the four species was seen higher in the rainy season than the spring season. Treatment with 1000 ppm of IBA and coumarin was effective in root length (and) or number of roots in either of the seasons in both propagation methods. Three species selected for the study adapted well to mini-cutting technology which shows that it is possible to propagate genotypes with superior characteristics through mini-cutting with a greater number of seedlings per source. However, complete propagation of plant was not possible through leaves.

Keywords: Clonal technology, Genotypes, Mini-cutting, Propagation

Propagation, either by sexual or asexual means, is one of the most crucial steps for the success of any tree improvement programme. Vegetative propagation plays a key role in large-scale multiplication of superior clones or tested plus trees (Palanisamy and Subramanian 2001, Prabakaran et al 2017). Most adopted methods of vegetative propagation includes cutting, grafting, budding, layering, micro-propagation and root sections/suckers (Kumar et al 2018, Thakur et al 2021). Recently, mini-cutting technology is gaining importance among commercial forestry companies to mass propagate clones of *Eucalyptus* (Almeida et al 2007) and is being expanded to other areas such as floriculture and fruit crops. A mini-cutting is essentially a miniature stem cutting and its length varies from two to six centimetres, depending on the size of the shoots emitted, the size of the leaves, and the phyllotaxis of the species. It is seen as an improvement of stem cutting, displaying variations for optimizing both the quality of clonal saplings and rooting (Xavier and Silva 2010). It allows a breeder to multiply any individual tree at an extremely rapid pace (up to 1,00,000 plants per year from a single individual), which is far superior when compared to the normal macro stem cutting methods. Mini-cutting in the production of forest species was first carried out with *Eucalyptus* in the 1990s (Brondani et al 2010). Today the technique has been expanded to propagate forest species such as Pine (Alcantara et al 2007), Pink cedar (Xavier et al 2003), and Ipe (Oliveira et al 2015) and in Casuarina (Palanisamy et al 2020).

Vegetative propagation through leaves is traditionally practiced in succulents such as Cactus, Bryophyllum, Agave, and *Aloe-vera*. Leaf cuttings are prepared by taking a single leaf from the plant, which itself is the propagating material (Welch-Keesey and Lerner 2002). The main advantage of this leaf propagation is that, with this technique plants can be raised throughout the year, with minimal disturbance to the mother plant unlike the stem cutting method (Uday et al 2014). However, vegetative propagation through leaves is not very commonly practiced and strangely there is a lack of interest among researchers in standardizing these techniques in forestry species. Despite having a huge potential to mass propagate clones through mini-cuttings and through leaves, there is a paucity of fine-tuning the techniques in important fruit species. Hence, the main objective of the study was to assess the potentiality of propagation through mini-cuttings and leaves in four fruit species as well as to understand the influence of seasons on success rate.

MATERIAL AND METHODS

The present study was carried out in a poly-house at the College of Forestry, Sirsi, Uttara Kannada district which comes under the hill zone (Zone 9) of Karnataka State, in the Central Western Ghats of India which lies between 14° 26' N latitude, 74° 50' E longitude and at an altitude of 619 m MSL.

Mini-cuttings of 5-7 cm in length and fully expanded middle aged leaves were collected from saplings of *Annona*

muricata, *Annona squamosa*, *Garcinia morella* and *Morinda citrifolia* growing in the College of Forestry, Sirsi campus. The experiment was conducted in two seasons of 2022 i.e., spring (Feb-Mar) and rainy (Jul-Aug). The leaves and mini-cuttings from either apical or intermediary shoots were placed in zip lock covers with coir pith as the rooting media after treatment. Three treatments viz., T₁= Control, T₂= Indole-3-butyric acid (1000 ppm in talc form), T₃= Coumarin (quick dip method in 1000 ppm solution) were imposed to leaves and mini-cuttings. Every treatment consisted of three replications with 15 mini-cuttings/leaves each which was set in Completely Randomized Design (CRD). After 8th week of planting leaves and mini-cuttings, the following observations were taken. The per cent leaves and mini-cuttings survived were estimated using formula: Per cent leaves and mini-cuttings survived=(Number of green leaves /mini-cuttings÷Total number of leaves)×100. Similarly, the per cent leaves and mini-cuttings rooted are worked out by the following formula: Per cent leaves and mini-cuttings rooted=(Number of leaves / mini-cuttings rooted÷ Total number of leaves / mini-cuttings planted)×100. Total number of roots produced and root length of each leaf/mini-cutting, were counted after the 8th week of planting.

RESULTS AND DISCUSSION

Mini-cuttings of four species remained green in both spring and rainy season by the end of 8th week, but *Annona muricata* and *A. squamosa* leaves showed drying in spring season. Survival per cent of all four species was good during rainy season in both mini-cutting and leaf propagation (Fig. 1). *Morinda citrifolia* is the only species which showed rooting through leaves in both the seasons (Plate 1). Three species such as *A. muricata*, *A. squamosa* and *M. citrifolia* showed

rooting through mini-cuttings (Plate 2). There was no rooting seen in the month of spring in *Annona muricata* mini-cuttings. Callus formation was noticed in *Garcinia morella* leaves in the month of rainy season. Promising rooting was observed in *M. citrifolia* mini-cuttings and leaves when compared to other species (Fig. 2). However, three species (*A. muricata*,

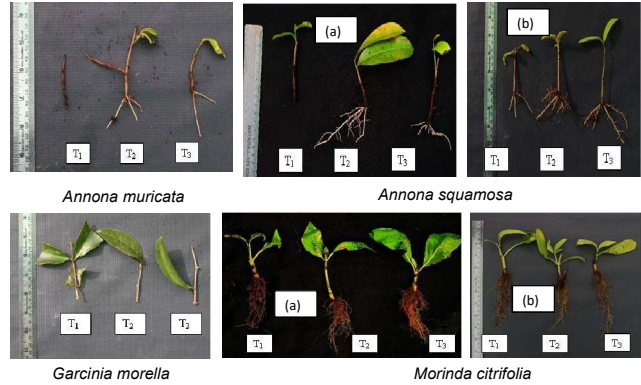


Plate 2. Influence of different treatments with plant growth regulators on rooting through mini-cutting propagation (a-spring season, b-rainy season)

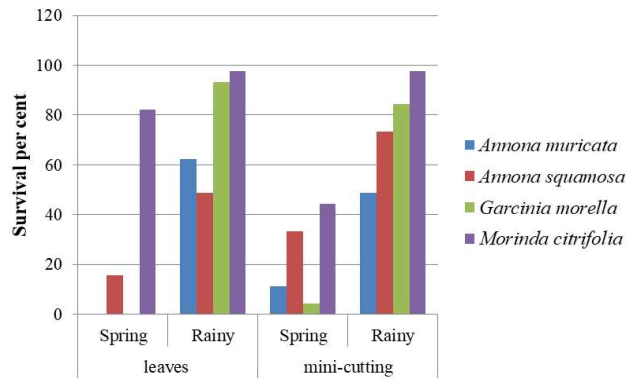


Fig. 1. Survival per cent of leaves and mini-cuttings of four fruit species in two seasons by the end of 8th week

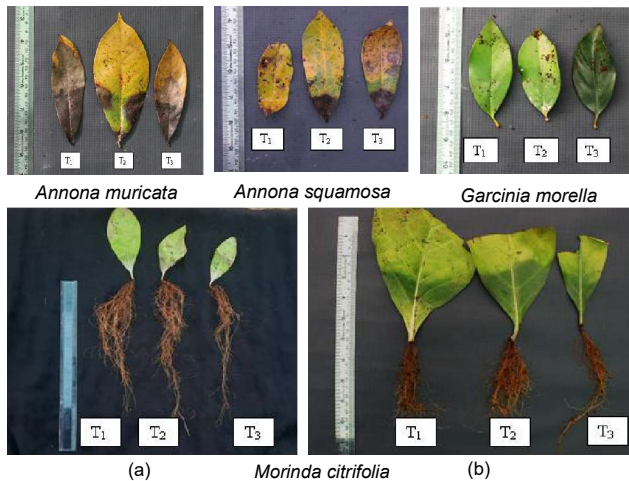


Plate 1. Influence of different treatments with plant growth regulators on rooting through leaf propagation (a-spring season, b-rainy season)

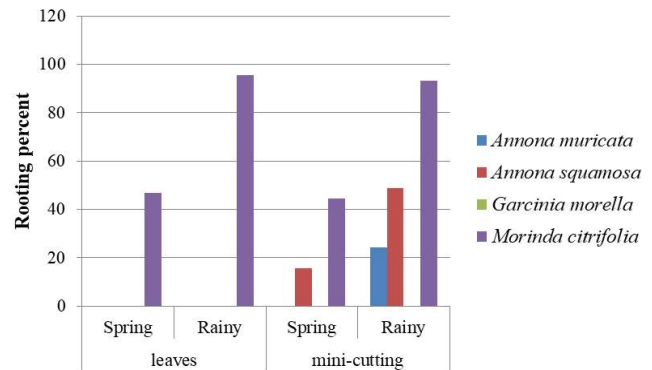


Fig. 2. Rooting per cent of leaves and mini-cuttings of four fruit species in two seasons by the end of 8th week

A. squamosa and *M. citrifolia*) showed successful propagation through mini-cutting and one species *i.e.* *M. citrifolia* through leaf propagation, whereas one species *G. morella* showed callus formation in leaf propagation. The survival per cent of all the four species was highest during rainy season than in spring season through both leaf and mini-cutting propagation perhaps due to congenial environment. Similar results have been reported by Palanisamy et al (2020) where in *Casuarina* clones showed highest rooting (86%) in rainy season compared to spring

(82%). Perhaps cuttings can hold more moisture which may help them to survive and to root vigorously in rainy season because of high humidity than any other months.

Treatment with IBA (250 mg/l) resulted in better results in terms of, number of roots (70.63), rooting per cent (80%), root length (11.13 cm) and number of leaves (5.25) per rooted mini-cuttings in *Azadirachta indica* (Gehlot et al 2014). Similar findings were found in this study where 1000 ppm of IBA was significantly effective in root length or number of roots in three species (Table 1, 2 and 3). Highest root length

Table 1. Influence of different treatments and seasons on mean number of roots and mean length of roots in *A. muricata* adopting mini-cutting propagation (The values are Mean \pm SD)

Treatment	Mean number of roots		Mean root length (cm)	
	Spring	Rainy	Spring	Rainy
Control (T ₁)	0	1.17 \pm 2.02	0	0.33 \pm 0.58
IBA 1000 ppm (T ₂)	0	3.50 \pm 0.50	0	3.33 \pm 0.35
Coumarin 1000 ppm (T ₃)	0	2.83 \pm 0.29	0	3.77 \pm 0.31
CD @ 1 %	0	1.80	0	0.87
P value	0	<0.01	0	<0.01

Table 2. Influence of different treatments and seasons on mean number of roots and mean length of roots in *A. squamosa* adopting mini-cutting propagation (the values are Mean \pm SD)

Treatment	Mean number of roots		Mean root length (cm)	
	Spring	Rainy	Spring	Rainy
Control (T ₁)	0	2.17 \pm 1.89	0	2.67 \pm 2.32
IBA 1000 ppm (T ₂)	4.67 \pm 0.58	3.93 \pm 3.48	8.67 \pm 0.32	4.23 \pm 3.67
Coumarin 1000 ppm (T ₃)	7.83 \pm 1.04	3.78 \pm 0.20	4.40 \pm 0.30	6.60 \pm 0.20
CD @ 1 %	1.40	N/S	0.52	N/S
P value	<0.01	0.60	<0.01	0.20

Table 3. Influence of different treatments and seasons on mean number of roots and mean length of roots in *M. citrifolia* adopting leaf propagation and mini-cutting propagation (The values are Mean \pm SD)

Treatment	Mean number of roots		Mean root length (cm)	
	Spring	Rainy	Spring	Rainy
a. Leaf propagation				
Control (T ₁)	5.42 \pm 0.52	2.63 \pm 0.35	27.77 \pm 0.21	14.50 \pm 0.20
IBA 1000 ppm (T ₂)	4.08 \pm 0.14	2.97 \pm 0.35	31.83 \pm 1.26	15.13 \pm 0.15
Coumarin 1000 ppm (T ₃)	3.40 \pm 0.35	1.80 \pm 0.20	30.97 \pm 0.45	23.60 \pm 0.10
CD @ 1 %	0.75	0.80	1.59	0.32
P value	<0.01	<0.01	<0.01	<0.01
b. Mini-cutting propagation				
Control (T ₁)	2.87 \pm 0.23	13.60 \pm 0.74	6.83 \pm 0.21	11.03 \pm 0.15
IBA 1000 ppm (T ₂)	4.87 \pm 0.23	7.08 \pm 0.25	6.33 \pm 0.15	11.47 \pm 0.45
Coumarin 1000 ppm (T ₃)	6.17 \pm 1.04	3.53 \pm 0.31	6.77 \pm 0.25	19.53 \pm 0.76
CD @ 1 %	1.28	0.98	N/S	1.05
P value	<0.01	<0.01	0.05	<0.01

(31.83 cm) was observed in *M. citrifolia* through leaf propagation with IBA treatment. Highest root length in mini-cutting propagation was seen in *M. citrifolia* (19.53 cm) with treatment of 1000 ppm of coumarin. Lipecki and Selwa (1977) recorded coumarin was generally more effective than other chemicals used to observe rooting in softwood cuttings of *Prunus mahaleb* L. clones. Highest mean number of roots in leaf propagation and mini-cutting propagation was observed in *M. citrifolia* i.e. 5.42 and 13.60, respectively with control as the treatment. Rana et al (2017) reported that bamboo cuttings without any hormonal treatment exhibited 83.33% rooting, which was significantly higher (34%) than the cuttings treated with 100 ppm of IBA.

CONCLUSION

The current study, perhaps for the first time, unveiled the potentiality of propagation of economically important fruit species through mini-cutting and leaves. Except *G. morella* all the three species showed rooting in mini-cutting in either of the season or both the seasons. *M. citrifolia* showed rooting through leaves in both spring and rainy season. *G. morella* showed callus formation through leaf propagation in rainy season. Three species selected for the study adapted well to mini-cutting technology. This shows that it is possible to propagate genotypes with superior characteristics through mini-cutting with a greater number of seedlings per source. However, in leaf propagation only rooting is achieved, which is the first and very crucial step in achieving complete regeneration, none of the rooted leaves showed signs of shoot formation during the study period. Perhaps more time is required to develop the shoots from the leaves. If leaf propagation method is perfected to achieve shooting, then it can be used for raising endangered species and it would be wonderful method to overcome tissue culture.

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