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## Viable Options and Factors in Consideration for Low Cost Vegetative Propagation of Tropical Trees

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**Abstract:** Vegetative propagation techniques are increasingly being applied to the domestication of tropical tree species as a means of producing planting stock and capturing genetic variation. However, the level of knowledge of cost-effective for vegetative propagation is not common and has slowed down domestication many of tree species. The current scenario of rapid climate changes and increasing land degradation in many tropical countries, certain multi-purpose tree species are becoming rare and threatened with local extinction. Thus, knowledge and efforts to develop low-cost vegetative propagation technologies to the people are necessary. In this study viable alternatives and potential factors to simplify various operations and reduce the costs in vegetative propagation for both micropropagation and macropropagation are reviewed. Viable alternatives and potential factors should be opted to lower the cost of vegetative propagation without compromising the quality of the propagules and plants.

**Key words:** Vegetative propagation, micropropagation, macropropagation, low cost

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### INTRODUCTION

Vegetative propagation is asexual propagation which involves removing sections of stem, leaf or root tissue from the parent or donor plant, treating this tissue with plant growth regulators and then inducing adventitious root or shoot formation under controlled environmental conditions. Vegetative propagation is an irreplaceable tool for tree domestication and breeding with many advantages which include rapid multiplication of a species under threat, offers alternative means for the domestication of trees because of their (1) relatively long generation times, (2) irregularity in flowering and fruiting, (3) predominantly outbreeding nature, with most traits being of low heritability and (4) assembly of germ plasma for base populations to maintain certain desired characteristics (Leakey and Simons, 2000; Hartmann *et al.*, 2002). Other advantages of vegetative propagation is to generate disease-free planting material (Hartmann *et al.*, 2002; Tchoundjeu *et al.*, 2004). Vegetative propagation also offers a unique opportunity of avoiding the problem of poor seed viability and recalcitrant seeds predominant in tropical tree species.

Vegetative propagation techniques are increasingly being applied to the domestication of tropical tree species (Leakey *et al.*, 1990). Although efforts have traditionally focused on economically important tropical tree species such as medicinal plants (Tchoundjeu *et al.*, 2004; Teklehaimanot *et al.*, 2004; Mng'omba *et al.*, 2008), with a current scenario of rapid climate changes (IPCC, 2008)

and increasing land degradation in many tropical countries, certain multi-purpose tree species are becoming rare and threatened with local extinction. These species frequently provide a combination of the following: food, medicine, fuel, fodder, wood and income through sale of products. The level of knowledge of cost-effective vegetative propagation of tropical and sub-tropical tree species is not common and has slowed down domestication of many trees (Akinnifesi *et al.*, 2006). The low cost vegetative propagation is a solution for domestication and conservation of indigenous and threatened tree species.

#### **The need for low cost vegetative propagation technology:**

Low-cost vegetative propagation technology is the adoption of practices and use of inputs and equipment to reduce the unit cost of propagule and plant production. Potential factors for low cost vegetative propagation discussed in this study are those which should be opted to lower the cost of production without compromising the quality of the propagules and plants. The broad application of existing vegetative propagation technologies is important for tree improvement and the most relevant aspects for programs involving indigenous species and impoverished communities (Leakey, 2005). However, the high costs in the technologies may be an obstacle to the direct use of propagules as planting stock. The review describes the potential factors in consideration for low cost vegetative propagation of tropical trees.

**Major types of vegetative propagation methods and costs comparison:**

A range of approaches can be utilized for vegetative propagation but the major types are macropropagation which involves vegetative propagation by cuttings and micropropagation through *in vitro* culture to multiply ontogenetically mature shoots that are very difficult to propagate by cuttings. Micropropagation offers the high multiplication rates of propagules and much quicker to capture of genetic gains in forest tree breeding programs than macropropagation through stem cuttings (Minghe and Ritchie, 1999). Micropropagation is also appropriate for tree improvements, to free tree from the known viral diseases or when there is a need for a wider cultivation (Mng'omba *et al.*, 2008). However, the current high costs for micropropagation in terms of the supply of culture containers, media, chemicals, equipment and instruments has caused a major impediments to the direct use of micropropagation in many programs (FAO, 2000). Even the relative cost of labour and expertise is very high in micropropagation compared to macropropagation by stem cuttings which is generally more cost effective (Abbasi *et al.*, 2007). In addition, for macropropagation the time required for plant regeneration is generally very short, often just days or weeks. Although, macro propagation by stem cuttings has low cost compared to micropropagation, in both methods there are potential factors to be considered in order to lower propagation costs.

**LOW COSTS IN MICROPROPAGATION**

In tropical tree species, the practical applications of micropropagation seem to concentrate on tissue culture techniques. A few woody trees have been propagated using tissue culture *Maytenus senegalensis* (Matu *et al.*, 2006) and *Pappea capensis* (Mng'omba *et al.*, 2007). Tissue culture-based plant propagation is carried out in highly sophisticated facilities which are expensive and often are not available in the developing countries of tropical Africa. For example, the cost media, chemicals, equipment and instruments used in micropropagation such as autoclaves for sterilization of media and instruments are often very expensive. Hence, options to expensive inputs and infrastructure are described and can be sought and developed to reduce the costs in micropropagation.

**Low cost option for culture media:** A conventional plant culture medium usually contains a basal solution with major and minor mineral elements, a source of carbon (that is, sucrose), vitamins, growth regulators, a gelling agent for semi-solid media and water (Trigiano and Gray, 2000).

In a conventional medium, if agar is used as a gelling agent, it can represent up to 70% of the total cost, followed by the minerals, water, sucrose and other minor media components (Prakash *et al.*, 2004). As a substitute for agar, cheaper alternative sources of gelling agent such as starches and plant gums can be used (Kaur *et al.*, 2005). Table sugar and molasses are suitable and cheap alternative to pure carbohydrates (Prakash *et al.*, 2004). Molasses is not only a source of carbohydrates but vitamins as well, especially inositol, niacin, pyridoxine, pantothenic acid, thiamin, riboflavin, biotin, choline and minerals (Santana *et al.*, 2009). Many laboratories have reported the use of table sugar (sucrose) on *in vitro* plant propagation medium (Kodym and Zapata-Arias, 2001; Prakash *et al.*, 2004; Kaur *et al.*, 2005; Tyagi *et al.*, 2007). Plant cultures can be maintained in rooms with air conditioners and tube lights instead of highly priced plant growth chambers. The conventional method of downward illumination can be replaced by sidewise lighting systems, which not only reduce the number of lights but also provide more uniform illumination to the cultures (Nagamori and Kobayashi, 2001; Savangikar *et al.*, 2002).

**Low cost option for washing and sterilising operations:**

Costly dishwashing machines for cleaning the culture containers can be replaced by manual washing if labour is relatively inexpensive. The washed culture containers can be dried in sun instead of costly hot air ovens (Prakash *et al.*, 2004). In small-scale laboratories, the autoclaves can be replaced by large sized pressure cookers, which are much cheaper (FAO, 2000). Instead of having one or two huge horizontal autoclaves, which generate hot air pockets in the sterilising rooms, it is better to have more of smaller vertical autoclaves, which keep the air cool. The normal practice to use costly aluminium foil for wrapping instruments for sterilisation can be replaced by stainless steel containers, which are autoclavable (Escobar *et al.*, 2005). Glass bead sterilizers can be used to sterilize forceps and scalpels instead of the conventional flame sterilization using spirit lamps or gas cylinders. Water is one of the major medium components. Distilled water obtained by electrical distillation is expensive and required a sophisticated distillation apparatus. However, rain water or tap water can be used as a cheap low cost substitute as it has previously reported by Kaur *et al.* (2005) and Tyagi *et al.* (2007).

**Low cost option for containers:** Proper choice of containers can reduce the cost in micropropagation. The expensive imported vessels can be replacement with reusable glass jars and lids which can reduce costs of production (Prakash *et al.*, 2004). Disposable plastic bags

eliminate the cost of washing and of lids; the system of using plastic bags has been reported to reduce labour cost per propagule by 60% (Savangikar *et al.*, 2002). Glass bottles and baby-food jars with polypropylene caps are the most widely used containers and the most economic and low cost option compared to conical flasks which are more expensive than glass bottles (Kodym and Zapata-Arias, 2001). The production cost of tissue-cultured plants can be reduced by 50-90% by using low cost containers (Abbasi *et al.*, 2007).

### LOW COSTS IN MACRO PROPAGATION

**Low cost option for propagation system:** There are many different types of propagation systems for stem cuttings in macropropagation but the most common are (1) intermittent mist, controlled by a range of different sensors and (2) air-tight, water-tight, high humidity, non-mist propagators (Hartmann *et al.*, 1997). The basic principles behind all these systems are that the cuttings are well supplied with water at the cutting base while the leaves are in a cool, shady environment with low vapour pressure deficit to minimise water stress. Intermittent mist and non-mist propagators are all very effective, but vary in cost of materials be used, sophistication and training needed. Mist propagation system involves a lot of cost in terms of materials needed for construction of the units and requires skilled personnel (Hartmann *et al.*, 2002). Mist units in intermittent mist propagation system usually are programmed to run continually or intermittently to ensure cuttings do not dry and wilt which also involves the cost of piped water and electricity.

The non-mist propagators (Leakey *et al.*, 1990) have the advantage of being very low cost in terms of materials needed for its construction and so are highly suitable for use in developing country or rural situations, where electricity and piped water are not available. Non-mist plant propagator can be constructed from cheap, locally available material in tropical areas (Mbile *et al.*, 2004). The system is made of a wooden frame enclosed in a single sheet of polythene such that the base is completely water tight and the lid is airtight. The system is covered by successive layers of small stones and gravel which may be covered by the sand as rooting media. A low-technology non-mist propagator has been developed that is extremely effective with low cost and meets the needs of most tree improvement projects in both the moist and dry tropics (Leakey *et al.*, 1990). Non-mist propagators have successful rooted cuttings of *Irvingia gabonensis*, *Dacryodes edules* (Tchoundjeu *et al.*, 2006), *Dalbergia melanoxylon* (Amri *et al.*, 2010) and several other tropical tree species.

**Low cost option for the choice of age of donor plant:** Effect of age of donor plant on rooting ability of stem cuttings has been reported in many trees (Stenvall *et al.*, 2004; Bhardwaj and Mishra, 2005; Husen and Pal, 2007). The ease of adventitious root formation tends to decline with the physiological age of the stock plants, due to several progressive changes in morphological, anatomical and biochemical traits such as decreased sensitivity of aging tissues to rooting promoters and/or accumulation of inhibitory substances which inhibit rooting (Hartmann *et al.*, 2002; Husen and Pal, 2006). Stem cuttings from mature donor plants have decreased content of endogenous auxins (Leakey, 2004; Husen and Pal, 2007), which requires application of higher quantity of exogenous auxins as rooting hormone than stem cuttings from juvenile donor plant as a results the overall costs of rooting hormones in mature cuttings may be higher than juvenile cuttings. In general, the more lignified the vegetative material, the higher the hormone doses should be applied (Hartmann *et al.*, 2002; Amri, 2009).

Significant interactive effect of age of donor and auxin such as indole-3-butyric acid (IBA) has been reported in many tree species such as *Robinia pseudoacacia* and *Grewia optiva* (Swamy *et al.*, 2002), *Khaya anthotheca* (Opuni-Frimpong *et al.*, 2008) and *Dalbergia melanoxylon* (Amri *et al.*, 2010) where percent rooting for IBA treated cutting from juvenile donor plant had higher rooting percentage than IBA treated cuttings from mature donor plant. Cuttings from mature trees have a low rate of propagative success and the number of people with the appropriate skills to carry it out may be a constraint to its widespread application in the future (Simons and Leakey, 2004). However, propagation by juvenile leafy cuttings is very easy probably over 90% of tropical trees are amenable to propagation by juvenile stem cuttings (Leakey *et al.*, 1990) and currently is the preferred option for participatory domestication in village nurseries (Amri, 2002; Mbile *et al.*, 2004; Mialoundama *et al.*, 2002).

### Low cost option for the type of stem cuttings to be used:

Stem cuttings of tropical trees can come in many forms, but the two major groups are leafy softwood cuttings from relatively unligified, young shoots and leafless hardwood cuttings from older and more lignified shoots which typically have already shed their leaves due to the onset of dry season. It is difficult and expensive to root leafless hardwood cuttings compared to leafy softwood cuttings in terms of labour and quantity of rooting hormones to be used (Amri, 2009). Hardness of cuttings determines the quantity of rooting hormones to apply. Generally, leafy softwood cuttings require 0.05-0.3%,

while semi-hardwood cuttings require 0.1- 0.5% of rooting hormone. Leafless hardwood cuttings require 0.25-1.0% or higher concentration of rooting hormone (Hartmann *et al.*, 1997; Amri, 2002; Amri and Friedrichs, 2006). More than 80% of tropical forest trees so far tested can be rooted as leafy stem cuttings in low-technology poly-propagators and/or under mist (Mudge and Brennan, 1999). By selecting for those characteristics of stem form and branching habit that show up early in life and making sure that cuttings come from a suitable shoot may lower the cost of propagules production.

**Low cost option for hormone (auxins) treatment:**

Typically cuttings treated with auxins root more rapidly and produce more roots and usually with a higher percentage than untreated cuttings. Indole-3-acetic acid (IAA), indole-3-butyric acid (IBA) and 1-naphthalene acetic acid (NAA) are three synthetic rooting chemicals that have been found to be reliable in the promotion of rooting in cuttings (Hartmann *et al.*, 2002). IBA is the most effective root promoting auxin and widely applied in general use because it is non-toxic to most wood plants over a wide range (Tchoundjeu *et al.*, 2004). IBA has an important role in the development of adventitious roots, increasing rooting percentage, improving quality of roots and uniformity in rooting of cuttings (Negash, 2002; Tchoundjeu *et al.*, 2002; Teklehaimanot *et al.*, 2004; Husen and Pal, 2007; Amri, 2009). Root formation process in cuttings is intensified by IBA which influences polysaccharide hydrolysis resulting in increased content of physiologically active sugar needed to provide energy for meristematic tissues and later for root primordia and roots formed as reported in *Tectona grandis* (Husen and Pal, 2007), *Swartzia madagascariensis* (Amri and Friedrichs, 2006) and *Dalbergia melanoxylon* (Amri *et al.*, 2010).

IBA is applied to plant cuttings for rooting using powder or liquid carrier. The use of powder may be expensive as it requires large quantity of powder and if a non-uniform amount of powder is applied there may be inconsistent results. Also much powder on the base of the cutting can sometimes stop the outgrowth of the new roots and the powder loses its effectiveness if not kept dry and in a refrigerator. Liquid carrier may be alcohol or water, while alcohol is very expensive water is cheap and easily available. Moreover, IBA dissolved in water is more effective for rooting most plant species than IBA dissolved in alcohol. High concentration of alcohol may dehydrate, injure and be toxic to basal stems, scions and other plant tissue.

**Low cost option for type of rooting media to be used:** The importance of rooting media for rooting of cuttings is widely known to provide sufficient porosity to allow good

aeration and ensure adequate oxygen availability for the developing rooting system (Hartmann *et al.*, 2002; Baiyeri, 2005; Allaire *et al.*, 2004; Shah *et al.*, 2006). Low cost option for rooting media should be single or mixtures of common and easily available materials, such as river sand, sharp sand, grit or fine gravel, forest top soil, old weathered sawdust and coconut palm husks. It is not necessary to buy expensive materials such as peat, vermiculite or perlite, except for special purposes.

Sand as low cost rooting media has been identified as the best rooting media for stem cuttings of *Gongronema latifolia* (Agbo and Omaliko, 2006), *Dalbergia melanoxylon* (Amri and Friedrichs, 2006; Amri *et al.*, 2009). The use fine sand and gravel as rooting media has also been reported to enhance high rooting percentage for stem cuttings of *Cordia alliodora* and *Vochysia hondurensis* respectively (Leakey *et al.*, 1990). Vegetative propagation by stem cuttings of *Irvingia wombolu* revealed high rooting percentage in coarse sand rooting media (Dickens *et al.*, 2009). Sawdust also as low cost rooting media has been reported with highest rooting percentages of *Milicia excelsa* (Ofori *et al.*, 1996). Stem cuttings of *Pausinystalia johimbe* rooted better in sand than in the mixture of sand/sawdust (Tchoundjeu *et al.*, 2004). Sand, gravel, forest top soil and saw dust are easy low cost locally available rooting media which can be used for propagation of tropical trees. However, forest top soil is not advisable because it is bulky, heterogeneous in mineral composition and could cause environmental hazard such as soil erosion (Ekwu and Mbah, 2001).

**Low cost consideration for sites of cuttings collection:**

There is a need to recognize the additional expense associated with site for cuttings collection and the cost of maintenance of these sources. Cutting sites or sources associated with factors such as diseases, insects and environmental stresses on donor plants may increase the cost of production (Amri, 2009). All donor plants sources should be relatively vigorous and free of insects and disease (Hartmann *et al.*, 2002). One year prior to taking cuttings, potential donors should be scouted during the growing season, their location marked and recorded and the plants revisited just prior to taking cuttings to assure the availability of an adequate quantity and quality of cuttings. Avoid plants showing signs of severe environmental stress or isolated groups of few individual plants.

The low-cost vegetative propagation of tree species is an important component of strategies to achieve the UN Millennium Development Goals through its contribution to eradication of extreme poverty, enhance food and nutritional security and ensuring environmental sustainability (Garrity, 2004). Low-cost vegetative

propagation will not only stay a high priority in forestry for tropical areas but also in agriculture, horticulture and floriculture of many developing countries for the production of suitably priced high quality planting material.

### CONCLUSIONS

Low cost vegetative propagation can be achieved through consideration of factors useful in improving process efficiency and better utilization of resources. For tropical trees, vegetative propagation should be particularly interested in methods for producing own rooted plants, which require low investments in equipment, inputs and personnel training. It is recommended that potentially low cost factors should be considered for vegetative propagation of tropical trees, since the resources of trained personnel and equipment are often not readily available. This review has focused on how to lower the cost for vegetative propagation, the factors discussed here can be adopted in tropical areas by forest tree breeders or farmers for domestication of economically important tropical tree species which are threatened by over exploitation.

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