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# Micropropagation of ornamental plants: Empirical implementation of the phase inclusive of its mercantile uses

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## ABSTRACT

Micro-propagation is an advanced and well-suited technology for the quick multiplication of plants. Due to the quick rate of multiplication, superior plant quality, and capacity to generate disease-free plants, it offers a significant potential for financial gain. To meet the demands of the expanding global population, it should be applied widely to create new vegetable types and boost vegetable output. A variety of methods for quick multiplication and enhancement of a wide range of horticultural crops and their production systems have been developed as a result of advancements in this field. One of the biggest benefits of micro-propagation is the minimal growing space needed in commercial nurseries. Within the culture vials, thousands to millions of plantlets can be kept alive. This approach is more beneficial for plants that grow slowly, where the seeds are the only propagule and are produced over an extended period of time. It is fairly common to employ attractive plant planting materials in both business and residential landscaping and household gardens. For growers to become more productive, high-quality planting material is a necessary requirement. Around the world, 700 million plants are produced in vitro, according to the most recent figures. Brazil, Ecuador, and Colombia are the top three countries for ornamental plant in vitro propagation, followed by Israel, the United States, and India. In vitro propagation has been heavily utilized in the ornamental industry for mass plant reproduction of superior elite varieties. Numerous plant tissue culture facilities have sprung up all over the world, especially in developing countries, due to the low cost of labour. Due to the higher cost of micro propagation technology compared to conventional propagation techniques, steps must be taken to cut manufacturing costs in order to lower the cost per plant. The use of biotechnological tools and techniques increase the productivity of ornamental plants is supported by a large number of articles.

*Key words: Ornamental, Micro propagation, Biotechnology, Landscaping*

## Introduction

The regenerative potentials of orchid propagation give rise to the concepts of micro-propagation or clonal propagation. When orchid explants are cultivated on the medium, protocorm-like bodies, a top-shaped structure, are produced (PLBs). When protocorms (PLBs) are cut into a variety of parts for

scientific research, each component regenerates into a different set of protocorms. During the entire protocorm multiplication process, a small number of millions of orchid plants were created in culture. Later, this method was used to other significant horticultural plants. As a good source of tissue for the multiplication of various woody plants, the micro propagation technique has been used to propagate

tree species, woody species, immature embryos, hypocotyl, young leaves, cotyledons, and other juvenile tissues. Micro-propagation techniques that utilise in vitro flowering can reduce the length of the juvenile stage. The problem caused by deforestation and the declining population of useful plants can be solved by improvising the techniques of a well-defined micro-propagation technique, even though micro-propagation is a new technique and must overcome several obstacles. To propagate plants, the majority of commercial laboratories nowadays use micro propagation methods.

### Micro-Propagation of Ornamentals

Growing attractive plants for the horticultural sector is one of the plant tissue culture's most varied uses. Many businesses in the USA, Europe, and Australia engage in the vast micro-propagation of ornamental plants meant for sale as pot plants, cut flowers, bulbs, and corms, as well as plants meant for re-planting. Shoot tip culture is used to multiply a large number of kinds of herbaceous ornamental plants. Because of the large commercial potential for anthuriums, extensive study has been done in Europe to develop ways for their propagation. The use of shoot tip culture for commercial multiplication has become widespread. Several taxa in the family Gessneriaceae, including *Santpaulia* and *Navtilocalyx*, may be easily multiplied by adding BAP and NAA to the adventitious shoots that grow on lamina petioles. The approach is expanded to include other genera. Methods for propagating bulbs and corms in vitro were standardised by European researchers. The methods rely on axillary shoot proliferation, induction of adventitious shoots on organs, and callus-mediated adventitious creation. In vitro methods can generate at least 1000 *Narcissus* bulbs in a year, but conventional methods take 6 years to produce the same amount. Shoot tip culture is used to multiply a large number of kinds of herbaceous ornamental plants. Because of the large market potential for anthuriums, extensive study has been done in Europe to develop ways for their propagation. The use of shoot tip culture for commercial propagation has become widespread. A number of gessneriaceae taxa, including *Santpaulia* and *Navtilocalyx*, can easily be multiplied from adventitious shoots developed on lamina petioles fed with BAP and NAA. The approach is expanded to include other genera. Methods for propagating bulbs and corms in vitro were standardised by European

researchers. The methods rely on axillary shoot proliferation, induction of adventitious shoots on organs, and callus-mediated adventitious creation. In vitro methods can generate at least 1000 *Narcissus* bulbs in a year, but conventional methods take 6 years to produce the same amount. Bulbils develop directly on the bulb scale as a result of exogenous auxin input. Compared to other bulbil species, tulips have proven to be more challenging to propagate using tissue culture. The issue of multiplication is, however, solved by two methods, including the culture of immature flower portions and the dissection of 3-5 mm axillary buds from sprung buds. It has been demonstrated that ornamentals like gerberas and geraniums can be successfully grown in vitro using modified Murashige and Skoog conditions. Owing to their stunning, long-lasting blossoms, orchids have been discovered to hold a distinctive place among ornamental plants. For orchid micro-propagation, in vitro flasking and mericlone procedures are frequently used. Mericlone refers to orchid species that were developed in culture from somatic protocorms and are descended from a single mother plant. Shoot tip culture is distinct from mericlone. Using micro-propagation techniques, several orchid genera including *Cymbidium*, *Dendrobium*, *Phaelonopsis*, and *Aranda* have been successfully multiplied.

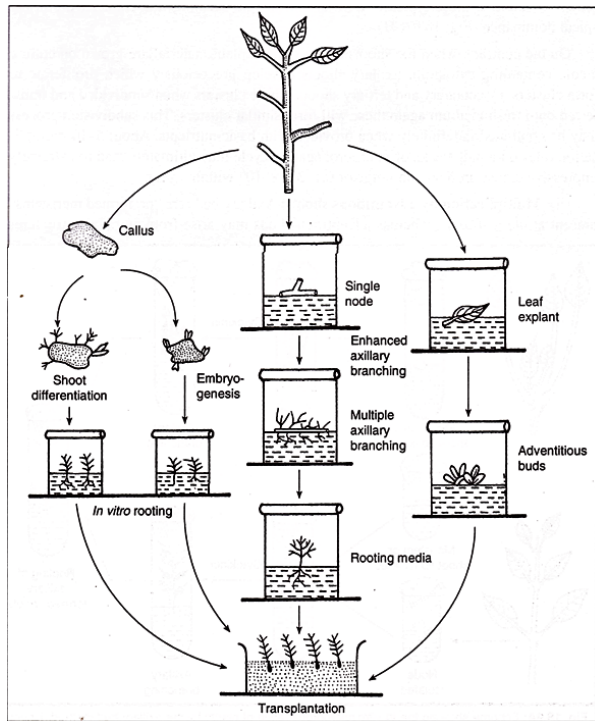
### Multiplication

Clonal propagation is the process of producing genetically identical duplicates of a cultivar through asexual reproduction. In vivo clonal propagation is frequently challenging, costly, and even unsuccessful. An alternate form of clonal propagation, referred to as micro-propagation, is provided by the tissue culture method. With this technique, a clone can quickly and efficiently grow a large number of small vegetative shoots. G started using tissue culture for micro-propagation.

### Methods of Micro-Propagation

Multiplication by Meristematic Tissue of Axillary and Apical Shoots:

Depending on the physiological state of the plant, the meristems in the axillary and apical shoots are dormant or active. These shoot tips often grow into single seedling-like shoots with high apical dominance when cultivated on a basal medium devoid of growth regulators. On the other hand, axillary shoots sprout prematurely and multiply to produce



clusters of secondary and tertiary shoots when the shoots of the same explant material are cultured on culture media containing cytokinin. This subdivision process may be continued indefinitely while essential nutrients are available. These clusters will create similar clusters when separated and put onto new medium once more. Approximately 5-10 multiplication rates in a micro-propagation cycle lasting 4 to 8 weeks might ultimately result in an incredibly impressive clonal propagation range of  $0.1-3 \times 10^6$  within that 1 Year cycle.

### Multiplication by Adventitious Shoots

Adventitious bud may originate from any plant structure, but a xillary bud is a prepared meristem that is present at the leaf axils. This regeneration is frequently reliant on the presence of organised plant tissue. The explants can be stems, internodes, leaf blades, cotyledons, root elongation zones, bulbs, corms, tubers, rhizomes, etc. If these explants are stimulated by adding the right amount of growth regulators to the medium, they will form meristematic zones that regenerate multiple shoots on an appropriate culture medium. A single epidermal cell can produce a large number of adventitious shoots. Two vertically split shoot bases can be grown to promote continuous adventitious shoot proliferation from bulbs and corms. The abaxial surfaces of grow-

ing leaves and scales may produce clusters of shoots. It has been discovered that trimming the shoot apices is a good strategy for maintaining consistently productive cultures of various hybrid plants for an unlimited amount of time.

### Multiplication by Adventitious Embryo Formation

It is yet another practical strategy used for numerous significant plant species. These adventitious embryoids are diploid in nature and can be used as clonal material for micro-propagation. Adventitious embryos can develop directly from a group of cells within the original explants or from primary embryoids. There are many plant species that develop embryos in vivo from diverse kinds of explants, for example, the orchid leaf tips produce large numbers of embryoids. Similar to this, in vitro adventitious embryos derived from various explants can be useful building blocks for clonal proliferation.

### Multiplication through Callus Culture

For micro clonal propagation, direct plantlet creation from the explants in culture is preferable. But shoot creation may occur by organogenesis or embryogenesis from the callus formed from the explant. The limitation of this method is that the callus cells are not genetically stable thus it cannot be called as a single clone and this procedure is more time consuming. Due to the genetic instability, it is also possible that plant regeneration capacity will decrease. Genetically stable calli have also been developed in some plant species; in these calli, the peripheral layer forms slow-growing meristematic zones. There are always diploid cells exhibiting totipotency in the meristematic layers.

### Stages of Micro-Propagation

Micro-propagation, also known as clonal propagation, is a difficult process that may be broken down into four stages of activity:

Stage 0: The stock plants must be grown under controlled conditions in this first stage of micro-propagation before being used to start the culture.

Stage I: Explants from stock plants are first prepared, and then they are established in a suitable culture medium. The steps involved in this stage are:

- (a) Isolation of explants,
- (b) Sterilization of surfaces,
- (c) Washing,
- (d) Setting up the explant on the proper culture media.

Stage II: A variety of methods are used for micro-propagation, including: - Multiplication through the growth and proliferations of meristems excised from apical and axillary shoot of the parent plant - Induction and multiplication of adventitious meristems through processes of organogenesis or somatic embryogenesis directly on explants. Multiplication of calli derived from any source. Typically, the harvest cycle lasts 4 to 8 weeks. The shoots are either promptly sold or carried on to the following stage for more growth.

Stage III: Shoots obtained from stage II are moved to the next rooting or storage media. As micro-cuttings, these shoots are directly planted in the soil to grow roots. The management of the shoots at this stage varies according to the species. If the shoots are placed directly in the soil, the following processes must be followed:

- A. Each shoot needs to be rooted separately.
- B. The shoots are hardened to strengthen their resistance,
- C. Allowing the establishment of autotrophic plants in place of heterotrophic plants in culture,
- D. Achieving the conditions needed to emerge from hibernation.

Stage IV: - It is accomplished to transfer plantlets to sterilised soil for hardening in a greenhouse environment. This step is to make sure that the plantlets from stage III or the unrooted shoot apices from stage II are successfully transplanted into the appropriate compost mixture or soil in containers under controlled lighting, temperature, and humidity conditions. These plants are sometimes grown in artificial growing media, such as soilless mixtures, Rockwood plugs, or sponges, for marketing purposes. The finished items are marketed between four and sixteen weeks.

### Micro-Propagation Advantage

A million shoot tips can be extracted from a tiny, microscopic piece of plant tissue in a short amount of time and space using the technique of micro-propagation, which is an alternative to traditional methods of vegetative propagation and has an enhanced rate of multiplication. The benefit of this type of propagation is that shoot multiplication typically has a short cycle (2–6 weeks), and each cycle results in a logarithmic increase in the number of shoots. Stocks of germplasm can be kept for many years using this method of propagation. This method is particularly appropriate if disease free

propagules are desired. The propagules can be maintained in a soil-free environment which facilitates their storage on a large scale. Micro-propagation is extremely helpful when dealing with dioecious plants because there the seed progeny yield is 50% male and 50% female, but this technique helps to get the progeny according to the desired sex. One of the biggest benefits of micro-propagation is the minimal growing space needed in commercial nurseries. Within the culture vials, thousands to millions of plantlets can be kept alive. This approach is more beneficial for plants that grow slowly, where the seeds are the only propagule and are produced over an extended period of time. This technique can get around the propagules being hard to come by. It's not always possible to produce seeds that will produce genetically uniform offspring. Micro-propagation is one of the best methods for plant multiplication via in vitro technique of plant tissue culture, and it will help to retain the genetic consistency in the propagules. As it is the simplest way to produce numerous propagules, the more recent tissue material obtained using r DNA technology, haploid culture, or somatic hybridization can be used as the source of tissue material for micro-propagation.

### Commercial Uses of Micro-Propagation

Apart from orchids, about 600 species of other ornamental plants have been successfully cloned; some of them are commercially exploited (Chrysanthemum, Carnation, Gerbera, and Anthurium), and the list of names is growing daily. The advantage is rapid cloning of selected colour individuals. Besides horticultural flowering plants, a large number of forest trees, fruit trees, and oil producing plants (Eucalyptus) have also been successfully cloned. Due to their lengthy generation times and difficulty in vegetatively propagating, sexual hybridization breeding and selection is a very slow process. In vitro cloning of more than 100 woody species across a wide range of families has thus been accomplished. Forest trees are significant contributors to our biodiversity as well as sources of our food, fuel, building materials, and industrial products, all of which are depleting at an unprecedented rate. For specific types of forest trees, including roughly 30 forest tree species of Gymnosperm and 70 forest tree species of Angiosperm, large-scale, cost-effective in vitro techniques have been established. The economics and plantlet unit cost associated in commercial Micropropagation are crucial factors. Research find-

ings in the area of micro-propagation are not always commercially feasible and are frequently rejected by business establishments. The cost of the laboratory setup, the type of plant to be propagated, and the skill required will all have a significant impact on the investment made in a commercial tissue culture business. The commercial nurseryman should begin with crop species for which published methods are available, and it is also crucial that the grower has some experience with tissue culture and plant husbandry. Automating micro-propagation at different stages is another strategy used. In this context, bioreactors are used for large-scale multiplication of somatic embryos, shoots, and bulbs. During phases III and IV of micro-propagation, shoots were sub cultured using automation. This lowers the labour component of micro-propagation.

### Conclusion

The micro-propagation technique, it is concluded, offers a high potential for financial gain, high plant quality, and the capacity to generate disease-free plants. To meet the demands of the expanding global population, it should be applied widely to create new vegetable types and boost vegetable output. A variety of methods for quick multiplication and enhancement of a wide range of horticultural crops and their production systems have been developed as a result of advancements in this field. Micro-propagation is an advanced and well-suited technology for the quick multiplication of plants. Due to the quick rate of multiplication, superior plant quality, and capacity to generate disease-free plants, it offers a significant potential for financial gain. Plant propa-

gation in vitro is a combination of art and science. Stock plant maintenance, explant section and sterilisation, medium modification to obtain proliferation, roots, acclimation, and growing plants under field settings are all processes in the propagation process. Micropropagation is typically done by hand since it demands a hygienic working environment, which makes the process expensive and boring for the employees. There are three different vegetative propagation methods used in micropropagation:- Somatic embryogenesis, in which structures with a shoot and root are formed and adventitious shoot production, which involves the meristem formation from callus tissue or directly from organised tissues, such as epidermal or sub-epidermal cells; and axillary shoot production, in which axillary buds and meristems give rise to shoots that are used to produce additional clonal shoots. Meristem culture, organogenic micro propagation from undifferentiated tissues, cells, or protoplasts, zygotic embryo culture, somatic embryogenesis, and gametic embryogenesis are only a few instances of effective uses that have been documented.

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