

PLANT TISSUE CULTURE TECHNOLOGY IN HORTICULTURE CROP PRODUCTION

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ARTICLE ID: 48

Introduction

Tissue culture is defined as the technique of isolating small portions of living tissues (explants) from an organism and growing them aseptically on a nutrition medium under controlled conditions such as light, temperature, humidity or Plant tissue culture is a set of procedures for maintaining or growing plant cells or tissues in sterile circumstances using a specified nutritional medium. It is commonly employed in the production of plant clones, a process known as micropropagation (Kruse and Patterson, 1973).

The most fundamental and characteristic function of plant cells is Totipotency. It is defined as the genetic potential of plant somatic cells to produce the entire plant. The first evidence of totipotency of plant cell was given by Haberlandt (1950) and he established plant tissue culture by exploiting totipotency. Therefore, he is known as father of Plant Tissue Culture.

The works of the teacher and student Murashige and Skoog (1962) duo has helped in the creation of the nutrient culture medium popularly known as MS media. As the requisite for any work in plant, tissue culture is MS media.

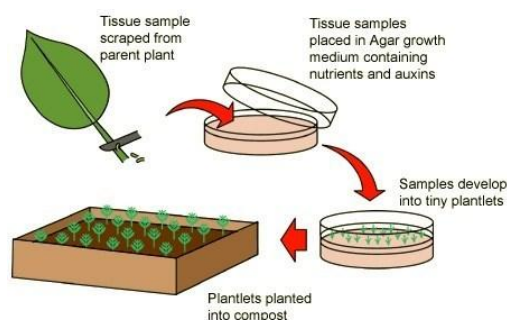


Fig.1 Plant Tissue Culture Technology

Scenario in India:

- First tissue culture laboratory set up in 1984 – for ornamentals, banana, and cardamom.
- Approximately 122 companies have plans for selling tissue cultured horticultural crops.
- Installed capacity: more than 245 million plantlets per annum.

Facilities:

The research as well as the production of the tissue cultured planting materials happens in the state-of-the-art plant tissue culture laboratory. This well-structured and well-kept plant tissue culture laboratory makes working here a truly enjoyable experience. It has a floor space of 1600 sq. ft. The plant tissue culture laboratory is separated into 2 divisions- the Research and Development unit and the production unit.

Production Unit:

- The media preparation room and the sterilization room have all the necessary instruments like Horizontal autoclave, vertical autoclave, pH meter, distillation unit and BOD incubator.
- The culturing room or the culture transfer room has some advanced and cutting edge florescent microscopes and laminar air flows, which are used for the transfer of explants into the sterilized media.

- The bottles containing the media and the explants are then transferred to the growth room, which can hold 50,000 culture bottles.
- The plant tissue culture laboratory also has a tunnel house where primary hardening of the regenerated plants takes place.
- The laboratory also has a green house where the saplings after primary hardening are transferred to for secondary hardening.



Fig. Inoculation room

Research and Development Unit:

The R and D unit also has all the above mentioned facilities and also some other instruments like the light intensity meter, humidity regulators, digital calipers, spectrophotometer and fluorescent microscopes. It also has a well-equipped observation rooms.

Benefits of Tissue Culture in Horticultural Crops

- Faster multiplication of plants in a shorter period of time.
- Somaclonal variation in varietal improvement.
- Haploid production.
- Protoplast fusion for somatic hybridization.
- Production of synthetic seeds through somatic embryogenesis.
- Multiplication of flower bulbs in bioreactors.
- *In vitro* preservation of germplasm.

- Year round production of plants in a less space.

Application of Tissue Culture Technology in Horticultural Crops:

Approach used Horticulture crops

- Micropropagation Banana, Papaya, Strawberry, Orchids, Anthurium, Cardamom
- Meristem culture Banana, Citrus, Dahlia, Gladiolus, Liliun, Rose, Gerbera
- Embryo rescue Liliun, Chrysanthemum, Pelargonium, Peach
- Haploid culture Datura, Pelargonium, Liliun, Gerbera
- Somatic embryogenesis Iris, Anthurium, Oil Palm, Date Palm, Coffee, Mango, Guava

Micropropagation:

- Most widely used biotechnological tool in horticulture crops.
- Well tested protocols available for banana, papaya, strawberry, orchids, Anthurium, Cardamom, etc.
- Efforts on to standardize protocols for mango, litchi, cashew.
- Adventitious shoot formation is widely adopted.
- On an average, 10-fold increase in shoot number per monthly culture passage (Arditti, 2009).

Meristem culture:

- Meristem culture *In vitro* used for the elimination of viruses and related pathogens from a large number of vegetatively propagated plants.
- The main method used in plant virus elimination programs.
- Well tested protocols standardized for Banana, Citrus, Dhalia, Gladiolus, Liliun, Rose, Gerbera (Kantha, 1985).

Embryo rescue:

- Embryo rescue: to overcome post-zygotic incompatibilities. Eg: Liliun, Carnation and Chrysanthemum.
- Rescue of embryos in early ripening cultivars Eg: Peach.
- Promotes germination in dormant seeds Eg: Iris, Maranta, Peach (Sharma *et al.*, 1996).

Haploid Culture:

- Culture of ovules, anthers and microspores.
- Used in breeding programmers as parents.
- Added benefits such as smaller flowers, prolonged flowering time, increased production of flowers, resistance to diseases etc.
- Attempted in Datura, Pelargonium, Liliium and Gerbera (Niizeki, and Oono, 1968).

Somatic Embryogenesis:

- Embryos originate from single somatic cells.
- Regeneration from somatic embryos attempted in Iris, Anthurium, Carambola, Mango, Guava, Oil Palm, Coffee.
- Bioreactors- to produce somatic embryos on a commercial scale.
- Fluid drilling and encapsulation also considered (Merkleet *al.*, 1995).

Somaclonal Variation:

- Phenotypically and genotypically variant plants are obtained.
- Frequently obtained when callusing stage is involved.
- Extensively studied in Carnation, Chrysanthemum, Potato, Pineapple and Tomato (Bairuet *al.*, 2011).

Low cost alternatives in tissue culture technology:

Media:

- **Water source:** Double distilled water, Single distilled water, Aqua guard filtered water, Membrane filtered water.
- **Carbon source:** Sucrose, Glucose, Fructose, Maltose, Market Grade Sugar, syrup, Sugar cane juice, Jaggery.
- **Gelling agent Agar :** Wheat flour, Tapioca starch, Corn starch, Potato powder

Sago, Rice powder, Sweet potato starch, Ragi powder.

- **Physical Matrices:** Luffa sponge, Coir, Cotton fibre, Glass wool cloth, Filter paper (Whatman), Polysterene foam block, Bagasse, Rock wool.

Other factors:

- **Culture Containers:** Baby jars, Test tubes, Conical flasks, Laxentapolypropelene Polypropylene bags.
- **Culture Transfer:** Laminar Air Flow Chamber, Wooden hood, Box (Glove Box), Open laboratory bench with bunsen burner
- **Light Source:** Harvest natural source, Exploit light and dark conditions.
- **Rooting:** *In vitro* rooting, *Ex vitro* rooting.
- **Hardenig:** Primary hardening, Secondary hardening.

Conclusion

The only technology which will have universal appeal and sustained usage will be the one which is practicably feasible and economically viable.

References:

Arditti, J.,(2009). Micropropagation of orchids. *John Wiley & Sons*.

Bairu, M.W., Aremu, A.O. and van staden, J.(2011). Somaclonal variation in plants: causes and detection methods. *Plant Growth Regulation*, 63(2):147-173.

Haberlandt, H., Hernegger, F. and Scheminzky, F. (1950). The fluorescence spectra of uranium minerals in filtered ultraviolet light. *Spectrochimica Acta (England) Divided into Spectrochim. Acta, Part A and Spectrochim. Acta, Part B*, 4.

- Kartha, K.K. (1985). Meristem culture and germplasm preservation. *Cryopreservation of plant cells and organs*.115-134.
- Kruse, P.F. andPatterson, M.K. (1973). Tissue culture. San Diego, CA: Academic Press.
- Merkle, S.A., Parrott, W.A. andFlinn, B.S. (1995). Morphogenic aspects of somatic embryogenesis. *In in vitro embryogenesis in plants*,Springer, Dordrecht. 55-203.
- Murashige, T. andSkoog, F. (1962).A revised medium for rapid growth and bio assays with tobacco tissue cultures. *Physiol. plant*. 15(3):473-497.
- Niizeki, H. andOono, K. (1968). Induction of haploid rice plant from anther culture. *Proceedings of the Japan Academy*. 44(6):554-557.
- Sharma, D.R., Kaur, R. andKumar, K.(1996). Embryo rescue in plants-a review. *Euphytica*. 89(3):325-337.