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Topic- Idea of tissue culture with special reference to plant propagation

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Idea of tissue culture with special reference to plant propagation

In biological research, tissue culture refers to a method in which fragments of a tissue (plant or animal tissue) are introduced into a new, artificial environment, where they continue to function or grow. While fragments of a tissue are often used, it is important to note that entire organs are also used for tissue culture purposes. Here, such growth media as broth and agar are used to facilitate the process.

While the term tissue culture may be used for both plant and animal tissues, plant tissue culture is the more specific term used for the culture of plant tissues in tissue culture.

Plant tissue culture is a collection of techniques used to maintain or grow plant cells, tissues or organs under sterile conditions on a nutrient culture medium of known composition. It is widely used to produce clones of a plant in a method known as micropropagation. Different techniques in plant tissue culture may offer certain advantages over traditional methods of propagation, including:

- The production of exact copies of plants that produce particularly good flowers, fruits, or have other desirable traits.
- To quickly produce mature plants.
- The production of multiples of plants in the absence of seeds or necessary pollinators to produce seeds.
- The regeneration of whole plants from plant cells that have been genetically modified.
- The production of plants in sterile containers that allows them to be moved with greatly reduced chances of transmitting diseases, pests, and pathogens.
- The production of plants from seeds that otherwise have very low chances of germinating and growing, i.e. orchids and *Nepenthes*.
- To clean particular plants of viral and other infections and to quickly multiply these plants as 'cleaned stock' for horticulture and agriculture.

Plant tissue culture relies on the fact that many plant cells have the ability to regenerate a whole plant (totipotency). Single cells, plant cells without cell walls (protoplasts), pieces of leaves, stems or roots can often be used to generate a new plant on culture media given the required nutrients and plant hormones.

Types of Tissue Culture

Seed Culture

Seed culture is the type of tissue culture that is primarily used for plants such as orchids. For this method, explants (tissue from the plant) are obtained from an in-vitro derived plant and introduced in to an artificial environment, where they get to proliferate. In the event that a plant material is used directly for this process, then it has to be sterilized to prevent tissue damage and ensure optimum regeneration.

Embryo Culture

Embryo culture is the type of tissue culture that involves the isolation of an embryo from a given organism for in vitro growth.

*Note, the term embryo culture is used to refer to sexually produced zygotic embryo culture.

Embryo culture may involve the use of a mature or immature embryo. Whereas mature embryos for culture are essentially obtained from ripe seeds, immature embryo (embryo rescue) involves the use of immature embryos from unripe/hybrid seeds that failed to germinate. In doing so, the embryo is ultimately able to produce a viable plant.

For embryo culture, the ovule, seed or fruit from which the embryo is to be obtained is sterilized, and therefore the embryo does not have to be sterilized again. Salt sucrose may be used to provide the embryo with nutrients. The culture is enriched with organic or inorganic compounds, inorganic salts as well as growth regulators.

Callus Culture

*Callus - This is the term used to refer to unspecialized, unorganized and a dividing mass of cells. A callus is produced when explants (cells) are cultured in an appropriate medium - A good example of this is the tumor tissue that grows out of the wounds of differentiated tissues/organs.

In practice, callus culture involves the growth of a callus (composed of differentiated and non-differentiated cells), which is followed by a procedure that induces organ differentiation.

For this type of tissue culture, the culture is often sustained on a gel medium, which is composed of agar and a mixture of given macro and micronutrients depending on the type of cells. Different types of basal salt mixtures such as Murashige and Skoog medium are also used in addition to vitamins to enhance growth.

Organ Culture

Organ culture is a type of tissue culture that involves isolating an organ for in vitro growth. Here, any organ plant can be used as an explant for the culture process (Shoot, root, leaf, and flower).

With organ culture, or as is with their various tissue components, the method is used to preserve their structure or functions, which allows the organ to still resemble and retain the characteristics they would have in vivo. Here, new growth (differentiated structures) continues given that the organ retains its physiological features. As such, an organ helps provide information on patterns of growth, differentiation as well as development.

There are a number of methods that can be used for organ culture. These include;

- Plasma clot method - Here, the method involves the use of a clot that is composed of plasma and chick embryo extract (or any other extract) in a watch glass. This method is particularly used for the purposes of studying morphogenesis in embryonic organ rudiments and more recently for studying the actions of various hormones, vitamins and carcinogens of adult mammalian tissues.
- Raft method - For this method, the explant is placed on a raft of lens paper/rayon acetate and floated on a serum in a watch glass.
- Agar gel method - The medium used for this method is composed of a salt solution, serum as well as the embryo extract or a mixture of various amino acids and vitamin with 1 percent agar. The explant has to be subcultured every 5 to 7 days. The method is largely used for the study of developmental aspects of normal organs and tumors.

- Grid method - Grid method, as the name suggests involves the use of perforated stainless steel sheet, on which the tissue of interest is placed before being placed in a culture chamber containing fluid medium.

Protoplast Culture

*Protoplast -cells without cell walls. A protoplast is the term used to refer to cell (fungi, bacteria, plant cells etc) in which the cell wall has been removed, which is why they are also referred to as naked cells.

Protoplasts may be cultured in the following ways;

- Hanging-drop cultures
- Micro culture chambers
- Soft agars matrix

Once a protoplast has regenerated a cell wall, then it goes through the process of cell division to form a callus, which may then be subcultured for continued growth.

Protoplast culture is an important method that provides numerous cells (single cells) that can be used for various studies. These include;

- Protoplast culture regenerated into a whole plant
- Development of hybrids
- Cell cloning
- Genetic transformations
- Membrane studies

In protoplast culture, a number of phases can be observed. These include;

- Development of a cell wall
- Cell division
- Continuous growth or regeneration to a whole plant

For plants, some of the special requirements include;

- Less amounts of iron and zinc and no ammonium
- Higher concentration of calcium
- High auxin/kinetic ratio for cell division and high kinetin/auxin ration for regeneration
- Glucose and vitamins

Some of the other types of tissue culture include;

- Single cell culture
- Suspension culture
- Anther culture
- Pollen culture
- Somatic Embryogenesis