# Plant tissue culture

**Plant tissue culture**: is the growth of explant (any plant part) or plant cells *in vitro* (in the laboratory culture media).

- Plant cell culture based on the unique property of the cell-totipotency.
- Cell-totipotency is the ability of the plant cell to regenerate into a whole plant under the laboratory conditions using artificial nutrient mediums.

## • Nutrient media

The nutrient media vary in type and quantity of materials used and the choice depends on the type of plant to be grown. Some have a better growth in solid media than in liquid media. The best known nutrient media are: White's medium, MS medium, B5 medium. The ideal pH of the culture medium is 5.8, and if the pH values of more than 7 or less than 4.5, there is inhibition of growth.

## Stages of plant tissue culture

There are four stages of plant tissue culture:

- Initiation stage. A piece of plant tissue (called an Explant) is (a) cut from the plant,
  (b) disinfected (removal of surface contaminants), and (c) placed on a medium. A medium typically contains mineral salts, vitamins, sucrose, antibiotics (optional), and a solidifying agent such as agar. The objective of this stage is to achieve an aseptic culture. An aseptic culture is one without contaminating bacteria or fungi.
- 2. **Multiplication stage.** A growing of explant can be induced to produce vegetative shoots by including a cytokinin in the medium. A cytokinin is a plant growth regulator that promotes shoot formation of growing plant cells.
- 3. **Rooting stage.** Growing shoots can be induced to produce adventitious roots by including an auxin in the medium. Auxins are plant growth regulators that promote root formation.
- 4. Acclimatization. A growing, rooted shoot can be removed from tissue culture and placed in soil. When this is done, the humidity must be gradually reduced over time because tissue-cultured plants are extremely susceptible to wilting.



### Stages of plant tissue culture

### Types of cultures

- Organ Culture (such as the ovary culture and endosperm culture).
- Explant culture (stem, root, flower.... etc).
- Callus culture (is a mass of undifferentiated parenchyma cells).
- Cell suspension cultures.
- Protoplast culture (The protoplast is a cell without cell wall and it obtained by using enzymes such as cellulases and pectinases).
- Embryo culture.
- Anther and Pollen Culture (They used in the production of haploid plants).



**Cell suspension cultures** 

#### Some Applications of Cell and Tissue Culture

### 1- Micropropagation /Clonal Propagation

- Clonal propagation is the process of a sexual reproduction by multiplication of genetically identical copies of individual plants.
- Micropropagation is the tissue culture methods of plant propagation. The micropropagation is rapid and has been adapted for commercialization of important plants such as banana, apple, and other plants.

### 2- Production of virus free plants

It has become possible to produce virus free plants through tissue culture at the commercial level. Among the culture techniques, meristem-tip culture is the most reliable method for virus and other pathogen elimination.

#### **3-** Production of synthetic seeds

In synthetic seeds, the somatic embryos are encapsulated in a suitable matrix (e.g. sodium alginate), along with substances like mycorrhizae, insecticides, fungicides and herbicides.

#### 4- Production of secondary metabolites

The most important chemicals produced using cell culture are secondary metabolites. These secondary metabolites include alkaloids, glycosides (steroids and phenolics), terpenoids, latex, tannins etc.

#### Transgenic plants with beneficial traits

**1-** Transgenic plants or transgenic crops are the plants, in which a functional foreign gene has been incorporated by any biotechnological methods that generally are not present in the plant.

2- Transgenic plants have many beneficial traits like insect resistance, herbicide tolerance, delayed fruit ripening, improved oil quality, weed control etc, but the main goal of producing transgenic plants is to increase the productivity.

#### Some of the traits introduced in these transgenic plants are as follows:

#### **Stress tolerance**

Biotic stresses (viral, bacterial infections, pests and weeds) and abiotic stresses (physical actors such as temperature, humidity, salinity etc).

#### Herbicide tolerance

Several biotechnological strategies for weed control are being used e.g. the overproduction of herbicide target enzymes (usually in the chloroplast) in the plant which makes the plant insensitive to the herbicide.

#### Virus resistance

There are several strategies for engineering plants for viral resistance, and these utilizes the genes from virus itself. The virus coat protein-mediated approach is the most successful one to provide virus resistance to plants.

### **Insect resistance**

The transgenic technology uses eco-friendly method to improve pest control management. The first genes available for genetic engineering of crop plants for pest resistance were Cry genes (popularly known as Bt genes) from *Bacillus thuringiensis*. These are specific to a particular group of insect pests, and are not harmful to other useful insects like butterflies and silk worms.



#### **Delayed fruit ripening**

The gas hormone, ethylene regulates the ripening of fruits, therefore, ripening can be slowed down by blocking or reducing ethylene production. This can be achieved by introducing ethylene forming gene(s) in a way that will suppress its own expression in the crop plant.



## Some of the uses of transgenic plants are:

- Improvement of Nutrient quality
- Improvement of seed protein quality
- Diagnostic and therapeutic proteins
- Edible vaccines
- Biodegradable plastics