

DEPARTMENT OF BIOTECHNOLOGY

ORGANOGENESIS

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INTRODUCTION

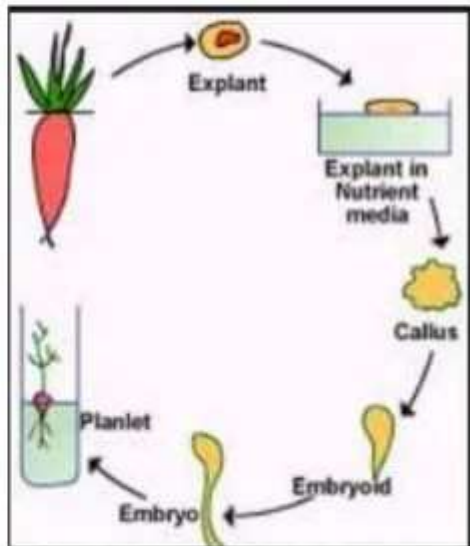
Plant tissue culture - **Plant tissue culture** is a collection of techniques used to maintain or grow plant cells, tissues or organs under sterile conditions on a nutrient culture medium of known composition. Plant tissue culture is widely used to produce clones of a plant in a method known as micropropagation.

The three common pathways of plant tissue culture regeneration are;

- (i) propagation from pre-existing meristems (shoot culture or nodal culture)
- (ii) organogenesis
- (iii) non-zygotic (somatic) embryogenesis

organogenesis

- A plant contains many organs like meristem, cortex, phloem, epidermis are consist of structural unit called cell. because an cell have to nature of create whole plant like any organ or tissue of plant also show same nature mean they also create to whole plant in in-vitro condition.
- If plant organs used in in-vitro conditions to generated new plant this process called organogenesis.



Definition

❑ **Definition of Organogenesis:**

- “The development of adventitious organs or primordia from undifferentiated cell mass in tissue culture by the process of differentiation is called organogenesis.

Or

- “The formation of roots, shoots or flower buds from the cells in culture in manner similar to adventitious root or shoot formation in cuttings is called organogenesis.

❑ **Caulogenesis:**

- Type of organogenesis by which only adventitious shoot bud initiation take place in the callus tissue.

❑ **Rhizogenesis:**

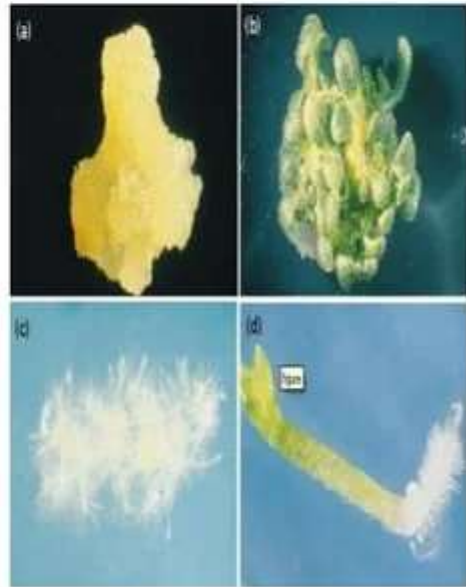
- Type of organogenesis by which only adventitious root formation takes place in the callus tissues.

Types of organogenesis

- In plant tissue culture, undifferentiated tissue is referred to as callus although a callus can contain meristematic nodules that may not be obvious to the naked eye but which never develop further unless suitable conditions are supplied.

Development of organised structures can follow one of three pathways:

- 1. shoot regeneration, based on a unipolar structure with a shoot apical meristem.
- 2. root regeneration, essentially a unipolar structure with a root apical meristem.
- 3. somatic embryogenesis in which there is a bipolar structure .



(a) disorganised
callus,

(b) Shoot
regeneration

(c) Root regeneration

(d) A single somatic embryo

(a) Indirect organogenesis

- Plant organ formation on callus tissues derived from explants.

plant growth regulators and differentiation

- The classic observations of Skoog and Miller that the direction of differentiation could be influenced by the ratio of the exogenously supplied growth regulators auxin and cytokinin.
- They observed in tobacco stem pith cultures that a high ratio of auxin to cytokinin led to initiation of roots whereas a low ratio led to development of shoots .
- Although there are many species for which this simple manipulation will not work, in general auxins e.g. IAA , NAA will stimulate regeneration of roots, and cytokinins e.g. BAP will promote regeneration of shoots or embryos.

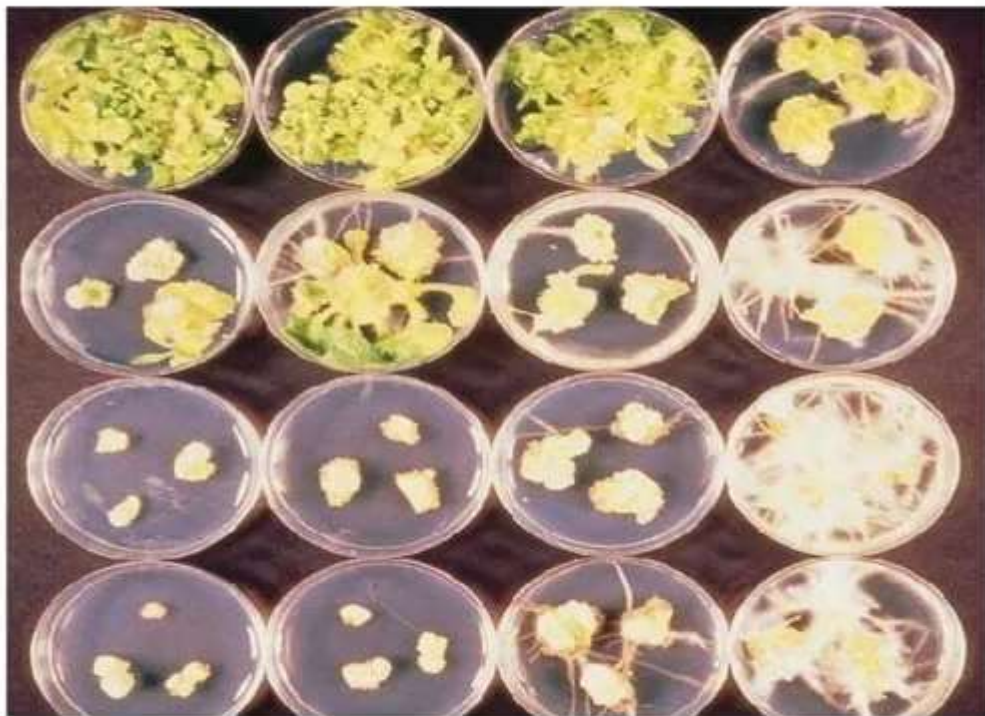


fig:-Tobacco leaf explants cultured on media with varying concentrations of an auxin (α -naphthaleneacetic acid; NAA) and a cytokinin (6-benzylaminopurine; BAP).

(b)Direct organogenesis

- ❑ Formation of organs directly on the surface of cultured intact explants. The process does not involve callus formation.
- **The role of growth regulators**
 - Direct organogenesis bypasses the need for a callus phase.
 - A good example is the formation of somatic embryos.
 - Most evidence suggests that direct embryogenesis proceeds from cells which were already embryogenically competent while they were part of the original, differentiated tissue.

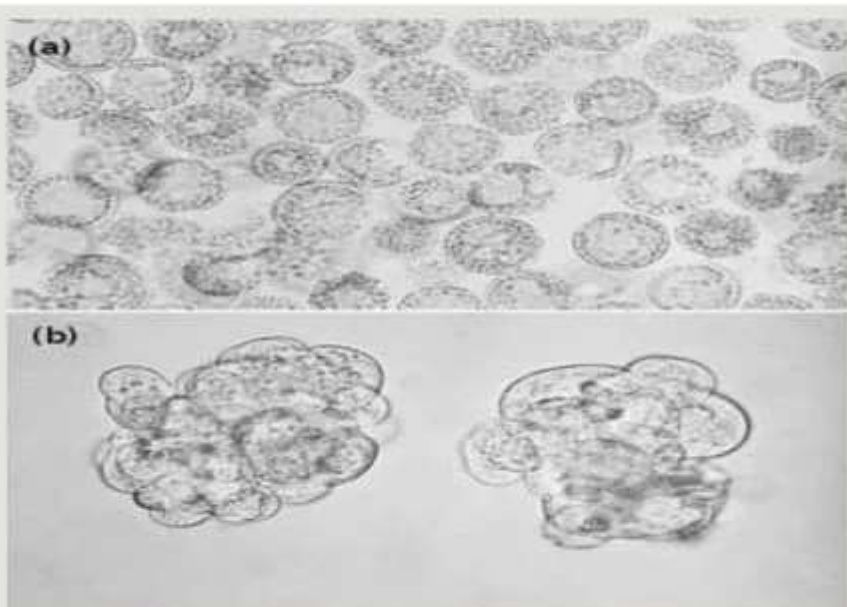
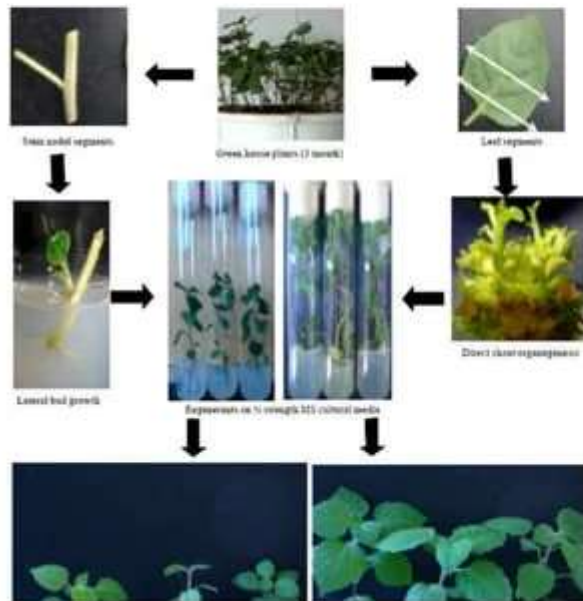


Figure 10.23 (a) Protoplasts of *Triticum aestivum* just after isolation from leaf mesophyll cell. (b) Cell division in two protoplasts of *Hyoscyamus muticus* has led to formation of two cell clusters or microcalluses.



direct organogenesis



(a) Indirect organogenesis

Factor Affecting the Organogenesis

In vitro organogenesis is controlled by a number of factors other than phytohormones such factors are discussed below:

1. **Size of Explant:** Organogenesis is generally dependent upon size of explant. The large explant consisting parenchyma, vascular tissues and cambium have greater regenerative ability than the smaller explant.
2. **Source of Explant:** The most suitable part of the plant for starting culture will depend on species. Leaves and leaf fragment of many plant species like Begonia, Solanum, Nicotiana, Crepis, etc have shown capacity to regenerate shoot buds.
3. **Age of the Explant:** Physiological age of explant is important for in vitro organogenesis. In Nicotiana species, regeneration of adventitious shoot is only noted if the leaf explant is collected from vegetative stage i.e. before flowering.

- 4. Seasonal Variation:** Bulb scales of *Lilium speciosum* regenerate bulblets freely in vitro when explant is taken during spring and autumn period of growth but same explants collected from summer or winter season does not produce any bulblets.
- 5. Oxygen Gradient:** In some cultures, shoot bud formation takes place when the gradient of available oxygen inside the culture vessel is reduced. But rooting requires a high oxygen gradient.
- 6. Quality and Intensity of Light:** The blue region of spectrum promotes shoot formation and red light induces rooting.
- 7. Temperature:** Most tissue cultures are grown successfully at temperatures around 25 °C. In bulbous species optimum temperature may be much lower of about 15-18 °C.

8. **Culture Medium: Medium** solidified with agar favours bud formation although there are some reports about the development of leaf shoot buds on culture grown in a liquid medium.
9. **PH of the Medium:** The PH of the culture medium is generally adjusted between 5.6 and 5.8 before sterilization. The pH may have a determining role in organogenesis.
10. **Ploidy Level:** Variation in chromosome number i.e. anuploidy, polyploidy, etc of plant cell in culture has been well documented. With the increase in chromosome instability there is a general decline in morphogenetic potentiality of callus tissue.
11. **Age of Culture:** A young culture frequently produces organs. But the Organogenic potential may decrease and ultimately disappear in old culture.

Application of organogenesis

- Plant tissue culture or organogenesis now has direct commercial applications as well as value in basic research into cell biology, genetics and biochemistry.
- micropropagation using meristem and shoot culture to produce large numbers of identical individuals.
- Screening programmes of cells, rather than plants for advantageous characters.
- large-scale growth of plant cells in liquid culture as a source of secondary products.
- Removal of viruses by propagation from meristematic tissues.

Conclusion

- Organogenesis is development pathway in which shoot or root have been induced to differentiation from a cell or cell clusters.
- In vitro plant regeneration by organogenesis usually involves induction and development of shoot from the explants tissue.

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