

Different tissue culture techniques

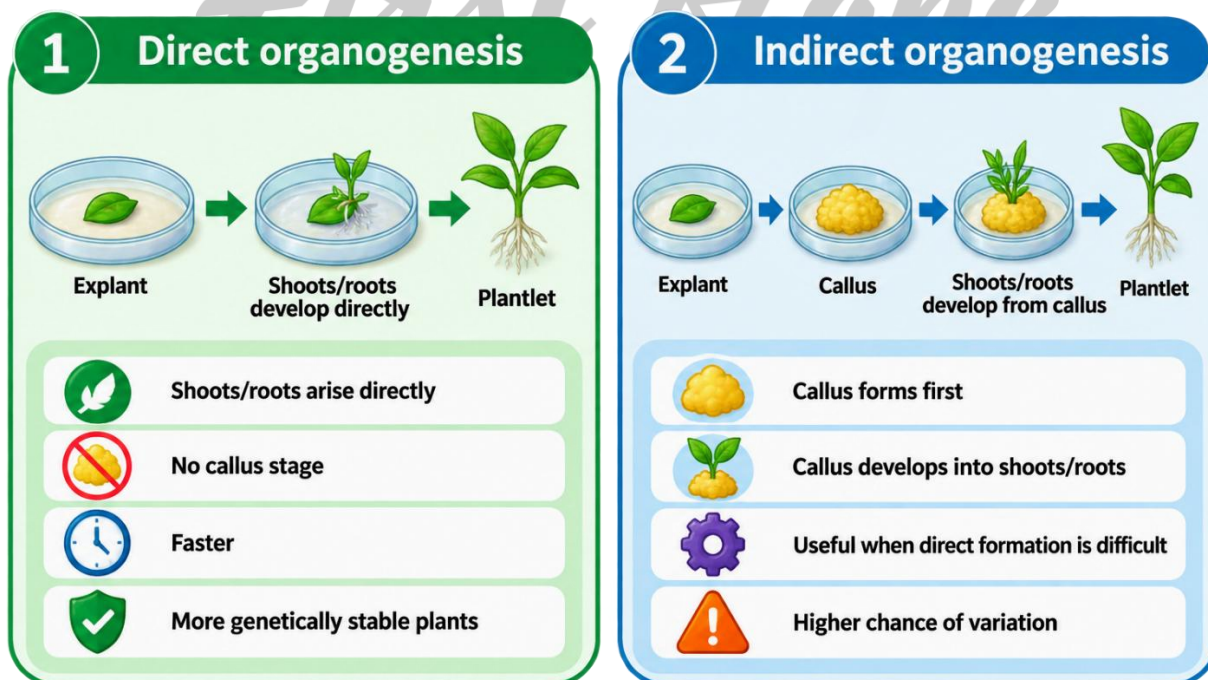
Organogenesis

- Organogenesis is a plant tissue culture method where new shoots and roots are formed from a small piece of plant (called an explant) grown on a nutrient medium in the lab.
 - Organo** = organ
 - Genesis** = formation
- So, it means formation of plant organs.

What is Needed

- Explant:** leaf, stem, root, cotyledon, etc.
- Nutrient medium:** water, minerals, vitamins, sugar, agar
- Growth hormones:**
 - Auxin** → helps root formation
 - Cytokinin** → helps shoot formation
- Conditions:** proper temperature, light, pH, and sterile environment

Types of Organogenesis



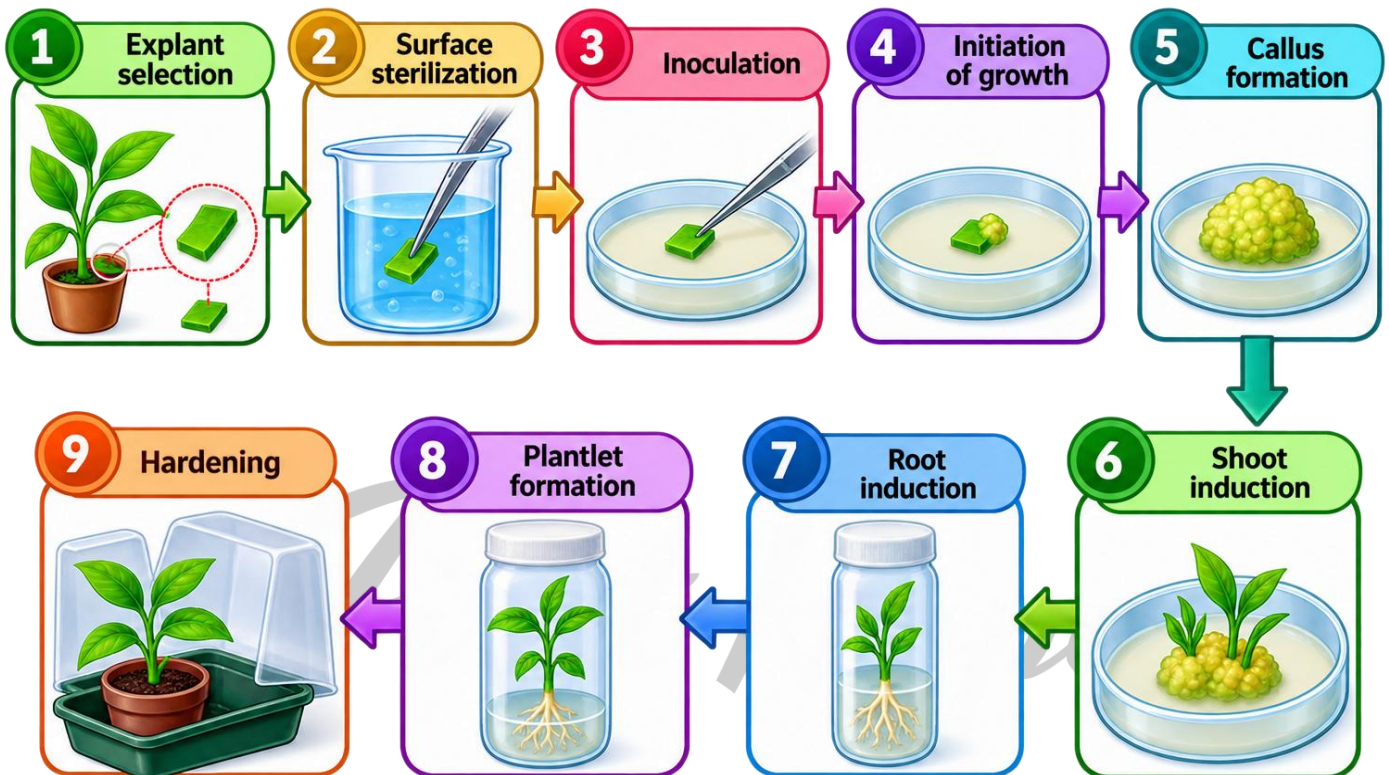
1. Direct organogenesis

- New shoots or roots arise directly from the explant
- No callus stage is formed
- It is faster
- It usually gives more genetically stable plants

2. Indirect organogenesis

- First, the explant forms an unorganized mass of cells called **callus**
- Later, this callus develops into shoots or roots
- It is useful when direct formation is difficult
- The chance of variation is often higher in this method

Stages of Organogenesis



A. Explant selection

- A suitable plant part is chosen
- Young and healthy tissues usually respond better

B. Surface sterilization

- The explant is cleaned to remove dust and microorganisms

C. Inoculation

- The sterilized explant is placed on the nutrient medium

D. Initiation of growth

- Cells begin to divide

E. Callus formation

- This happens in indirect organogenesis
- A soft mass of dividing cells appears

F. Shoot induction

- Shoots develop from the explant or from the callus

G. Root induction

- Roots are induced on a separate medium or sometimes on the same medium

H. Plantlet formation

- The shoot and root system together form a small complete plant

I. Hardening

- The plantlet is gradually adapted to outside conditions before field transfer

Role of Hormones in Organogenesis

- **High cytokinin and low auxin**
 - Usually leads to shoot formation
- **High auxin and low cytokinin**
 - Usually leads to root formation
- **Balanced amount of auxin and cytokinin**
 - Often leads to callus formation

Importance

- Produces many plants quickly
- Useful for rare or slow-growing plants
- Helps in research and plant breeding

Applications

- Micropropagation
- Producing disease-free plants
- Regeneration after genetic modification
- Conservation of rare plants
- Commercial plant production

Advantages

- Fast and efficient
- Many plants from small tissue
- Useful for difficult plants

Limitations

- Not all plants respond well

- Needs strict sterile conditions
- Hormone levels must be precise
- Indirect method may cause variation

Somatic Embryogenesis

- Somatic embryogenesis is a process where **normal plant body cells (somatic cells)** form **embryo-like structures** without fertilization, and these embryos can grow into complete plants.
- Unlike seeds, no male and female fusion is involved.

Key Idea

- **Somatic** = body cell
- **Embryogenesis** = embryo formation
- So, it means **formation of embryos from normal plant cells**

Features of Somatic Embryos

- Not formed by fertilization
- Produced in lab conditions
- Can grow into a full plant
- **Bipolar** → form both shoot and root

Types of Somatic Embryogenesis

1. Direct somatic embryogenesis

- Embryos arise directly from the explant
- No callus stage
- Less chance of genetic variation
- More suitable when true-to-type plants are needed

2. Indirect somatic embryogenesis

- First, callus is formed
- Then embryos arise from the callus
- More common in many plants
- Greater chance of variation

Steps in Somatic Embryogenesis

A. Selection of explant

- Young tissues are usually better
- Leaves, cotyledons, hypocotyls, immature embryos, and nucellus may be used

B. Culture initiation

- Explant is placed on nutrient medium

C. Induction of embryogenic cells

- Certain cells become capable of forming embryos
- Usually induced by auxins such as 2,4-D

D. Formation of proembryogenic masses

- Small clusters of embryogenic cells are formed

E. Development of somatic embryos

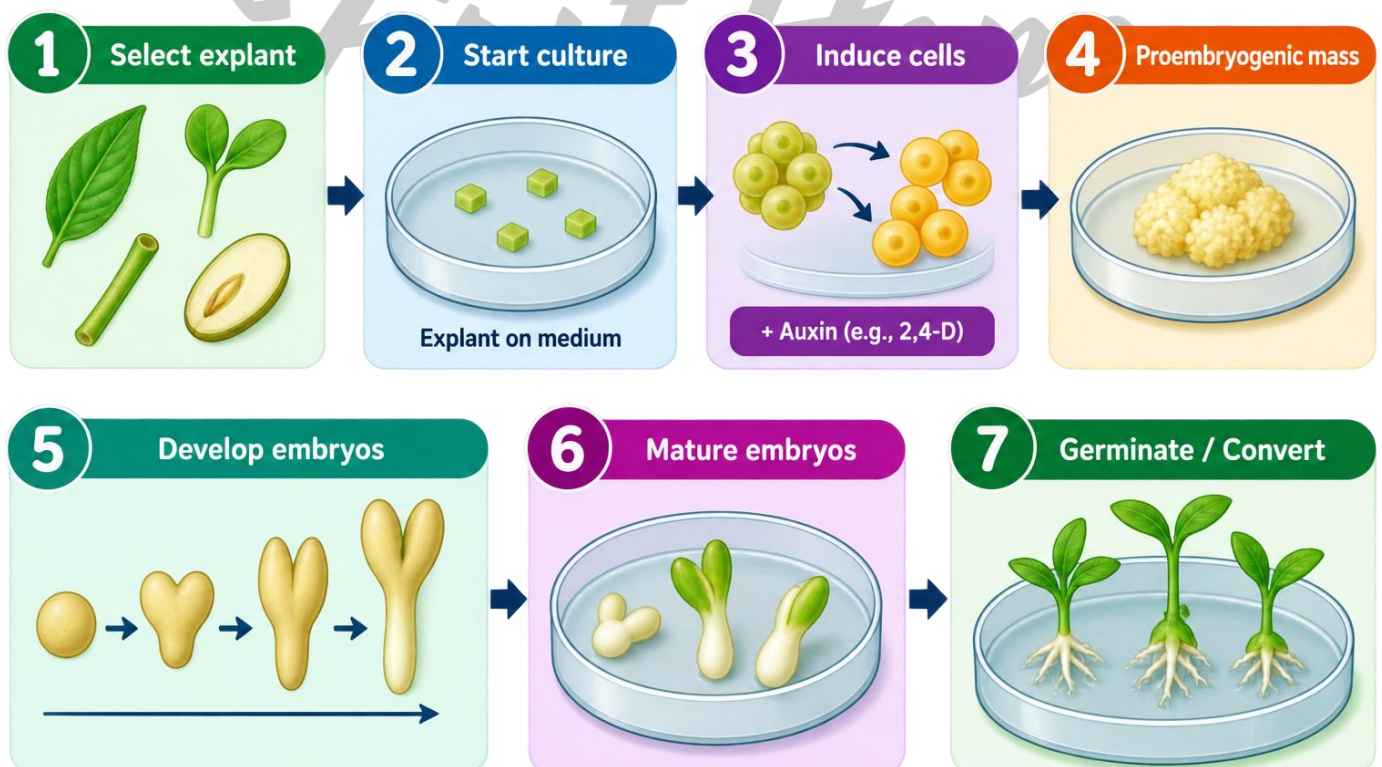
- These pass through several stages

F. Maturation of embryos

- Embryos become fully developed

G. Germination or conversion

- Embryos are transferred to another medium where they grow into plantlets



Stages of Somatic Embryo Development

These stages are similar to the stages of zygotic embryo development:

1. Globular stage

- The embryo is round in shape

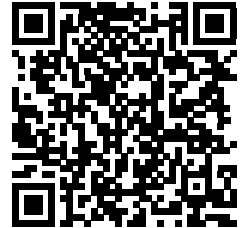


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