

# Introduction To Embryo Culture:

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A decorative graphic element consisting of several horizontal lines of varying lengths and colors (teal, light blue, and white) extending from the right side of the slide.

# What is embryo?

- A seed plant embryo is part of a seed, consisting of precursor tissues for the leaves, stem and root as well as one or more cotyledons.
- The young sporophyte of a seed plant usually comprising a rudimentary plant with plumule, radicle, and cotyledons

# Embryo culture??

- Embryo culture similarly can mean the growth of any species, during its embryonic stage, in an artificial medium in order to enhance its likelihood of survival.
- Embryo culture can also mean the use of an artificial medium for embryonic growth during in vitro fertilization, which can last two to five days, after which the embryo is transplanted into a surrogate mother.

# Embryo Rescue:

- Embryo rescue is one of the earliest and successful forms of in-vitro culture techniques that is used to assist in the development of plant embryos that might not survive to become viable plants.
- Embryo rescue plays an important role in modern plant breeding and ornamental plant crop hybrids.
- The most widely used embryo rescue procedure is referred to as embryo culture, and involves excising plant embryos and placing them onto media culture

# How can we do embryo culture?

- For embryo culture, embryos are excised from immature seeds and introduced in a medium.
- Providing aseptic and sterile area.

- The culture plates or culture tubes with excised embryos are transferred to a culture room maintained at a suitable:
  - Temperature.
  - Photoperiod.
  - Humidity.
- The frequency of excised embryos that gives rise to seedlings generally varies greatly.

- The hybrids raised through culture have been utilized for:
- Phylogenetic studies.
- Genome analysis. Transfer of useful agronomic traits from wild genera to the cultivated crops.
- To raise synthetic crops.

# Advantages of Embryo Culture:

- Recovery of distant hybrids.
- Recovery of haploid plants from Interspecific crosses.
- Propagation of orchids.
- Shortening the breeding cycle
- Overcoming dormancy.
- In addition ovule and ovary can also be cultured



# Importance Of Embryo Rescue And Embryo Culture

- This technique is used to develop plants that are viable.
- Embryo rescue plays an important role in modern plant breeding.
- Allowing the development of many interspecific and intergeneric food and ornamental plant crop hybrids.
- This technique nurtures the immature or weak embryo, thus allowing it the chance to survive.

- Tissue culture techniques in combination with molecular techniques have been successfully used to incorporate specific traits through gene transfer
- In vitro techniques for the culture of protoplasts, embryos have been used to create new genetic variations in the breeding line.

# Factors.

Embryos are manually excised and placed immediately onto a culture media that provides proper nutrients to support survival and growth:

- **Time** of culture.
- **Composition** of the medium.
  - Two main types of basal media are the most commonly used for embryo rescue studies, i.e. Murashige and Skoog medium (MS) and Gamborg's B-5 media (Bridgen, 1994)
  - The composition of the media will vary in terms of the concentrations of media supplements required this will generally depend on the stage of development of the embryo

- Young embryos would require a complex medium with high sucrose concentrations, while more mature embryos can usually develop on a simple medium with low levels of sucrose.

#### □ **Temperature and light:**

The temperature and light requirement is generally species specific and thus its usually regulated to be the within the same temperature requirement as that of its parent with embryos of cool-season crops requiring lower temperatures than those of warm-season crops.

- Embryos sometimes grow best when maintained in darkness for the first 1 to 2 weeks of culture and then transferred to light to allow chlorophyll formation.
- The optimum temperature depends on plant species, but normally a high range of 25 to 30C is used.
- The growth conditions of the mother plant are also a consideration in embryo culture. The endosperm and the cotyledons will develop more if the mother plant is grown under well-controlled conditions; embryo growth will consequently be promoted.

# Techniques & Handling:

- Embryos are located in the sterile environment of the ovule and surface sterilization of embryos is not necessary.
- Embryos are removed aseptically from the surrounding tissues.
- Embryo is often well-protected by surrounding tissues, harsh procedures may be used in surface disinfection.
- Direct disinfection of embryos is needed if seedcoats are cracked or if endophytic pathogens exist inside the seedcoats, as with fescue (*Festuca* spp. L.), corn (*Zea mays* L.), and dogwood

# Techniques & Handling:

- Small embryos require the use of micro dissecting tools and a dissecting microscope to excise without injury.
- The process of excising immature embryos varies with species.
- However, many times an incision can be made at the micropylar end of the young ovule and pressure applied at the opposite end to force the embryo out through the opening.

# Techniques & Handling:

- When heart-stage and younger embryos are excised, it is important to keep the suspensors intact.
- The nutritional requirements of young embryos vary greatly and the chances of damaging the embryos are great. In such situations, it may be possible to rescue embryos by ovary or ovule culture methods.
- Ovaries are excised after pollination and the calyx, corolla, and stamens are removed.



# Techniques & Handling:

- Depending on the organ cultured, it may be referred to as :
  - Ovule Culture.
  - Ovary Culture.
  
- It is technically difficult to isolate the tiny intact embryos.

# Techniques & Handling

- **Ovule Culture:**

Ovule embryo culture is a modified technique of embryo rescue where embryos are cultured while still inside their ovules to prevent damaging them during the excision process.

- **Ovary Culture:**

Ovary or pod culture, on the other hand employs the use of an entire ovary into culture. The ovary is surface-sterilized and cultured and inserted into the nutrient medium

# Applications:

- Embryo culture is now days used in alot of fields such as agriculture ,Industry etc .
- Its use is made more specific in the area of agriculture foccussing mostly on plant tissue culture.
- Embryo culture is being routinely used in some crop improvement programmes.

# Applications:

- Shortening breeding cycles and overcoming dormancy thus reducing the time for the next generations which may be grown two or three weeks earlier by embryo culture than from seeds.
- Example of Iris whose breeding cycle is shortened to less than one year from two years through this .

# Applications:

- Seed sterility is another problem faced in the field of crop production .
- Early ripening fruit cultivars have seeds that do not germinate as embryo is immature.
- Thus using these seedlings are raised from sterile seeds and the embryos are recovered.

# Applications:

- Production of Monoploids helps in achieving the desired trait in less time as normally several generations of testing required before desired trait achieved in selective breeding .
- Plants derived from tissue culture techniques are monoploid and can be treated with chemicals to double chromosome number.

# Applications:

- Micropropagation is done to rapidly produce large number of progeny plants by multiplying stock plant material .
- This is done to achieve virus free stock .
- To multiply plants whose multiplication rate is too slow .
- For germplasm conservation
- For genetic transformations

# Embryo culture:

- **Embryo:**

A mature zygote of animals while Fertile part of seeds in plants

**Embryo Rescue:**

Embryo rescue is the process when plant breeders rescue inherently weak, immature or hybrid embryos to prevent degeneration.

Common in lily hybridizing to create new interspecific hybrids between the various lily groups (such as Asiatic, Oriental, Trumpet, etc).



# Embryo culture

- Embryo culture.
- Artificial growth of individual during embryonic stage is known as embryo culture
- In vitro technique
- Embryo rescue
- Embryo rescue is synonym of embryo culture
- It is also in vitro preservation and culturing of embryo.

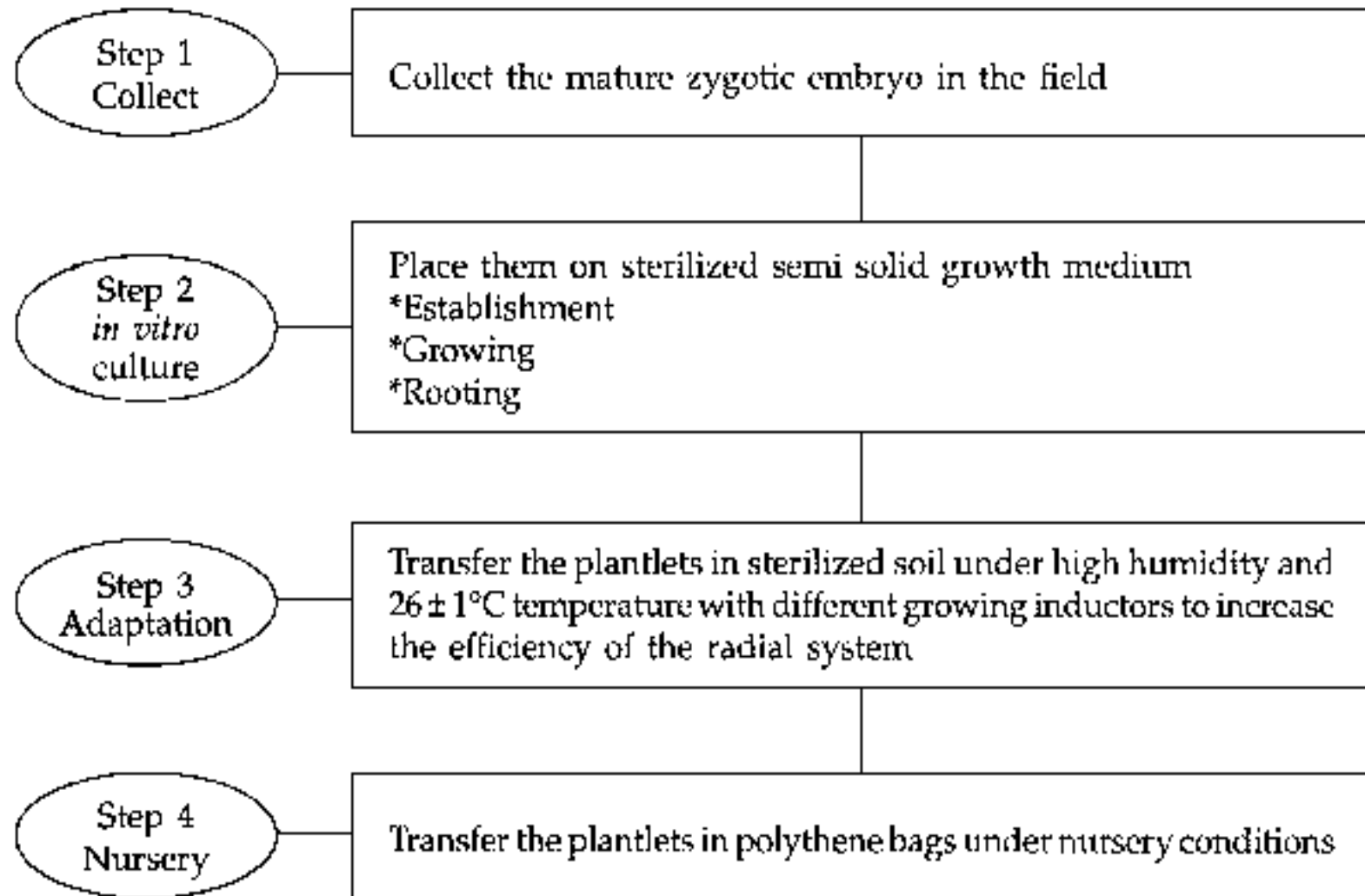
# Purpose of embryo culture /rescue

- To overcome seed dormancy & immaturity
- To enhance breeding
- To rescue genotypes (preservation).
- To avoid abortion
- Crop improvement
- Cryopreservation( Cryo conservation).

# Types Of Embryo Culture:

- Ovary Embryo Culture.
- Ovule Embryo Culture.

# Stages Of Culture:



<http://www.biologydiscussion.com/plants/plant-tissue-culture-environmental-condition-methods-types-and-application/1359>