




EXPLANT MANAGEMENT

Online Training Course on
Horticulture Seed Propagation with Tissue Culture

Indonesian Center for Agricultural Training Lembang
The Agency for Agricultural Human Resources and Development
Ministry of Agriculture, Republic of Indonesia

Non-Aligned-Movement Centre for South-South Technical Cooperation

ABD. ROHIM
LECTURER
ICAT LEMBANG

elm_kalop@yahoo.co.id

Contact me via

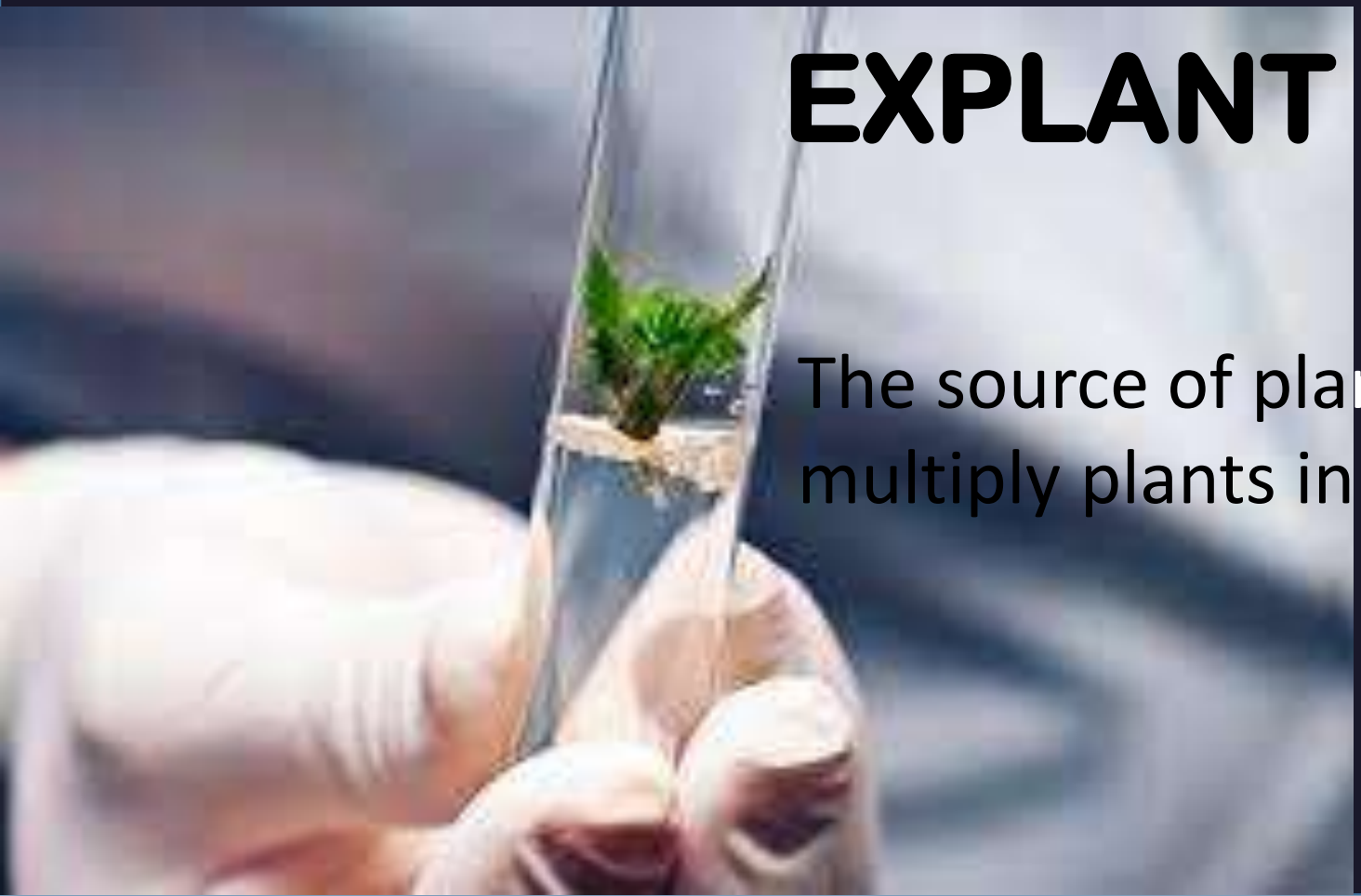
+62 813 77 8181 99

+62 813 77 8181 99

Abdurrohim El-Mukromy

abu_mizyfa

@mizyfa



EXPLANT ?

The source of planting material used to multiply plants in tissue culture technology.

4 keys to success in explant management:

1) Where did the explants come from?
2) How do you remove sources of contaminants (fungi and bacteria)?
3) What culture medium do you use to grow the explants?
4) Do you have the skills to grow explants in culture bottles?

Good Explants ?

"source of planting material that **meets the requirements**, to **grow** and **develop properly**"

Criteria for good explant:

1) Originates from the “mother plant” which is known where it comes from.
2) Originates from the “mother plant” that grows normally and healthy.
3) Originates from the “mother plant” that grows in a healthy environment.

Note: Explants can be taken from plant tissue that is actively dividing (meristematic).

Explant Sterilization

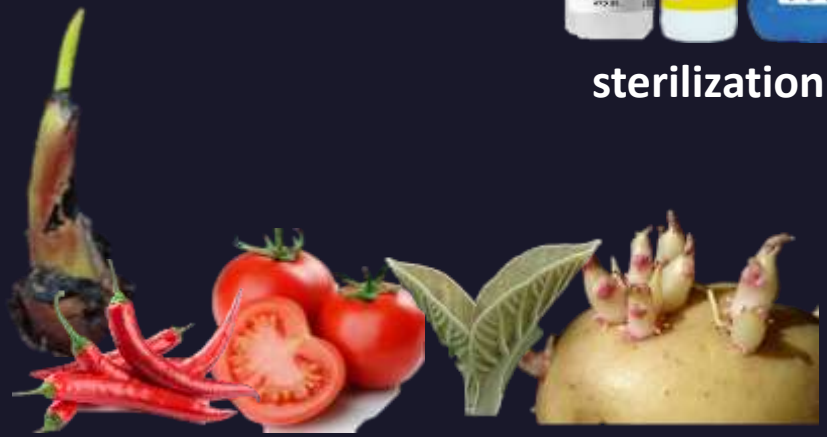
?

"to remove sources of contaminants (fungi and bacteria)"

4 Important Points on Sterilizing Explants

sterilization material

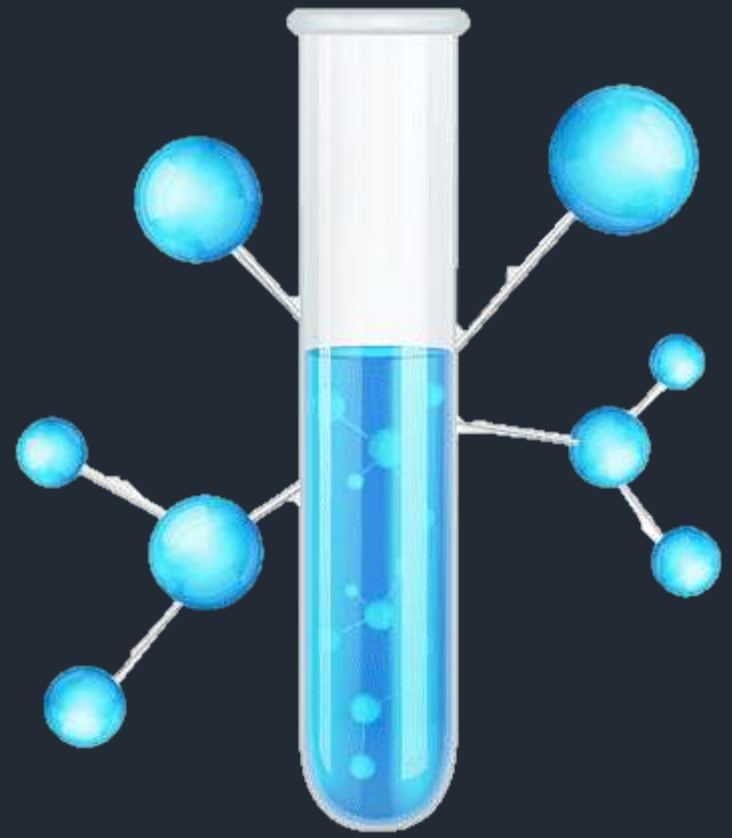
sterilization duration



explant condition

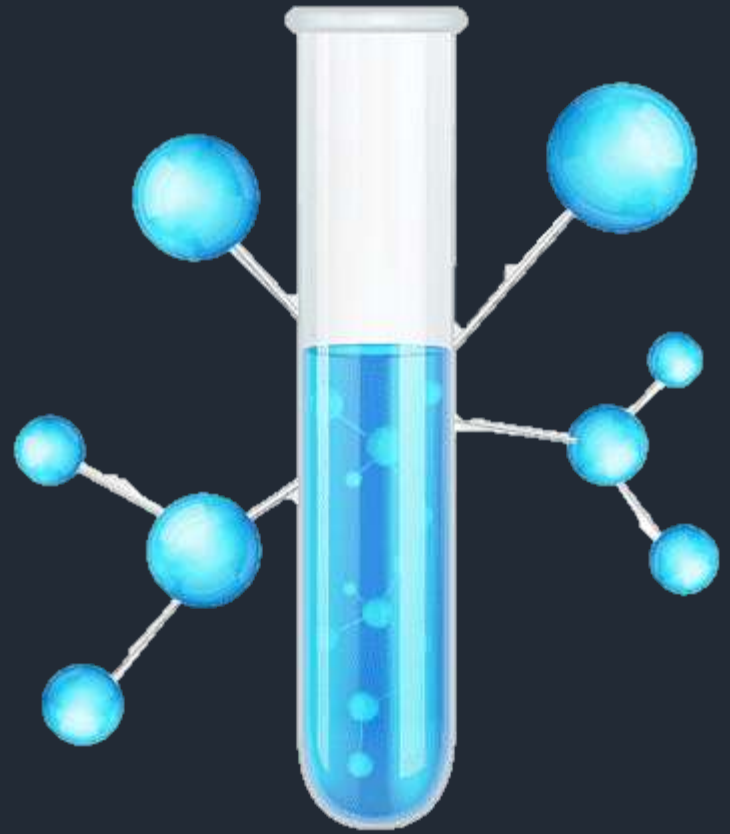
the concentration of the sterilizer

- ✓ The basic principle of explant sterilization is to eliminate the source of contaminants as small as possible to eliminate the interference of the sterilizing material to the explant tissue.
- ✓ You can start the test with a single sterilizing material, low concentration, and short time. And you can periodically upgrade if there is still contamination of the explants.



- ✓ There is no standard for the amount of material, concentration, and explant sterilization time. You can try and learn.
- ✓ As a reference, you can follow these guidelines:

  “Explants with a high possible source of contaminants, usually from the soil, such as banana shoots or potato tubers, you can use a higher material, concentration and sterilization time than explants with a low possible source of contaminants, usually those not in direct contact with the soil, such as chili or tomato.”



Some of the alternative sterilization materials that can be used are:

clean the dirt on the explant

clean the dirt on the explant

Some of the alternative sterilization materials that can be used are:

Antiseptic that functions to inhibit the growth of viruses and kill bacteria and fungi.

A type of disinfectant that can kill bacteria, fungi and viruses

Some of the alternative sterilization materials that can be used are:

a sterilizing material that can control bacteria

a sterilizing agent that can control fungi

a type of sterilizing material that can help open the explant tissue

Some of the alternative sterilization materials that can be used are:

a type of sterilizing material that can help open the explant tissue

a type of disinfectant that can kill bacteria, fungi, and viruses

$\sum$ %

Sterilization Materials

$<$

How to treat bacterial contamination with silver nanoparticles?

- The principle of explant sterilization is to understand the characteristics of antiseptic substances related to the characteristics of explants.
- Silver nanoparticles were only effective in killing gram-positive bacteria (on the explant surface).
- Sources of contaminants often come from indophytic microbes (from inside explants).
- Microbial handling using heavy metals (silver nanoparticles) in explants can damage explant tissues and genetic mutations.

How to treat bacterial contamination with silver nanoparticles?

- Antiseptic is a solution to kill bacteria without having to kill plant tissue. Perform a combination of treatments: type of material, amount of material, concentration and duration of the sterilization process.
- What if the microbes are not killed with an antiseptic/disinfectant?
  - ✓ Quarantine the explant broodstock.
  - ✓ Use antibiotic treatment for 3 months.
  - ✓ Take explants shoots are 5 days old (less than 1 week), the hope is not infected with microbes.
  - ✓ Take the meristem (0.5 mm). If it is difficult, take the explant as small as possible (5 mm).

What are the effects of silver nanoparticle treatment on fungal contamination control?

- Silver nanoparticles are only effective at killing fungus on the surface of the explants.
- The fungus often infects the inside of the explant.
- Still needed an antiseptic to kill the fungus in the explant.

How to plant explants correctly ?

1) Make sure the tools and materials for planting the explants are **sterilized**.

How to plant explants correctly ?

2) Make sure the explants are planted in the right place, such as **Laminar Air Flow**.

How to plant explants correctly ?

3) Make sure before planting explants, the culture media **is not contaminated with fungi and bacteria.**

How to plant explants correctly ?

4) Make sure you do the planting method correctly:
   - ➢ Burn the planting tool before use.
   - ➢ Take the right part of the explant.
   - ➢ Put the explant into the bottle quickly and precisely.

How to plant explants correctly ?

5) Make sure the explants are reproduced (subcultured), if ...
   - ➢ the culture media has run out.
   - ➢ for certain growth conditions on the explants.
   - ➢ the explants were fully grown in the culture bottle.

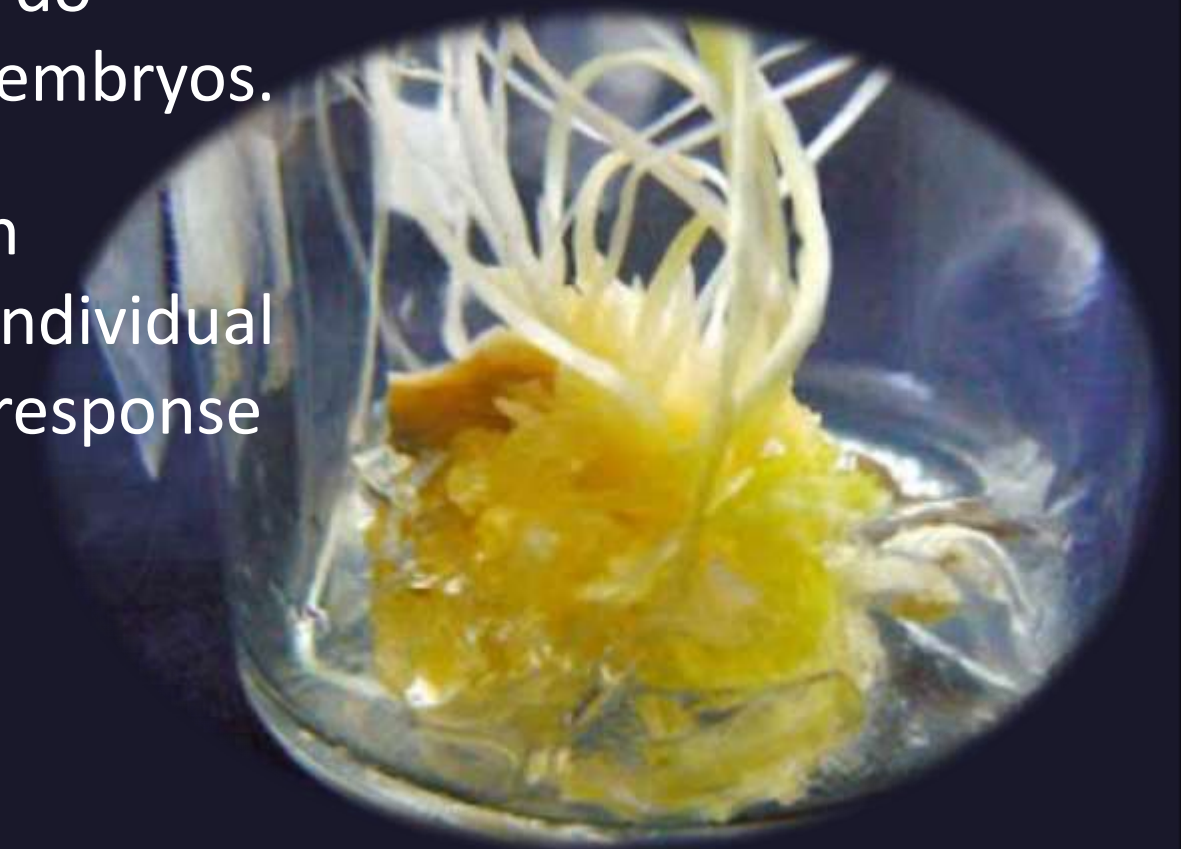
How to plant explants correctly

6) Make sure the explants that have been planted are stored in the incubation room with ideal conditions. Check regularly:

- ✓ Incubation room temperature
- ✓ Incubation room humidity
- ✓ Incubation room light
- ✓ Explant condition

How may somaclonal variation occur in tissue culture practice?

- Somaclonal variation occurs when we do multiplication using callus or somatic embryos.
- The callus cell or somatic embryo is an independent individual, so that each individual has a different reaction and action in response to the given stimulus.



How do plants react to various LED wavelengths?

- Plants need light radiation according to their needs.
- Plants have a variety of different leaf dyes, which determine the variety of light spectrum needed.
- Vegetative growth requires an infra-red light spectrum, and the generative phase is more dominant in the ultra-violet spectrum.
- Sunlight is white light with a complete spectrum of light, so that it fulfills all kinds of leaf dyes of various plants.
- Thus, attention is focused on the intensity of the light spectrum and the duration of irradiation.

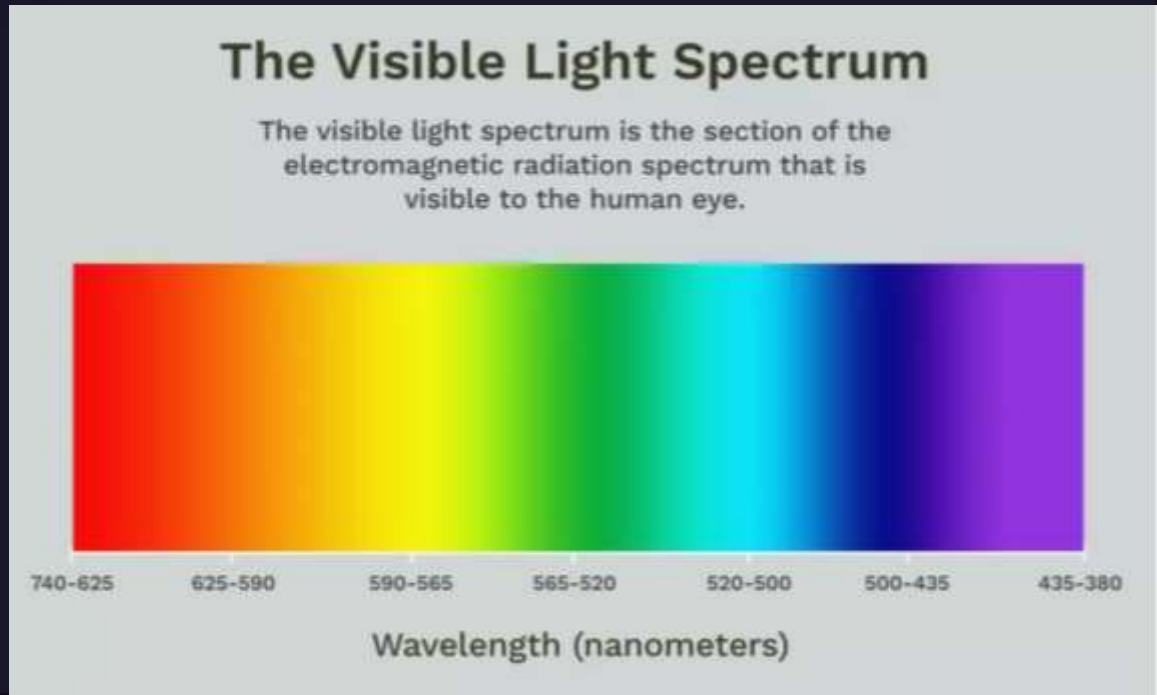
How may narrow-band illumination treatment be effective to regulate plant growth?

- Specific spectrum irradiation treatment is needed to optimize the spectrum requirements of light and the duration of irradiation, so that the photosynthesis process is optimal.
- Infrared spectrum is a spectrum used for vegetative growth.
- The provision of narrow band illumination is very efficient to increase the required light requirements, so that vegetative growth can be more optimal.

How does the supply of blue light and temperature become crucial to the efficiency of the plant?

- Different colors of light help plants to achieve different goals equally well. For example, blue light helps promote vegetative leaf growth. Red light combined with blue makes plants flower. Cool color fluorescence is excellent for the growth of plants grown indoors.
- Blue light and red light are the most effective and green light is the least effective in carrying out photosynthesis.

(Loveless, A.R. 1991. Principles of Plant Biology for the Tropics. Logman Group Limited.)



How does the supply of blue light and temperature become crucial to the efficiency of the plant?

- The rate of photosynthesis in tropical plants increases from a minimum temperature of 5ºC to a temperature of 35ºC, above this temperature range the rate of photosynthesis decreases.
- Temperatures above 35ºC cause temporary or permanent damage to the protoplasm resulting in decreased the speed of photosynthesis, the higher the temperature the faster the rate of photosynthesis decreases.

(Loveless, A.R. 1991. Principles of Plant Biology for the Tropics. Logman Group Limited.)

How may tissue culture help to satisfy human demands of horticulture crops in developing countries?

- Tissue culture technology in Indonesia, in an effort to meet the needs of horticultural plant products is focused on commercial commodities and seed propagation requires large quantities and a fast time which cannot be met through conventional seed propagation.
- For example, commodity potatoes, certain varieties of bananas, orchids, etc.
- For the case of chili and tomatoes, we can still handle it conventionally.

How to strengthen tissue culture research capacity?
How to employ tissue culture?

- Develop commercial commodities based on local wisdom.
- Develop applicable research that produces agricultural products are needed by many people.
- Provision of infrastructure and human resources according to the needs of tissue culture research.

Thank You