# Biotechnology: Through In Vitro Cultutre Of Plants And Its Applications In Agriculture And Anvironmental Conservation

*by* Sutini Sutini

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# BIOTECHNOLOGY: THROUGH *IN VITRO* CULTURE OF PLANTS AND ITS APPLICATIONS IN AGRICULTURE AND ENVIRONMENTAL CONSERVATION

Sutini [1*],Widiwurjani[1],N Augustien[1],Guniarti[1],DU Pribadi [1],A P Djoko [2],WMuslihatin [3].

*1 Department of Agrotechnology, Faculty of Agriculture, UPN "Veteran" East Java, Surabaya, Indonesia.*

*2 Department of Pharmaceutical Chemistry, Faculty of Pharmacy, Universitas Airlangga, Surabaya, Indonesia.*

*3 Department of Biology, Faculty of Mathematics and Natural Sciences, Sepuluh Nopember Institute of Technology, Surabaya, Indonesia.*

**Corresponding Author: tien.basuki@gmail.com*

**Abstract:** *In vitro* culture is an important aspect of biotechnology which concerns plants. This *in vitro* culture technique is based on the power of cells or plants tissue to produce callus biomass or to become multicellular plants derived from differentiated single cells. The problem of *in vitro* culture, especially in developing countries is the lack of knowledge about improving the quality and quantity of *in vitro* culture of plants results. To overcome the problem in the formation of callus or plant biomass is through the provision of precursors, elicitors, or the addition of microorganisms. The aim of this scientific work is to explore the role of biotechnology of *in vitro* culture of plants in agriculture and environmental conservation. The method used is by reviewing the literature related to the biotechnology of *in vitro* culture techniques of plants and the application of the results. The results obtained from this study are profiles of several callus and plant biomass products from the biotechnology of *in vitro* culture techniques of plants that have a role in agriculture and the environment.

**Keywords**: **Keywords:** Agriculture, Biotechnology, Environment, *In vitro* culture

## I. INTRODUCTION

Biotechnology products through *in vitro* culture of plants including secondary metabolites in the form of callus biomass and plants derived from single cells can be produced already. The result from this *in vitro* culture that is the secondary metabolite biomass products can be applied in various fields. This callus biomass is a basic asset that can be developed into various forms according to research objectives. The research that has been conducted by Sutini et.al (2020) has succeeded in producing callus containing secondary metabolites of epicatechin by inducing callus with the addition of phosphorus metal elicitor. Multicellular plants originating from single cells have been produced by Gladfelter et.al (2020). Gladfelter et.al have developed an in vitro Franklinia alatamaha plant propagation system to increase the availability of rare or endangered Franklinia alatamaha trees for commercial and restorative horticulture. Biotechnology through *in vitro* culture of plants plays an important role in the protection, use and diversification of plants, especially in plant conservation. Therefore, biotechnology through *in vitro* culture of plant is an indispensable tool not only for sustainable management of plant genes but also for the creation of new diversity resources.

The problem that exists in *in vitro* culture, especially in developing countries, is the lack of knowledge about increasing the quality and quantity of *in vitro* culture of plant results. To overcome this problem, the formation process of callus or plant biomass is through the provision of precursors, elicitors, or the addition of microorganisms. The aim of this scientific work is to explore the role of the biotechnology of *in vitro* culture of plants in agriculture and environmental conservation.

## II. METHODS

The method used in writing this paper is to examine the literature related to the biotechnology of *in vitro* culture of plant techniques and the application of the results. The literature review that discussed is *in vitro* culture methods, *in vitro* culture applications in agriculture, and *in vitro* culture applications in environmental conservation.

### *In vitro* Culture Methods

The *in vitro* culture of plant method is a technique that is carried out completely aseptically with plant subjects starting from plant organs and cells in a fully controlled environment. The steps taken in the *in vitro* method include: disinfection and start of axenic culture, initiation of culture, multiplication, elongation and promotion of shoot and root development, acclimatization and hardening (Khayat, 2012).

Disinfection and start of axenic culture is an absolute requirement so that in vitro culture conditions run on target. According to Monokesh et.al (2013), disinfectants are applied to the plant materials, equipment used and personal work, including lab clothes worn.

Initiation of culture is the initial stage by preparing the media, usually using MS media (Murashige and Skoog, 1962) which is enriched with growth regulators auxin and cytokinins with the addition of anti-oxidants of 200 mg $L^{-1}$ to prevent auxin and cytokinin degradation. Then the culture was being incubated in a dark room.

Multiplication is the multiplication process of callus biomass in the same medium, such as during culture initiation. In this multiplication step, callus biomass has been formed. Callus generally has a fragile structure, large, when observed under a microscope; the cells are differentiated, but not organized. This callus can be increased its secondary metabolites by adding the iron oxide elicitors (Moharrami 2017). This callus can be extracted to be applied in various activities such as applications in the health sector for medicinal substances.

Elongation and promotion of shoot and root development, is a step to grow embryos (Shirin et.al 2020) that can be in the form of roots and shoots by increasing the ratio of the same auxin and cytokinins, but each plant for this elongation requires specific auxin and cytokinin concentrations.

Acclimatization and hardening are steps to make culture as newborn babies that require adaptation treatment from the laboratory environment to the ex vitro environment or can be placed in a greenhouse. When it has adapted in the greenhouse, it can be slowly planted on land that is given a lid for a while so that small plants are ready to be released with care for growth and development (Uyen et.al 2013).

### *In vitro* Culture Applications in Agriculture

The applications of *in vitro* culture in agriculture have been widely researched which are useful for herbicides, allelochemistry, nurseries, embryogenesis, fungicides and phytotoxics, which are shown in Table 1.

*Tabel 1. In vitro Culture Applications in Agriculture*

| ***In vitro* culture Form** | **Bioactive Function** | **Secondary Metabolites** | **Origin of Plants** | **References** |
|---|---|---|---|---|
| **Solid** | herbicide | epicatechin | Camellia sinensis | Maria John et al. ( 2009). |
| **protoplast co-culture** | allelochemistry | canavanine | Vicia villosa Roth | Sasamoto *et al*. (2019) |
| **Solid** | Phytotoxic | catechin | Camellia sinensis | Inderjit et al. (2008). |
| **Solid** | Nursery | lignin | *Dyera lowii* | Rodinah *et al*. (2016) |
| **Solid** | Fungicide | *Azadirachta indica* | *Phomopsis theae* | Linner et. al. (2017) |
| **Suspension** | embriogenensis | Gallic acid | Camellia sinensis | Seran et al. (2008). |
| **Solid** | biodiesel plant | Lectin, saponin | Jatropha curcas, | Kumar et. al. (2011) |

***In vitro* Culture Applications in Environmental Conservation**

The application of *in vitro* culture in environmental conservation includes can be used for mass propagation, producing plants that are tolerant of polluted soil conditions, plants that are tolerant of climate change which are shown in Table 2.

*Table 2. In vitro Culture Applications in Environmental Conservation*

| **Types of *in vitro* cultures** | **Types of Plants** | **Culture morphology** | **Types of Conservation** | **References** |
|---|---|---|---|---|
| **Suspension** | horticulture, forestry - breeding | somatic embryo | Conservation of Endangered Species | Ojo et. al. (2018) |
| **Suspension** | *Crocus* | embryogenesis | preservation of germplasm | Freytag et. al. (2017) |

| | | | | |
|---|---|---|---|---|
| **Solid** | *Rhanterium epapposum* | organogenesis | city landscape | Sudhe et. al. (2003) |
| **Suspension** | *Ruscus* | Embryogenesis & biosynthesis | Conservation & biomass | Ivanova et. al. (2015) |
| **Suspension** | *Brassicaceae, Poaceae, Biofuel species* | freeze storage and DNA protection | conservation | Demir A. (2015). |
| **Suspension** | *Eucalyptus perriniana* | Embryogenesis & biosynthesis | Phytoremediation | Pauline MD. (2009) |
| **Solid** | *L. anagyroides* | Micro propagation | landscaping & pharmaceutical. | Timofeeva et. al. (2016) |
| **Suspension** | *Podophyllum hexandrum* | Micro propagation | selected medicinal herbs | Shyamal et. al.(2016) |
| **Suspension** | *Parkia timoriana* | Micro propagation | inventorisation- food security | Thangjam R, (2016). |

## III. RESULTS AND DISCUSSION

*In vitro* culture method is an effective and efficient technique because the process and aseptic are controlled, using a small area of land, independent of climate and weather. *In vitro* culture method in this discussion was adapted from the Mathu method (2013) with a range of micro propagation, protoplasts, synthetic seeds, haploids, secondary products. The micro propagation method in Table 2 that has been carried out by Shyamal et. al. (2016) conducted for selected herbal plant materials from Podophyllum hexandrum plants, in addition to selecting them for conservation so they do not become extinct. Meanwhile, synthetic seeds aim to manufacture artificial seeds by including the encapsulation of non-endospermic seeds for species with high commercial value. Catechin secondary metabolites have also been produced by Sutini et.al. (2020) through callus *in vitro* culture of the Camellia sinensis L.

*In vitro* culture applications in agriculture have been widely applied, including for allelochemicals, herbicides, anti-pests, phytotoxics and biodiesel. The use of biodiesel from *in vitro* culture of the Jatropha curcas plant, which is mentioned in table 1, has been implemented with a growth rate of up to 90% (Kumar et. al. 2011). *In vitro* culture of J. curcas plants by direct organogenesis from J. curcas petiole explants using Murashige and Skoog (MS) media supplemented with various thidiazuron (TDZ) concentrations.

The application of *in vitro* culture in environmental conservation has been mentioned in Table 2. Other researchers, namely Ashok et. al. (2015) succeeded in producing sugarcane plants that were tolerant of soil conditions containing salt by the radiation method with gamma ray mutagens.

## IV. CONCLUSION

*In vitro* culture method is a technique that has prospects to be applied in various fields including agriculture and environmental conservation. Biomass from *in vitro* culture is in the form of callus and embryos as well as multi-cellular plants that are ready to be planted in the land for conservation.

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