NILAMANI MAHAVIDYALAYA

RUPSA,BALASORE

A
PROJECT REPORT OF
"DEPARTMENT OF BOTANY"

6TH Semester +3 Examination 2022

Guided By:-

Dr.Ardhendu Kumar Dash

Lect. In Botany

Submitted By:-

Group 2 Student

+3 3rd Year Botany(H)

...e of the Project-Plant Tissue Culture of Tobacco Plant(Nicotiana Tabacum)

A project report submitted in partial fulfillment of the degree of bachelor of Science by

The Group -2 Students.

Group -2 Students

Name	Roll No	Regd.No
Jyoti Rekha Behera	5208B19010	02698/19
Parvati Sahoo	5208B19011	02699/19
Rashmi Ranjan Panda	5208B19013	02701/19
Saijiban Giri	5208B19014	02702/19
Subhalaxmi Dey	5208B19015	02703/19
Subhasmita Hota	5208B19017	02705/19
Suman Roul	5208B19019	02707/19
Tapan Kumar Patra	5208B19020	02708/19

Under the Guidance of

(Dr. Ardhendu Kumar Dash)

Nilamani Mahavidyalaya , Rupsa,Balasore

Nilamani Mahavidyalaya , Rupsa,Balasore

DECLARATION

We Group-2 Students do hereby certify that the project report entitled"**Plant Tissue Culture of Tobacco Plant(Nicotiana Tabacum)**"being submitted to nilamani mahavidyalaya, rupsa,balasore,Odisha for the award of bachelor of science is an original piece of work done by us and the same has not been submitted elsewhere for any other academic degree or diploma to this college or any other college/university

Group -2 Students

Name	Roll No	Regd. No	Student sign.
Jyoti Rekha Behera	5208B19010	02698/19	Jyoti Rekha Behera
Parvati Sahoo	5208B19011	02699/19	Parvati Sahoo
Rashmi Ranjan Panda	5208B19013	02701/19	Rashmi Ranjan Panda
Saijiban Giri	5208B19014	02702/19	Saijiban Giri
Subhalaxmi Dey	5208B19015	02703/19	Subhalaxmi Dey
Subhasmita Hota	5208B19017	02705/19	Subhasmita Hota
Suman Roul	5208B19019	02707/19	Suman Roul
Tapan Kumar Patra	5208B19020	02708/19	Tapan Kumar Patra

CERTIFICATE

This is to certify that the project report entitled "**Plant Tissue Culture of Tobacco Plant(Nicotiana Tabacum)**" submitted by group-2 student . for the a ward of the degree of bachelor of Science form Nilamani mahavidyalaya Rupsa, Balasore, Odisha, India is a bonafide record of work carried out by them under my guidance. Neither this project report nor any part of it has been submitted for any degree of academic award elsewhere.

Group -2 Students.

Name	Roll No	Regd. No	Student sign.
Jyoti Rekha Behera	5208B19010	02698/19	Jyoti Rekha Behera
Parvati Sahoo	5208B19011	02699/19	Parvati Sahoo
Rashmi Ranjan Panda	5208B19013	02701/19	Rashmi Ranjan Panda
Saijiban Giri	5208B19014	02702/19	Saijiban Giri
Subhalaxmi Dey	5208B19015	02703/19	Subhalaxmi Dey
Subhasmita Hota	5208B19017	02705/19	Subhasmita Hota
Suman Roul	5208B19019	02707/19	Suman Roul
Tapan Kumar Patra	5208B19020	02708/19	Tapan Kumar Patra

28-6-20

Signature of Guide

ACKNOWLEDGEMENT

We the all the group 2 students here by declare that the project report entitled" **Plant Tissue Culture of Tobacco Plant(Nicotiana Tabacum)**"Beang submitted to Nilamani Mahavidalaya ,rupsa, Balasore,Odisha for the award of bachelor of science & in this project report the every student in group-2 are coporate each other & the work is done by the guidance of our department sir Dr. Ardhendu kumar Dash so we thank full to our sir . we also thankfull to our Dr.Santanu Kumar Jena sir for its kindly help. we also thankfull to our Principal Sir For his valuables Suggestion. we also thank full to all staff of Nilamani Mahavidlaya,Rupsa,Balasore, For this Coporation

ABSTRACT

Plant Tissue Culture Successfully Carried out by leaf this method .In this tissue Culture Nicotiana Tabacum Leaf Is taken and tissue culture done Successfully.From the leaf tissue culture tobacco plant Successfully generated.

CONTENTS

List of Figures

Total Figer-8

Chapter-1

Introduction:- Tissue culture by leaf disc method [Nicotiana Tobacum]

Tissue culture is the most demanding particle in plant technology in 21st century for crop improvement and commercial exploitation of which providing greatest achievements in obtaining tissue cultured crop plant varieties. Tissue culture is the modern practice in which isolated plant tissue can be grown into produce plantlets. In course of tissue culture processing requisite technological treatments can suitable done achieve desirable varieties of crop plant. Isolate plant tissue was first successfully grown in artificial culture medium under laboratory condition by P.R. White, R.J. Gautheret and P. Nobecourt [1939]. They were able to grow cambial cells of tobacco stem and carrot root on artificial culture medium. Around 1955 single cell culture was achieved. The advantage of culturing isolate tissues was embryos, anther and other aerial parts have been taking place.

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 micro propagation.

Tissue culture commonly refers to the culture of animal cells and tissue, with the more specific term plant tissue culture being used for plants.

Tissue culture is the growth of tissues of cells in an artificial medium separate from the organism. This is typically facilited via use of a liquid, semi-solid or solid growth medium, such as both or agar. Tissue culture commonly refers to the culture of animal's cells and tissues, with the mare specific term plant tissue culture being used for plants. The term tissue culture being used for plants. The term tissue culture was coined by American pathologist.

- It is easy to select desirable traits directly from the culture setup thereby decreasing the amount of space required for field trials.
- Once established a plant tissue culture line can give a continuous supply of young plants throughout the year.
- The time required is much shortened no need to wait for the whole life cycle of seed development for species that have long generation time, low level of seed production or seeds that readily do not germinate rapid propagation is possible.
- In vitro growing plants usually free from the bacterial and fungal diseases. Virus eradication and maintenance of plants in virus free state. This facilitates movement of plant across international boundaries.
- Plant tissue banks can be frozen and then regenerated through tissue culture. It preserves the pollen and cell collections from which plants may be propagation.

Types of tissue culture:-

Callus culture:-

- Callus culture may be defined as production and maintenance of an unorganized mass of proliferative cell from isolated plant cell, tissue or organ by growing them on artificial nutrient medium in glass vials under controlled aseptic conditions.
- Callus culture need to be subculture every 3-5 weeks in view of cell growth nutrient depletion and medium drying.

Organ culture:-

- That may allow differentiation and preservation of the architecture.
- The organ culture refers to the in vitro culture and maintenance of an excised organ primordial or whole or part of an organ in way and function.

Single cell culture:-

- Single cell culture is a method of growing isolated single cell aseptically on nutrient medium under controlled condition.

Suspension culture:-

- Suspension culture is a type of culture in which single cell or small aggregates of cell multiply while suspended in agitated liquid medium. Suspension cultures are used in induction of somatic embryos and shoots, production of secondary metabolites, in vitro mutagenesis, selection of mutants and genetic transformation studies.

Anther culture:-

- Androgenesis is the in vitro development of haploid plants originating from potent pollen grains through a series of cell division and differentiation.

Embryo culture:-

- Embryo culture may be defined as aseptic isolation of embryo from the bulk of maternal tissue of mature seed or capsule and in vitro culture under aseptic and controlled physical condition in glass vials containing nutrient semisolid or liquid medium to grow directly into plantlet.

Pollen culture:-

- Pollen culture is the in vitro technique by which the pollen grains are squeezed from the intact anther and then cultured on nutrient medium where the microspores without producing male gametes.

Somatic embryogenesis:-

- Somatic embryogenesis is the process of a single or group of cells initiating the development pathway that leads to reproducible regeneration of non zygotic embryos capable of germinating to from complete plants.

Protoplast culture:-

- It is the culture of isolated protoplasts which are naked plant cells surrounded by plasma membrane which is potentially capable of cell wall regeneration, cell division, growth and plant regeneration on suitable medium under aseptic condition.

Shoot tip and meristem culture:-

- The tips of shoots can be cultured in vitro producing clumps of shoots from either axillary or adventitial buds. The method can be used for colonel propagation.

Explants culture:-

There are variety of forms of seed plant viz, trees, herbs, grasses, which exhibit the basic morphological units i.e. root, stem and leaves. Parenchyma is the most versatile all types of tissues. They are capable of division and growth.

Plant in vitro culture techniques:-

The promise of plant in vitro technologies in three major areas, namely micro propagation somatic cell genetics and generation of transgenic plant.

Micro-propagation:-

Propagation in tissue culture is used to develop high quality colonel plant. The main advantages are attributed to the potential of rapid, large scale propagation of new genotype, the use of small amount of original germ plasma.

Somatic cell genetics:-

Contribution of in vitro methods to plant breeding i.e. somatic cell genetics is most significant, mostly in terms of haploid production and somatic hybridization.

Transgenic plants:- Expression of mammatian genes or other plants gene is becoming routine for several plant species. One of the successful approaches has been engineered for resistance against insects, virus and other pathogens as well as herbicide.

Application:-

- Micro propagation is widely used in forestry and in floriculture. Micro propagation can also be used to conserve or endangered plant species.
- A plant breeder may use tissue culture to screen cell rather than plants for advantageous characters.
- Large scale growth of plant cell in liquid culture inside bioreactors as a source of secondary products, like recombinant proteins used as bio-pharmaceuticals.
- To cross pollinate distantly related species and then tissue culture the resulting embryo this would otherwise normally die.
- To cross distantly related species by protoplast fusion and regeneration of the novel hybrid.

For production of doubled monoploid plants from haploid culture to achieve homozygous lines more rapidly in breeding programs, usually by treatment with colchicines which causes doubling of the chromosome numbers.

sic technique required for tissue culture:-

here are several basic technique and step required to produce a tissue culture plant they as follows:-

(1) Acceptic condition in laboratory
(2) Selection of an explants
(3) Sterilization
(4) Selection of culture media
(5) Inoculation of tissue
(6) Growth of callus
(7) Regeneration of callus to shoot and root
(8) Synthesized plant transfer to green house for acclimatization
(9) Acclization plant transfer to the field condition

Following all these basic steps are can synthesize a tissue culture plant.

(1) **Acceptic condition in laboratory**:-

For the plant tissue culture highly sterilized laboratory is required. The laboratory should be neat and clean and regularly room should be properly sterilized. The erective condition in the tissue culture.

(2) Selection of an explants:-

Micro propagation is preferred because of genetic stability for the tissue culture selection of the explants is another important step of the tissue culture selection of an explants mean to selection a proper plant tissue which has power of totipotency.

A plant or piece of tissue used to initiate a culture almost all parts of plant are amenable to in vitro plant regeneration. However, in certain plants some organs may be more regenerative than the others glycogen max, the hypocotyls exhibits higher potentially for shoot formation than the root segments.

The regenerability of explants is influenced by several factors.

i. Organ from which it is derived.
ii. The physiological state of explants.
iii. Size of the explants.

iv. Orientation of the explants on the medium.

v. Its inoculation density.

(3) **Sterilization**:-

Sterilization is the process of inactivating or removing all living organisms from a substance or surface. Different kinds of sterilization procedures were adapted in plant tissue culture.

They are,

a. Heating

b. Radiation

c. Chemicals

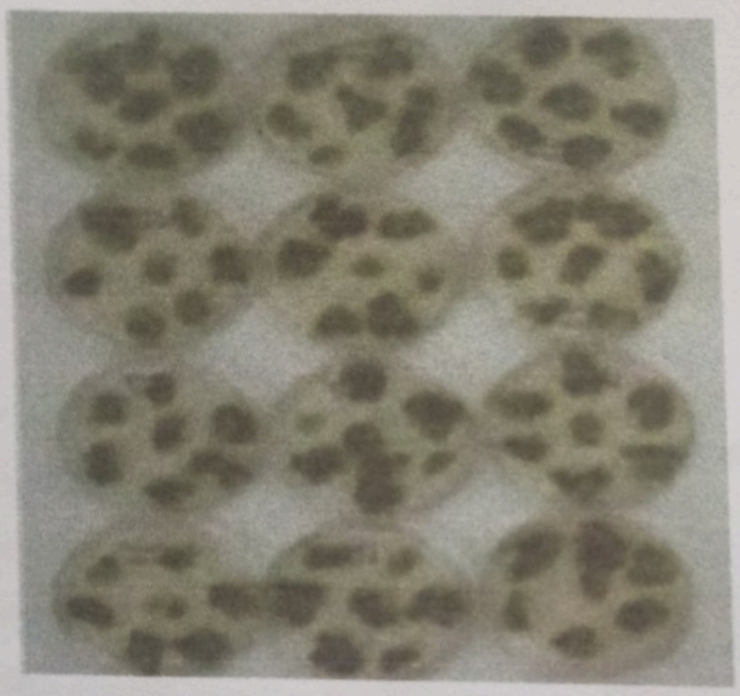
d. Ultra filtration

e. Explants in micro propagation

- In the tissue culture used all material like explants nutrient media, conical flask test tube, peterplate, cotton gauge, scissor, hormones are properly sterilized by different sterilized procedure.
- Media all instrument used by are sterilized by low of allconol and sodium pilloried by micro filters.

(4) **Selection of culture media**:-

To prepare the medium, many researchers mix the stock solutions which were made previously since the medium compositions are generally complicated. The present studies pertained to the micro propagation of dendrocalamusstrictus. For the tissue nutrient media play a vital role M.S media are used.



(5) **Inoculation of tissue**:-

For the tissue culture explain should be inoculation inside the culture media. Inoculation tissue must be kept in laboratory. Light condition should be properly maintained for the inoculation lumana follow as required.

The medium is solidified with 0.5 – 1.0% agar murashingeand skoog standard medium is used for tissue culture.

Methodology of tissue culture has following steps:-

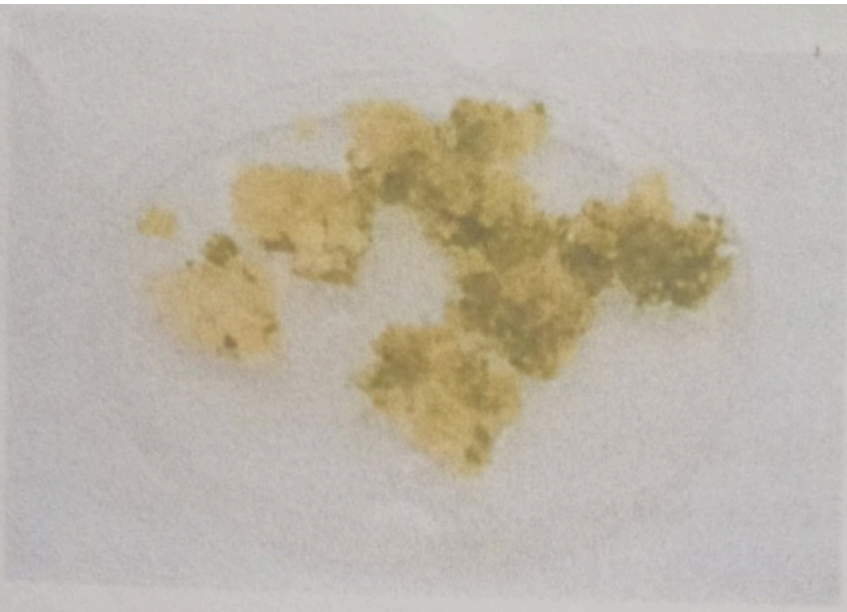
a) Selection of plant material

b) Cultivar is selected and explants is separated

This is known as inoculation of explants.

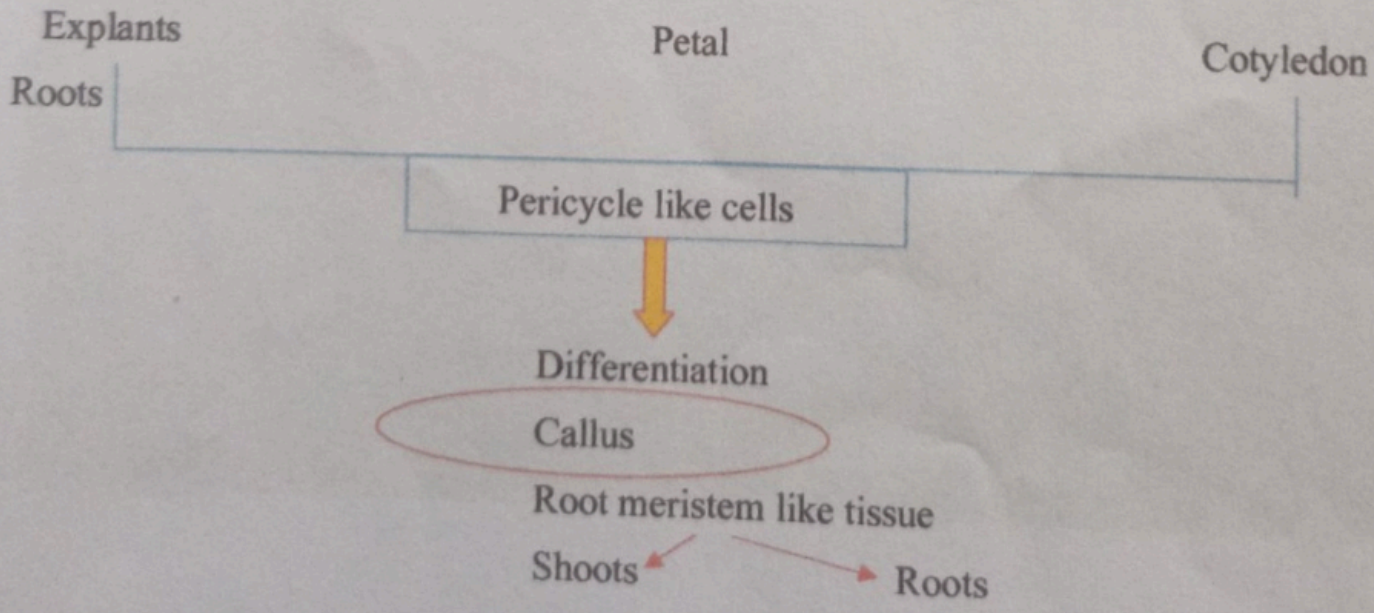
(6) **Growth of callus**:-

- After inoculations in the plant tissue are different sized give rise to a group of cells is called as callus.
- Plant callus is a growing mass of organized plant parenchyma cell. In living plants, callus cells are those cells that cover a plant wound. In biological research and biotechnology callus formation is induced from plant tissue samples after surface sterilization and plating onto tissue culture medium in vitro. The culture medium is supplemented with plant growth regulators, such as auxins, cytokines and gibberellins to initiate callus formation or somatic embryogenesis. Callus initiation has been described for major group of land plants.
- Plant species representing all major land plant groups have been shown to be capable of producing callus in tissue culture.
- A callus cell culture is usually sustained on gel medium. Callus induction medium consists of agar and a mixture of macronutrients and micronutrients for given cell type.



(7) **Regeneration of callus to shoot and root:-**

- After callus formation the callus tissue are transported to the organogenesis for organogenesis. 1st the conclave tissue transfer to the shoot induction medium.
- Getting to the root of regeneration adventitious rooting and callus formation. The
- Capacity of plants to regenerate is impressive. During tissue culture, adventitious roots or shoots can be induced by transferring the callus to medium containing different ratio of auxin and cytokines.
- During tissue culture, adventitious roots or shoots can be induced by transferring the callus to medium containing different ratios of auxin and cytokine, in some cases in nature denovo organogenesis from detached organs leads directly to the formation of a new plant at a wound site, without production of callus.



8) **Synthesized plant transfer to green house for acclimatization;-**

- Tissue culture generated plant are transfer the green house effect plant for acclimatization after few days plants are actimized to the sun light and green house.
- Substantial numbers of micropropagated plants do not survive transfer from in vitro conditions to green house or filed environments. The green house and filed have substantially lower relative humidity higher light levels and septic environments that are stressful to in vitro conditions mast species grown in vitro require an acclimatization process in order to insure the sufficient numbers of plants survive and grow vigorously when transferred to soil.

(9) Acclimation plant transfer to the field condition:-

- The green house acclimatized plants are transfer to the field conditions.
- During the field transfer the in vitro grown plantlets are unable to compete with soil. Plants cultivated in vitro are different from field acclimatization to exvitro conditions leaf thickness.
- The ultimate success of micro propagation on a commercial scale depends on the ability to transfer plants out of culture on a large scale at low cost and with high survival rates. During field transfer the in vitro grown plantlets are unable to compete with soil microbes and to cope with the environmental condition

M.S Media:-

Murashinge and Skoog medium is a plant growth medium used in the laboratories for cultivation of plant cell culture.MSO was invented by plant scientists Toshio Murashinge and Folks K. Skoog in 1962 during Murashinge search for a new plant growth regulator. A number behind the letters MS is used to indicate the sucrose concentration of the medium. For example, MSO contains no sucrose and MS20 contains 20 g/l sucrose.

Along with its modifications it is the most commonly used medium in plant tissue culture experiments in the laboratory.

For the tissue culture nutrient media are used like M.S media are generally like this. In nutrient medium inorganic nutrient like glucose, sucrose, extracted coconut water, peptonetc are used. Then vitamins are also in vitamin-B_1. B_6 V-C etc. Amino acid are used in like L-argin, L-aspatic acid are used solid flying agent and P_H was maintain very strongly above organic used in proper ratio to form a good tissue culture medium.

The number of people killed each year by tobacco will double over the next few decades unless urgent action is taken. But just as the epidemic of tobacco caused disease is manmade, people acting through their government and civil society can reverse the epidemic.

The WHO framework convention on tobacco control with over 150 parties demonstrates global commitment to taking action and identifies key effective, tobacco control policies. Through this landmark treaty, country leaders affirm their citizen's right to the highest. To fulfill this fundamental human right. The M Power package of six effective tobacco control policies, if fully implemented and enforced will protect each country's people from the illness and death that the tobacco epidemic will otherwise inevitably bring. The impact of the M Power policies can turn the vision of the framework convention into a global reality.

Although the tobacco epidemic can be countered, countries need to take effective steps to protect their populations.

Furthermore, the tobacco epidemic is making health inequalities worse both within countries, were in most cases the poor smoke for more than the wealth and internationally with poor countries soon to make up more than 80% of the illness and death cause by tobacco.

Tobacco is unique among today leading public health problems in that means to curb the epidemic are clear and within its reach. If countries have the political competent and technical and logistic support to impart that M Power that M Power policy package. They can give millions of lives.

These report shows that the overwhelming majority of the world population

- Is not fully protected from other peps smoke.
- Is not adequately protected from tobacco company advertising promotion and sponsorship.
- Is not paying tobacco prices that are high enough to substantially reduce tobacco use. Does not desire sufficient health information for tobacco use. And in more than half of the world there is little accurate information on the full scope of the epidemic.

Government around the world collect more than us 200 billion in tobacco takes each year. They spend less than one fifth of 1% of that amount on tobacco control. In many low and middle income countries, government receives about US$ 5000 in tobacco tax revenues for every US$ 1. They spend on tobacco control activities. Yet the costs for the most effective tobacco control interventions. Taxation smoke free public places, advertising, promotion and sponsorship bans and graphic pack warnings are very low only anti-tobacco advertising and cessation services require signification financial resources.

Which are many cases can be covered through increased tax revenues and partnerships.

But all tobacco control measures require political commitment. Because the tobacco industry is far better funded and more polically powerful then those who advocate to protect children and non-smokers from the tobacco and to help tobacco users quit much more needs to be done by every country to reverse the tobacco epidemic. By taking acting to implement the M Power policies, governments and civil societies can create the enabling environment necessary to help people quit

Tobacco use 'WHO' with the help of its global partners, stands ready to support member states as they face the challenges ahead.

Unless urgent action is taken more than one billion people could be killed by tobacco during this century. But this dire future can be changed by the leaders of government and civil society. As the tobacco epidemic is entirely manmade, the end of the tobacco epidemic must also be manmade. We must act now.

Chapter-2

Leaf disc method for plant tissue culture:-

Genetic engineering of plant involves several independent yet closely related steps. First a gene must be identified and isolated. Second, a mechanism for the introduction of the gene into plant tissues must be developed. And third production of whole plants from transformed cells must be performed. A great deal of research time and effect has been spent in the engineering of commercially important plant genes. Herbicide resistance and pathogen tolerance have been the most successful to date.

The introduction of these genes through the use of transformation vectors has also been a major research focus. Isolated genes can be introduced into plants several ways.

Microinjection, electroporation, ballistic incorporation, agrobacteriatumefaciensco incubation, and virus infection are potential methods for the introduction of foreign genes into the plant genome of these methods, co-incubation of plant cells with agrobacterium has been used most effectively for the transformation of plant tissues.

Transformation with agrobacterium was first proposed as means of incorporating DNA into plants by exposing plant protoplasts to the organism. Disadvantages to this technique included the difficulty in obtaining large numbers of high quality of protoplasts and the difficulty in regenerating whole plants from protoplasts. Relatively few plants can be regenerated from protoplasts.

In 1985, scientists at Monsanto developed a leaf disc method for the co incubation of tobacco petunia and tomato leaves with agrobacterium for the transformation of plant tissues. In this method, surface disinfested leaf discs are incubated with an agrobacterium strain containing a Ti plasmid. The leaf discs are then transferred to a selective plate on which only transformed cells will grow. This plate also contains antibiotics that arrest the growth of any bacteria present. The leaf disc technique has been widely accepted and continuous to show great promise for the genetic transformation of many plant species.

Tobacco leaf disc:-

The gram negative soil bacterium agrobacterium tumefacienscauses crown gall diseases in plants. This disease is characterized by the formation of tumors on plants after infection of wound sites by the bacterium. The crown gall disease in plants. This disease is characterized by the formation of tumors on plants after injection of wound sites by the bacterium. The crown gall cells given two genetic properties not proposed by normal plant cells. These are the ability to grow in culture in the absence of externally added hormones and the production of unusual compounds called opines. These opine are used by the bacterium as sources of carbon and nitrogen. The bacterium therefore creates a niche in the crown gall which is favorable for growth.

The molecular basis for this disease was identified in 1974 with the discovery of a 200 kb plasmid called the Ti plasmid. Removal of the plasmid leads to a virulence and reintroduction restores virulence.

The significant finding was that a 13 kb piece of the Ti plasmid is transferred into the genome of the recipient cell. This piece of transferred DNA is called the T DNA. The transformation of cells with this T DNA results in the development of the neoplastic growth called the crown gall tumor this is accomplished by the products of three genes, tms-1, tms-2, and tmr. Tms-1 and 2 codes for the synthesis of cytokine, the two phytohormones involved in cell growth and development. The increased levels of auxin and cytokine directed by the T DNA leads to the timorous morphology in transformed plant tissues.

The transfer of the T DNA into the plant genome is determined by a series of virulences genes located elsewhere on the Ti plasmid and a short 25 base pair regions located at each end of the T DNA.

Through the interaction of several vir gene products and the T DNA boarder sequences, the T DNA is transferred into the host genome. The transfer of T DNA to plant cells is the key step in using agrobacterium tumefaciens as an agent for directed transformation of

Plants with foreign genes are easing constructed by disarming the tiplasmid and by inserting any gene(s) to be transferred within the boarder sequences. In the presence of the vir genes either on the same plasmid or another plasmid in the same bacterial cell the genes are transferred to the plant genome. A growing number of plant species have been successfully transferred with foreign genes using such artificially constructed plasmid vectors. These foreign genes include selectable antibiotic resistance markers, easily scorable bacterial genes and genes for useful agronomic traits such as herbicide tolerance, virus resistance and insect resistance.

Leaf disc transformation of tobacco is a very simple and robust method. It is used with success in many laboratories. The protocol presented here is simplified version of that of horch at all, basically, it consists of immersing the leaf disks in a liquid culture of agrobacterium carrying the chosen transformation vector. The plant tissue and agrobacterium are then co-cultivated on regeneration medium supplemented with an antibiotic to kill the bacteria (cefotaxime) and a selective agent against untransformed plant cells. It takes about 2 mo to obtain rooted plantlets that can be transferred to soil. The protocol presented here works well in our hands with Nicotianatobacum cultivar "petit Havana" mutant SRI (2) and agrobacterium tumefaciens strain LBA 44 04(3) harboring binary vectors conferring kanamycin resistance (100 mg/L). We have also used PB1BHYG (4), which confer hygromycin resistance (50 mg/L).

Leaf disc transformation of tobacco is the paradigm for agrobacterium mediated transformation of plant tissues and subsequent selection and regeneration of transgene plants. This system permits efficient gene transfer, selection and regeneration to be coupled together in a simple process. Tobacco is an excellent host for a tumefaciens and also responds exceedingly well in culture. While the technique is most easily practiced with tobacco, it has been applied to a number of other species. This example will be described for tobacco, using a vector that confers kanamycin resistance, pMON200. Tobacco is a model plant for agrobacterium mediated genetic transformation due to the simplicity of its transformation procedures. The traditional technique does not require

Expensive machinery or complicated procedures. Some significant advantages of using tobacco leaf pieces through organogenesis transformation are:-

(1) Tobacco plants can be easily regenerated from tobacco leaf pieces through organogenesis.

(2) Short acclimatization time and a high transporting survival rate: up to 100% of the in vitro green-house raised plants transferred from lab to ex-vitro. The acclimatization is brief only a matter of days.

(3) Ability to maintain a hemizygous state with T DNA cassette can be maintained at hemizygous state status in a sterile environment through simple vegetative propagation. This can be achieved through cutting of tips or stems and reproduction in solid MS medium without phytohormane. Roots will grow from the cut region and the root plants can then be transferred to soil. If needed the simplicity of the hemizygous T DNA cassette is necessitated in some research products. For example, hemizygous T DNA structure has been used for site specific deletion or integration experiments using site specific recombinases.

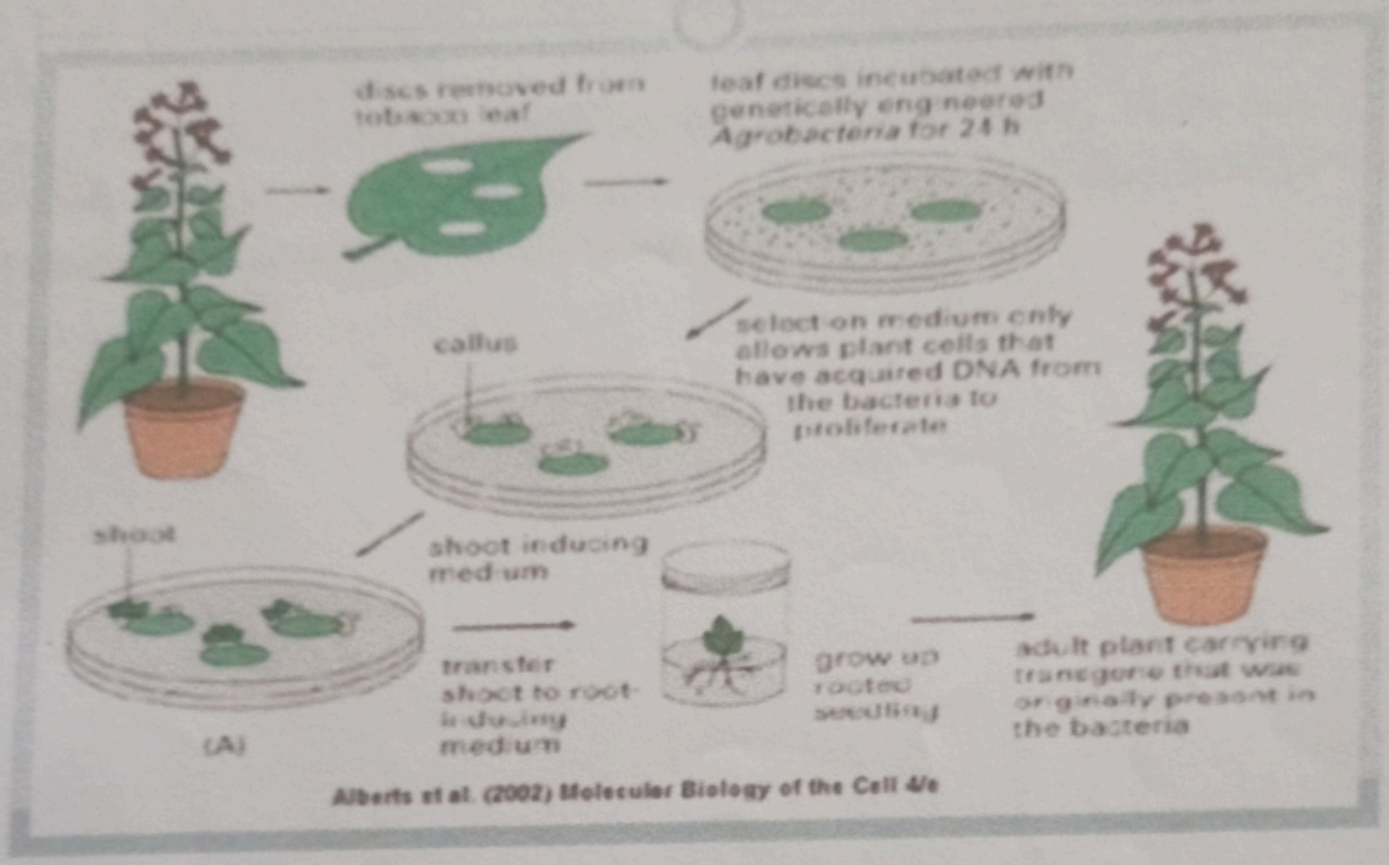
(4) Easy crossing, due to large flower size, hand pollination is easily accomplished.

(5) Longevity by removing the flowering buds or tips, plants continue growing in green house conditions for extended periods of time, which provides supplemental experimental material, particularly for the W 38 tobacco species.

(6) Prolific seed production for sustaining lines and testing results.

(7) Increased biomass with the potential for molecular forming to produce recombinant proteins due to tobacco's high biomass yield.

(8) This plant is commonly used for ago infiltration transient assays. Due to the variety of advantages mentioned above, most plant research scientist view tobacco as a prime choice for genetic transformation in proof of concept. Experiments with the added benefit of multiple practical uses.



Alberts et al. (2002) Molecular Biology of the Cell 4/e

CONCLUSION

Plant Tissue culture successfully carried out by leaf this method. In this tissue culture of **Nicotiana tabacum** leaf is taken and tissue culture done successfully. From the leaf tissue cultured tabaco plant successfully generated.

Examiner
28.06.2022
(External)

28.6.22
(Internal)