Propagation and Hardeningof Ornamental Plants via Hydroponic System

Submitted in fulfillment of the requirement for the degree of

Bachelors of Technology

IN

BIOTECHNOLOGY

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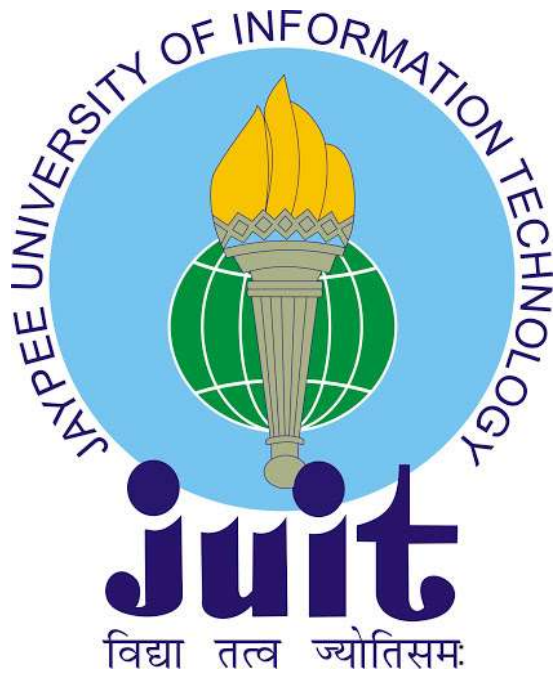
UNDER THE SUPERVISION OF

DR. HEMANT SOOD



DEPARTMENT OF BIOTECHNOLOGY AND BIOINFORMATICS

JAYPEE UNIVERSITY OF INFORMATION TECHNOLOGY

SOLAN (H.P)

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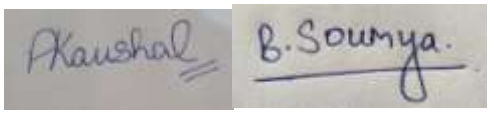
DECLARATION

We hereby declare that the project work entitled "**Propagation and Hardening of Ornamental Plants via Hydroponic System** " submitted to the Department of Biotechnology and bioinformatics ,Jaypee University Of Information Technology Solan(H.P), is an authentic record of original work done by us . The work was carried out under the guidance of **Dr**.**HemantSood**.



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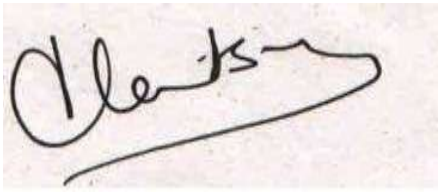
Date:15th July, 2020

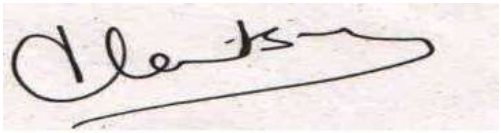
This is to certify that the above statement made by the student is true to the best of my knowledge.



Dr. Hemant Sood

SUPERVISOR CERTIFICATE

This is to certify that the work titled " **Propagation and Hardening of Ornamental Plants via Hydroponic System** "by Arush Kaushal and B Soumya during the end semester in June 2020 in fulfilment for the award of degree of Bachelor of Technology in Biotechnology of Jaypee University of Information Technology ,Solan has been carried out under my supervision .This work has been submitted partially or wholly to any other University or Institute for the award of any degree or appreciation



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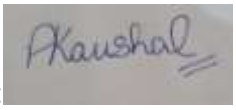
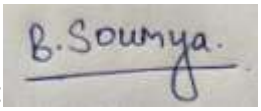
ArushKaushal : 

B. Soumya : 

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ABSTRACT

Hydroponic system is used to grow different plants but we have carried out additional extension usage of hydroponic for carrying out *ex-vitro* rooting and hardening of tissue culture plantlet .We have carried out growth and multiplication of *OrientalLilium, Catalognafrastagliata, Origano vulgare*[1] *and Orchids Cymbidium*under invitro condition and optimized condition of Hydroponic set up .For invitro multiplication of these plantlets MS media along with different growth hormones were being simultaneouslyused in Hydroponic System having NFT modes and also it is being used for carrying out rapid multiplication under NFT mode including flow rate (1-2l/min for each channel) ,pH (5.5),and EC (1.6) temperature25 º ±C and humidity ranging from 70-80% and media containing MS media + NAA+BAP for *Catalognafrastagliata,*MSmedia +NAA +BAP+IBA for *Origanovulgare*[2], MS media + NAA+BAP+KN+GA for *OrientalLilium,*MS media + NAA+BAP+ KN for *Orchids Cymbidium* were multiplied .Under optimized hydroponic condition *ex-vitro* rooting and hardening were carried out to save time and expenditure .So we attained *ex-vitro* plantlet of*OrientalLilium, Catalognafrastagliata and Orchids Cymbidium* where as*Origanovulgare*didnot respond effectively. Therefore in this study we achieved exceptional results in an average of 10 days under hydroponic system along with significantly higher survivability of these maintain plants after that these plantlet were transplanted inpotting aggregate vermiculite, perlite and cocopeat in ratio of 1:1:3respectivelyforgrowth and development .

CHAPTER 1

INTRODUCTION

Tissue cultured [3]plant life are most appropriate as they produce the precise copies of the plant with the applicable trait .Furthermore, this method also produces vegetation that isextraordinarily unexpected compared to thetraditional method of developing plant life and in addition diverse vegetation may be acquired through from tissue lifestyle when seeds or plant pollinator are absent . For asuccess breeding of plantlife they must be acclimatized in an ex vitro scenario also know as hardening , it truly is the

fundamental bottleneck of this method. So, hydroponic is the best solution for hardening.

1.1 Hydroponics-Art of developing of plant without the utilization of soil in nutrient rich solution. Moreover Plants grab nutrients from their roots . The hydroponic medium has vitamin, growth hormone enriched in that encourage plant enlargement. The unwanted nutrient awareness can be avoided with the removal of soil. Nutrimentnoxiousand dearth[4]each are essential for plant boom and these problem can be modified by means of hydroponics. It was cited that the hydroponically grown plants develop faster as comparison with the traditional system of growing plant in soil due to the fact the vegetation are immediately supplied withample amount of nutrients.

1.2 Cultural medium - They are stuffthat bolster up the plant extension. Cultural mediumis precisely planned to guide vegetation boom and it can be robust or fluid. Various kinds of developing medium are being used to domesticate numerous vegetation. Cultural mediumis also known as called increase medium. Hardening of vegetation requires medium which can bepure component aspect. Hardening of plant life need a substrate that could each be a mixture of the beneath mechanism or a solitaryconstituent.

1. **MS media**-Murashige and Skoog medium is a vegetation boom medium used within the laboratories for crop growing of vegetation mobile manner of lifestyles.

2. **Coconutdust**- coir also referred as coconutshell dust, are the remains of most outer sheath of the coconut's after its fibre. It also includes the Trichoderma fungi that safeguard roots and sell root increase.

3. **Perlite-** Light weighted glass pebbles that were acceleratedin a manner of volcanic rocks being over heated. Retains much little water and ample quantity of air.

4. **Vermiculite**- possesample amount of irrigate than perlite.

Table1 - The desk constitute the constitute of inventory suspension of MS media

Stock solution and components	*for MS medium g/L*	*for DKW medium g/L*
Stock solution A (nitrogen):		
Ammonia nitrate	82.5	98.0
Potassium nitrate	95.0	—
Calcium nitrate	—	98.0
Stock solution B (sulfates):		
Magnesium sulfate heptahydrate	18.5	37.0
Potassium sulfate	—	78.0
Stock solution C:		
Calcium chloride dihydrate	22.0	7.35
Potassium phosphate	8.5	13.0
Stock solution D (chelated iron):		
Ferric sulfate heptahydrate	1.39	1.65
Sodium ethylene dinitrotetraacetic acid	1.88	2.25
Stock solution E (micronutrients):		
Manganese sulfate monohydrate	1.110	1.700
Zinc sulfate heptahydrate	0.430	—
Zinc nitrate hexahydrate	—	0.850
Boric acid	0.310	0.250
Potassium iodide	0.042	—
Sodium molybdate dehydrate	0.013	0.020
Cupric sulfate pentahydrate	0.0013	0.0125
Stock solution F (organics):		
Myo-inositol	5.00	5.00
Glycine	0.10	0.10
Pyridoxine hydrochloride	0.025	—
Nicotinic acid	0.025	0.05
Thiamine	0.005	0.10

1.3**Hardening**-Hardening [5]is the manner used for the transplantation of vegetation grown in artificial medium to outdoor environment by using attune them to the barbarousnature situation oneafter the other . This is accomplished by way of manifest them to fluctuating temperature, moisture and luminosity conditions.

1.4TYPES OF HYDROPONIC SYSTEM

There are specific sorts of hydroponics however we've defined here NFT which we've used :

1: **Nutrient film method (NFT):** The key is pressured hooked on the channel that preserve a differentquantity of vegetation. The structure is somewhatlean, the solution run through the channel touching the roots and then revert reverse inside the hydroponic system.

1.5 Growth in Hydroponic is affected By Following Factors

A:Electrical conductivity:

Electrical conductivity of whichever medium used for the vegetationextension and improvement of crop regulate the supply of vitamins andquantity of nutrient persist in that unique media. This limit is calculated using tool called EC meter. The electrical conductance varies amongplant .Salts within the media is directly propositional to the EC of the medium. The stretch of EC extendfrom 1.5-1.8intended the most advantageous boom of the plant life. In order to decorate the electric conductivity of the media used additional fertilizers can be optedin orderto decrease the assess pure water can be utilized that let you to form medium more amicable for the vegetation increase. Unit for the electric conductivity is:Siemens meter^2 per mole (S/m^2/mole)

B:pH:

Plants for his or her boom in hydroponic device calls for most beneficial limitof pH of their fluid in which they are grown-up . It additionally contributethe supply of vitamins to the vegetation. The scale of the pH is from 5.8-6.2,consequently normally plants grows within the ultimate variety of pH.pH of the fluid can be regulated using acid (HCL)or base(NaOH).This limitis calculated the usage of pHmeter. If medium wherein plants are full-grown does

no longer contain the most appropriate variety of the pH, they will not take up nutrients.

C:Temperature:

Temperature is some other important component which have an effect on the boom of the plant. Higher temperature result in the evaporation of the moisture and while decrease temperature contribute poor vegetation extension . Best limitrange is from 19°-24°C.Extortionateor much little temperature can also purpose trouble to vegetation.

D:Dissolved oxygen:

Dissolved oxygen is overall quantity of gaseous oxygen exist inside the fluid. If the plant in the systemhave disadvantaged of the oxygen .The outcome isto little or absence of absorption of the minerals from roots with the intention to sooner or later cause end of the vegetation consequently to hold the viability of the vegetation dissolved oxygen is mandatory. Designed forthe uniformity of dissolved oxygen in the fluid oxygen is provided by using oxygen pump and consequently enables in plant increase.

CHAPTER 2

REVIEW OF LITERATURE

Hydroponic

Many elements are answerable for the damage of land used for cultivation of flowers main the principal cause is worldwide climate change , distinctive motives encompass soil erosion , over use of chemicals which results in damage of fertile land. Influence of those elements can be visible in flora, which go phuttowardsmeet the requirement of enormous population. Hydroponics[6],in this case performs a important part that could contribute in filling the gaps. This is the nutrient rich lifestyle which enables in dealing with diverse problem.

Hardening of in vitro vegetation plant is one more software of the top soil less subculture.

Approaches of hydroponics:

1) continuous strategies (move go with the flow solution cultured gadget)/non-stop

2) Discontinuousmode

Thus hydroponics no longer most effective meets the aspiration of the inhabitants but moreover influences the productivity of manufacturing of the flower[7].Hydroponics pivot no longer handiest on the enlarge of medicinal vegetation however hardening is the primary assignment. Several studyprofessthat the eye of secondary metabolite substance can be numerous whilst those vegetation are located under managed environment.

Therefore hydroponic tool is a beneficial approach of raising numerous flora such as Catalognafrastagliata and different veggiesby controlling numerous situations like the temperature ,pH, humidity, slight, fertigation means irrigation and fertilizer we are both to the plant in the form of nutrients. The aim ofraisingexceptional medicinal plants in hydroponic structures in order to attain sufficient increase relationship with tremendous crop terrific and huge quantityof secondary metabolite compound. Cheap source like the low price and chemical blessings , the hydroponic structures of raising medicinal plant life, serve in defence of numerous plants and also fashion of untamed species[8].Hydroponics is likewise used in investigation a biotic pressure forbearance.

Hydroponics permits scientist to without problems take a look at the consequences of compound deficiencies and toxicities and to have a look at extraordinary additives of plant improvement below precise situation. By hydroponics humans infer that plant life take up their food inseveralways and in various awareness.

Shift of ofinvitro grown flower to hydroponic device differ from one flower species to some other fashionable boom solution possesof the following essential macro-factors: nitrogen (N), potassium (K), phosphorus (P), calcium (Ca), magnesium (Mg) and sulphur (S); and micro-elements: iron (Fe), boron (B), copper (Cu), manganese (Mn), etc.

Molybdenum (Mo) and chlorine (Cl), and, for fabaceous species requiring Nitrogen fixation, cobalt (Co).Also additives solution is boosted with carbon source both organic or inorganic.

pH of the solution is also important aspect to notice because it influences the dissociation of ions and uptake of nutrients with the aid of the flower .pH may be regulated with the aid of the usage of computerized structures linked with the system. Acid and Base may be confer into solution on every occasion there can be changes within the pH of the solution. Hydroponics can manipulate the state of affairs in a biotic pressure condition such as salinity, toxicityandcompound deficiency in the plant[11].Several nutrient answer affects the fulfilment of drug compounds in medicinal plant[10].

Tissue Culture and Hardening :

Tissue Culture is the cultivation of plant cell, tissue,explants on specific nutrient media. Under the synthetic state of affairs. Artificial state of affairs include way of life room temperature 20-30°C,humidity 20-98%,fluorescent mild 10,000 Lux. Both moderate and temperature ought to be adjusted in step with 24 hours length .Tissue Culture is an important strategy within the developing countries for the producing of healthy plant, production of high quality plant . It can be classified in 2 category :Invitro rooting , Exvitrorooting . Invitro rooting means cultivation of explants, plant cell wherein micro cutting are rooted at the floor of solid media or immersed in liquid media in test tube or jar. On the alternative hand exvitro rooting of micro decreasing rise up the use of hydroponic media. Furthermore exvitro rooted plant have better advanced root tool and extra danger of survival , tons less time consuming due to simultaneous incidence of rooting as examine to invitro rooted flora.

For the establishment of tissue culture existence plant to the outer surroundings , hardening is the method which account for the most survival of invitro grown plants.

Hardening in fact manner exposing plant to severalaspectssuch asdaytime , dry air , humidity and so on. It is typically a uninteresting technique which incorporates numerous strategies and techniques.

Direct shift of tissue way of life expanding flora to situationbecause of excessive price of demise, Direct switch to daytime moreover motives charring and sizzling of leaves and drooping of the plant .It is important to adjust plant to its encircling through the hardening approach [9]. Hydroponic is a way of growing flowers inside brief period of span in nutrient rich medium . In this technique required amount of nutrient is supplied to plant ,nutrient toxicity and its deficiency are the maximum essential element in plant increase and that can be with out issue manipulated in hydroponic tool. There are many types of hydroponic system . Numerous features affecting the increase of in vitro grown plant at the same time as hardened in hydroponics. Salinity is an important vital thing which donates to the increase of the flora underneath one of a kind conditions and one of a kind media . This is calculated with tool known as electrical conductivity meter consequently variety at which vegetation suggests best increase is among one.Five- 2.5 ds m [12]. Dissolved oxygen is every other crucial detail donating to the vegetation boom .Several studies showed the impact of dissolved oxygen within the multiplication of the roots [13]as a result it's miles important to complement the media with oxygen the use of air pump.

One of the green approach of developing plant life in hydroponic gadget is nutrient movie technique (NFT) . In Nutrient movie approach , skinny film of nutrient answer inside the channel wets the basis and allow plant to grow . Channel of the nutrient film is slightly slanted to allow water to move into the tank at the way to save you Oxygen deficiency in nutrient medium. In this studies we've got optimized the subculture situation for the fastest possible boom of Catalognafrastagliata, Oriental Lilium[14],Oregano vulgare[15] , orchid cymbidium . Ex vitro rooting and hardening had been additionally optimized by way of using the usage of hydroponic tool as an opportunity fast approach .

Table 2:Data collected from preceding examine for the following flora , where scientist grew those plant in hydroponic device and desire end result received by way of the scientistduring the experiment.

S.NO	Plants	Scientist	Method adopted by the Scientist	Hydroponic media	Result obtained by the scientist
1	*Catalogna frastagliata*	Lovro Sinkovic Janez Heribar,Drgean Znidarcic	Seed sowing	Murashige and Skoog media+IBA+BAP.	**5 out of 8 pots**
2	*Oregano vulgare*	Stapleton ,Suzanne C, Robert C Hochmuth	Vegetative propogation	Fe-EDTA Nutrient solution	**6 out of 9 pots**
3	*Black out*	**Hassan M. Asker**	Vegetative propogation	Black out were grown in these different nutrient solution :Rain water, Hoagland, Basal Salt Mixture, Murashige and Skoog Basal Salt Mixture and White's Basal Salt	**8 out of 9 pots**
4	Orchid *cymbidium*	Shahram Sedaghathoor, Gholamreza Golzari Dehno, Rohangiz Naderi	Vegetative propogation	water	12 out 15 pots

2.1*Catalognafrastagliata*

Catalognafrastagliata is grown in Italy . It belong to Asteraceae circle of relatives and itbelonges a dandelion range. Maximum height of the plant is 18 cm tall .This sour crunch vegetable is also referred as Pantarelle (Roman name). The roots of the variety are used as espresso alternative . June – October are beneficial situation for developing this plant[4] . It is high in diet A and C and correct source of minerals like calcium , iron, folic acid, iron. Stem of Catalogna has high dietary great[18]. It is also rich in antioxidant, flavonoids,phenoliccompound[19].

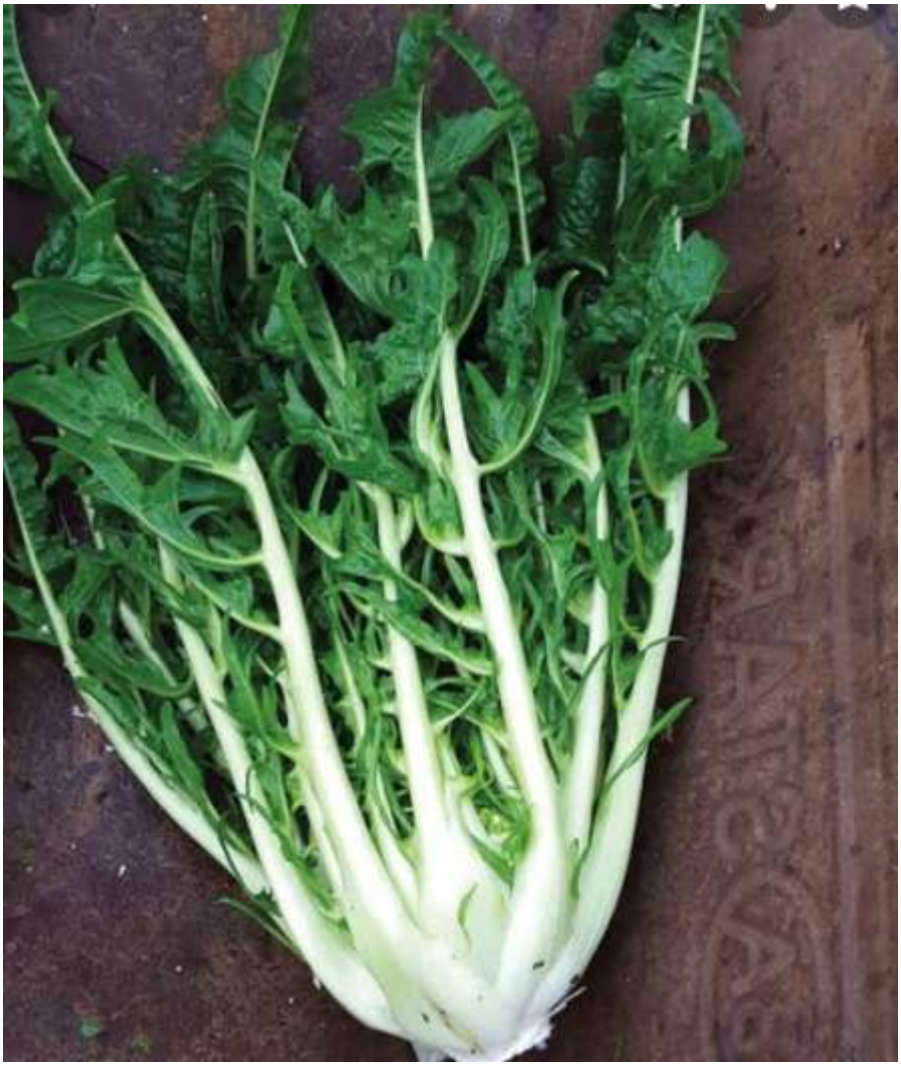


Figure 1 :*Catalognafrastagliata*

2.2*Oregano vulgare*

Oregano vulgare is a Perpetual developing to 0.6 m by using 0.8 m on an average price. It is strong to region (UK) 5 and is not chill tender. They grows mostly in July till September (Best period for their growth). The species is hermaphrodite and is cross-pollinated in such a way of Bees, Lepidoptera .

Oregano play important role in the field of gastronomic and therapeuticbasil for lots of years. It is effectivein gastric and respirational structures and is also

used to sell menstruation. It thought to now not to be therapeutically by way of prenatal women though it's miles flawlessly safe in small amounts for gastronomic functions. The trees and pinnacle stems are strongly antiseptic[1], antispasmodic, stomachic and mildly tonic[3].

The essential oil remains used fashionable aromatherapy to deal with the same sorts of lawsuits that the herb is used for[2] and chemical composition of the essential oil from 3 chemo forms of Origano vulgare develop in wild in Campania (Southern Italy [20].

Figure 2:*Origano vulgare*

2.3*Oriental Lilium*

Lilium is a kind of herbaceous pinnacle flowers developing from rhizomes, all through massive outstanding floras. It belong to Liliaceae circle of relatives. Lilies are a set of pinnacle flora which remainsignificant in culture and works in a great deal of the biosphere. They grow mostly in northern areas . Many different plants have "lily" in their mutual call but aren't linked to actual lilies.

In the Chinese medicinal drug as skillful in Japan, the legitimate Japanese administrative pharmacopeia includes the use of lily bulb in conventional pharmacological , list the usage of the following species: Liliumcifolium, Liliumbrownii, Liliumbrownii var. Colchesteri, Lilumpumilum[21] The balancescrumbled off from the rhizomes are used, commonly steamed.[22]

In South Korea, the lilium class which can be formally indexed for medicinal use areLilumlancifolium Thunberg; Lilium brownii var. Viridulun Baker;[23][24]

Figure 3 :*OrientalLilium*

2.4 *Orchids Cymbidium*

Orchid, any of almost thousandstypes and greater than twenty two thousands class of delightfullyflower-patternedflowers disbursed through the arena, particularly in drizzly tropics. Orchidaceous is a associate of Asparaguses, an command of monocotyledonous pinnacle plant life that still contains the asparagus and iris households. The term orchid is resulting from the Greek saying (orchids) for testicle due to the form of the origincorms in some class of the kind Orchid. These non-woody perpetual vegetable lives are generally native or epiphytic basils (i.E., developing on different flowers instead of fixed in mud). Folks connected to different plant life frequently are vine like and

feature a softorigincover known as the velamen that engrosseswater from the nearbymid-air. Maximumclass manufacture their individualmeal, however nearly stay on lifeless carbon-based fabric (saprophytic) or are helped to reap food via a mushroomresiding of their origins.

Figure 4: *Orchids Cymbidium*

2.1 Rationale and Research gap

- As the degree of existence of in vitro flora throughout hardening is actualabundant less throughpredictable hardening strategies. So, this trial makes a speciality of cumulative the existencedegree of in vitro grown flora when transported from laboratory to grounds.

- As the traditional hardeningapproach could be aperiodoverwhelming and limits for optimization of hardening proceduresdiffers from plant to plant .Therefore, which will reduce time for hardening, one of a kind hydroponic device could be enhanced.

- Hydroponic systems are by and large cast-off for increase of propagation and herbalplantswhereas, few medicinal vegetation had remained targeted for spread.

- No Hardening in hydroponics be there achieved but ex-vitro rooting along with hardening is not yet reported in tissue culture plants by using hydroponic system .

2.2 Objectives:

- To carry out invitro multiplication of(*Oriental Lilium ,Catalognafrastagliata ,Origano vulgare and Orchids Cymbidium*).

- Designing of Hydroponic device for multiplication and Hardening ofplants (*OrientalLilium ,Catalognafrastagliata ,Origano vulgare and Orchids Cymbidium*).

- Optimization of condition for Ex-vitro rooting and hardening of tissue culture plantlets .

CHAPTER 3

MATERIAL AND METHOD

3.1 Propagation of invitro multiplication

(a) Media preparation and culture conditions

Numerous MS media (Murashige and Skoog 1962) complementedby distinctive concentrations besides combos of IBA, KN and BAP through sucrose 3% (w/v) were organized. The pH of every media became adjusted to 5.7 the use of zero.1 N HCl and zero.1 N NaOH and ultimately agar-agar 0.8% (w/v) become introduced as a crystalizing agent. All the media have been autoclaved at 121°C at that time 15 lb/in stress for 15 - 20 min. The cultures were incubated in plant tissue culture chambers maintained 25 ± 2°C having 70 % qualifiedmoisture, 16 day/8 h night time photoperiodat photosynthetic photon flux density of forty µmol m-2 s-1 supplied by calm white glowing tubes (Philips, India).
So in table 3 media composition is mentioned for invitro multiplication of (*Catalognafrastagliata,Origanovulgare,OrientalLilium,Orchid cymbidium)*

Table3:MS Mediacomposition for the following plants

S.No	Name of Plants	MS media composition
1	*Catalognafrastagliata*	MS media +NAA+BAP
2	*Origanovulgare*	MS media +NAA +BAP+IBA
3	*Oriental Lilium*	MS media + NAA+BAP+KN+GA
4	*Orchidcymbidium*	MS media + NAA+BAP+ KN

3.2 Methodology for carrying out propagation of (*CatalognafrastagliataandOrigano vulgare, Oriental Lilium , Orchid Cymbidium):*

(a)For Germination of seed in petriplate

Blotting sheet was cut according to the diameter of the petriplate

Blotting sheet was put in the petriplate

And inside that few seeds were placed

Distilled water was sprinkled on the seed

Petriplate was kept in culture room

(b)Culturing of plantlet in liquid media

1. Weighed 0.5 gm of bavistin and dissolved in 100ml of distilled water

2. Weighed 0.1 gm of mercuric chloride and dissolved in 100ml of distilled water

3.In LAF sterilization of plant was done with both of them and was placed in liquid media

(c)Subculturing of *Oriental Lilium and Orchid Cymbidium*

In LAF sub culturing of Lilium was done.

Petri plate , Blade, Forceps, Jar was sterilized.

Explants was removed from the jar with the help of forceps.

With the help of forceps and blade incision were made on the shoot.

Then these small shoot was transferred into fresh jar with the help of forceps.

The jar was again sterilized and kept in culture room for future growth.

3.3HYDROPONIC SYSTEM

Formation of Media

1. BAP,NAA , KN and GA growth hormone was weighed 20 mg/ml ,18 mg/ml , 10 mg/ml and 30mg/ml respectively using weighing balance.
2. 10 l distilled water was measured using measuring cylinder .
3. Then in bucket 10 l distilled water ,15 ml N,P,K each and growth hormone was added.

4. Mixing is done properly .
5. Electivity conductivity of media was set (i.e. 1.5 ppm) using EC meter .
6. pH of media was set (i.e.5.8) using pH meter .
7. Temperature was maintained to be ±25° C in glass house
8. Humidity was maintained at 90%

Figure 5: Nutrient Film technique

CHAPTER 4

RESULTS

AND

DISCUSSION

Propagation of plants (*Origanovulgare,Catalognafrastagliata, Oriental Lilium ,and Orchids Cymbidium*) were carried out by using seed as explants. Culturing of seeds were carried out in liquid as well as solid MS media as mentioned above. Growth parameters were recorded at regular intervals of 15 days from each experiment.

Table4:Growth parameters observed under culture condition

Plants	MS media Composition	Number of invitro roots grown	Number of days
Origano vulgare	MSmedia+NAA+BAP+IBA+Sucrose +Agar	6	16
Catalognafrastagliata	MSMedia+NAA+BAP+ Sucrose+Agar	18	14
Oriental Lilium	MSmedia+ NAA+BAP+KN+GA + Sucrose + Agar	10	15
Orchids Cymbidium	MSMedia+NAA+BAP+ KN+ Sucrose+ Agar	10	15

4.1 Invitro rooting in culture room

(a)*Origano vulgare*

(a) (b)

Figure 6:(a) Seed germinated in petri plate

(b)Invitro growninpetriplate

Figure 7: Culturing of plantlet in liquid media

(b)*Catalognafrastagliata*

(a) (b)

Figure 8:(a)Invitro grown in petri plate

(b) Culturing of plantlet in liquid media

All these plants were kept in Hydroponic for the induction of exvitro rooting and along withacclimatizationof these plantlets to outside environment growth parameters and multiplication were observed in table 5 and mentioned in figure :(11,13,16,17a)

Table 5: Data Recorded in Hydroponic system

Plants	Media used in Hydroponic	Number of Exvitro roots grown	Number of days
Origano vulgare	Distilled water + NAA +BAP+IBA	10	10
Catalognafrastagliata	Distilledwater+NAA+BAP	25	8
Oriental Lilium	Distilled water+ NAA+BAP+KN+GA	16	9
Orchids Cymbidium	Distilledwater+NAA+BAP+ KN	16	9

4.2*Exvitro*rooting In Hydroponic system

(a) Origano vulgare

(a) (b)

Figure 9:(a)Germination of*Origano vulgare*

(b)Growth after 2weeks

Figure 10: Growth of after *Origano vulgare* 2 weeks

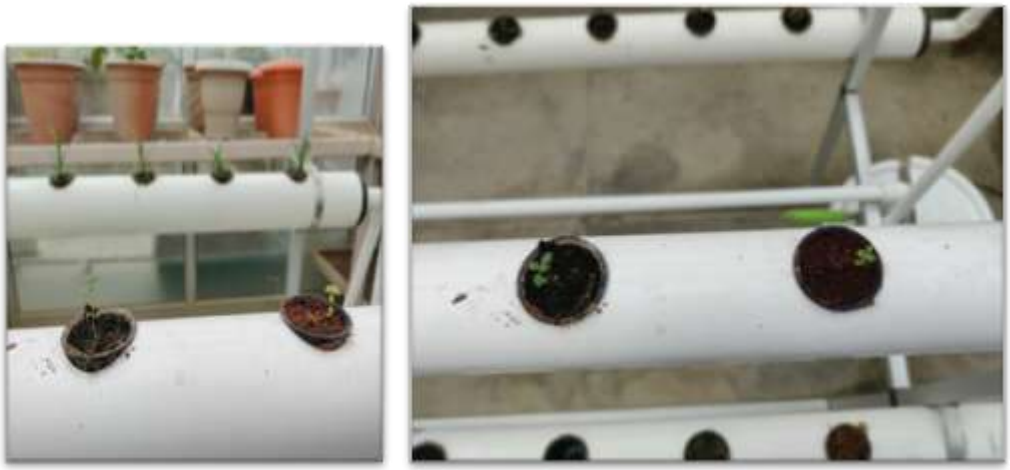


Figure 11: Growth of *Origanovulgare*after 4weeks

(b)Catalognafrastagliata

(a) (b)

Figure 12:(a)Growth of plantlet as shown in table number 5

(b)Hardening of *Catalognafrastagliata* in Hydroponicsystem

Figure 13:*Exvitro* rooting and hardening of plants

Figure 14: Transfer of fully grown *Catalognafrastagliata* to pots

(a) (b)

Figure 15: (a) Growth in pots after 3 week

(b) Transfer of fully grown *Catalognafrastagliata* to pots

(C)*Oriental Lilium*

(a) (b)

Figure 16:(a) Hardening of *OrientalLilium* in Hydroponic system

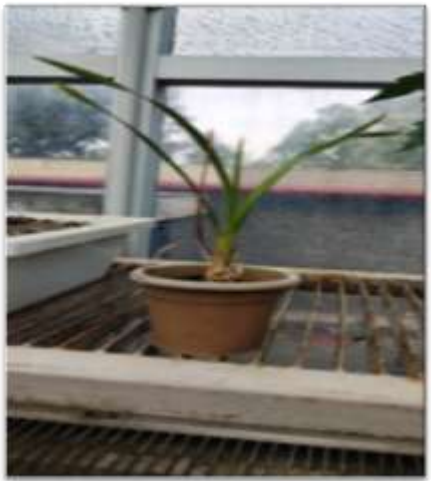
(b) Growth after 4 weeks

(d)*Orchid Cymbidium*

(a) (b)

Figure 17: (a)Hardening of *Orchid cymbidium*inHydroponic system

(b) Transferred to pots and Growth after 1 week



(a) (b)

Figure 18 :(a)Growth in pot after 3 weeks

(b)Growth in pots after 5 weeks

Discussion

Tissue culture is the growth of tissue or organ in artifical medium detached part from the whole plant . This is generally facilitated through use of liquid , semisolid and solid medium inclusive of liquid suspension or agar.Tissue culture of following plants : like *Catalognafrastagliata ,Oriental Lilium, Oregano vulgare ,OrchidCymbidium*in invitro we got good result .After that we made an attempt to harden these plant and transfer them to hydroponic system . Hydroponic system is a process of growing plants without soil providing required nutrients to the plants.After 2 weeks we transferred these plant in Nutrient film technique. Providing optimized environment to these plants and we achievedwonderful result in all the 4 plants in hydroponic system .Best growth was achieved in *Catalognafrastagliata*as compare to previous study performed by these scientist(*LovroSinkovic ,JanezHeribar,DrgeanZnidarcic*)[25]in hydroponic system.Where as in case of*OrientalLilium* results were obtained similar to the previous study performed by scientist*Hassan M. Asker* [26].In hydroponic system we observed maximum root formation as compare to in-vitro .In ex -vitro we achieved maximum growth in hydroponic system . After6 weeks we transferred these plants into pots and providing them vital environment condition like temperature , water ,sunlight, humidity and these plants were kept in green house.

CHAPTER 5

CONCLUSION

Hardening method have been multiplied and advanced over the last limited year but still many bottlenecks exist. So with the passage of time many parameters were tested and many systems were designed to improve propagation potential and hardening status of plants .The presence study explored many aspects of Hydroponic system to be used for propagating plants (*Origanovulgare,Catalognafrastagliata, Oriental Lilium and Orchids (Cymbidium*)) and concludes that in*Catalognafrastagliata*which is an Italian variety best growth has been achieved under invitro conditions and also in hydroponic system within2-3weeks.Where as for *Origanovulgare, Orchids (Cymbidium) and OrientalLilium*optimize growth has been achieved in both invitro and hydroponic system within 3-4 weeks . Also this study provided us ample of opportunity for exploration of theseplants for their commercial growth.

CHAPTER 6

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