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RESEARCH ARTICLE

MICROPROPAGATION AND STANDARDIZATION OF PHOTOPERIOD IN G9 BANANA (GRAND NAINE) *INVITO* CONDITION

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ABSTRACT

Banana fruit is found to be one of the essential source for all the nutrient content in it. Due to increase in the population the demand for the food crops increases. Microprapagation in invitro condition is one of the best methods adapted to increase the productivity. In this study suckers from the matured banana plant was taken and surface sterilized and media preparation was done using different composition of macromolecules, micro molecules and trace elements for the multiplication of propagated plantlets in the MS medium this step was followed by the standardization of photoperiod for the micro propagation. Medium containing BAP, Ascorbic acid and agar composition gave a good result and the shoot induced plantlets were transferred to the root inducing medium were standardization of root inducing medium was done with different composition of charcoal in which charcoal containing 2% charcoal gave a good yield after which the plantlets were transferred to the primary hardening stage. In this stage we can eliminate the poorly developed plants and the diseased plantlets.

Key words: Micropropagation, g9 banana.

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INTRODUCTION

Banana is the common name for a fruit and also the herbaceous plants of the genus which produce the commonly eaten fruit. They are native to the tropical region of south Asia and Australia. Today, they are cultivated throughout the tropics. Banana plants belong to the family musaceae. They are cultivated primarily for their fruit, and to a lesser extent for the production of fibre and as ornamental plants. As the bananas are mainly tall, upright, and fairly sturdy, they are often mistaken for trees, when the truth is the main or upright stem is called a pseudo stem literally meaning "fake stem" Bananas are grown in at least 107 countries. In popular culture and commerce, "banana" usually refers to soft, sweet "dessert" bananas that are usually eaten raw. Micro propagation is the art and science of plant multiplication in vitro. The process includes many steps-stock plant care, explant selection and sterilization, media manipulation to obtain proliferation, rooting, acclimation, and growing on of liners.Micropropagation is the practice of rapidly multiplying stock plant material to produce a large number of progeny plants, using modern plant tissue culture methods

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Health benifts of banana

Bananas consist mainly of sugars (glucose, fructose and sucrose) and fiber, which makes them ideal for an immediate and slightly prolonged source of energy. It is also used in Reducing depression (Queenly N Y Lee january8th – 2010), Anemia, Constipation and Diarrhea: (Queenly N Y Lee january8th – 2010)Eyesight Protection, Healthy bones, Stress Relief: (Queenly N Y LEE JANUARY 8TH – 2010)Mosquito Bites and Stroke Risk

MATERIALS AND METHOD

Explant collection

G9 is a type of plant which was collected from chembrankulam ottanchatram TamilNadu.
Media must be maintained at 5.7 pH

Explant preparation

- From the mother banana plant, stems (suckers) are taken.
- From the suckers outer layers were removed.
- The sucker size was reduced into the bottle size and the bottom part was spliced off.
- Spliced meristem was taken for the further sterilization procedure.

MEDIA PREPARATION

Macro nutrient preparation

Macro nutrients	Stock solution for 1litre	For 50 litres	Dissolved media	Volume to be taken for 1l of media
Ammonium Nitrate	1650mg	82.5g		
Potassium Nitrate	1900mg	95.0g		
Calcium Chloride	440mg	22g	Dissolved in 5 litre	100ml/litre
Magnesium Sulphate	370mg	18.5g		
KH2Po4	170mg	8.5g		

Micro nutrient preparation

Micro nutrients	For 1litre	for 50 litres	Dissolved volume	Volume to be taken for 1 l of media
Boric acid	6.2mg	310mg		
Potassium Iodide	0.83mg	41.5mg		
Manganese Sulphate	22.3mg	1115mg	Dissolved in 1 litre	
Zinc Sulphate	8.6mg	430mg		20ml/litre
Copper Sulphate	0.0025mg	0.125mg		
Coboltous Chloride	0.0025mg	0.125mg		
Sodium Molyphate	0.0025mg	0.125mg		

Preparation of vitamins

Vitamins	For 1 litre	For 50 Litre	Dissolved volume	Volume to be taken for 1l of media
Nicotinic acid	0.5mg	25mg		
Pyridoxin	0.5mg	25mg	Dissolved in500ml	10ml/litre
Thiamine	0.5mg	25mg		
Meso inositol	100	5g		
Glycine	2mg	100mg		

Surface sterilization procedure

- Initially spliced meristem was washed with the tap water.
- Washed in 2 to 3 drops of Tween20 and maintained in Tween20 for 20 minutes. After the tween20 wash, meristem was treated in the fungicide for 20minutes.
- Fungicide treated meristem was taken immediately into the chamber before that chamber was surface sterilized with 70% of ethanol.
- Now sterilized meristem was transferred into the MS gel medium under aseptic condition.
- Bottle containing the meristem was screw capped tightly and it was covered with thin film and it was placed inside the plant growth room (PGR) for incubation.
- During incubation care must be taken to check contamination and growth must be observed.
- After 3 to 4 days initiation starts
- Initiation can be measured by the colour change in the meristem.

Different composition of multiplication media

- MS +5mg BAP + Normal agar
- MS + 5mg BAP + 20mg Ascorbic Acid + Agar
- MS + 5mg BAP + Suspension
- MS + 5mg BAP + 0.5mgNAA + Normal Agar
- MS + 5mg BAP + 0.5mgNAA + Gelrite
- MS + 5mg BAP + 0.5mgNAA + Suspension
- MS + 5mg BAP + Gelrite.

Multiplication of banana

- After initiation, grown plants were removed from the media using sterilized forceps.
- Leaf parts were spliced off.
- Clumps were inoculated in different compositions of media under aseptically condition.
- Bottles were covered with thin film and kept inside the tissue culture room
- Good multiplication was obtained in MS + 5mg BAP + 20mgAscorbic acid + agar

Standardization of photoperiod for multiplication

Multiplication was done at different photo period to standardize the multiplication rate, plantlets were illuminated at 6, 8, and 12 hours but only at 8 hours illumination optimum growth was obtained.

Standardization of rooting media

- Rooting was the third step after initiation and multiplication.
- Plants which were grown in the multiplication process were removed and inoculated inside the rooting media
- Transplantation was done aseptically using sterile knife and forceps.
- Charcoal agar media and MS+ Hormones and MS + charcoal agar media was used for rooting
- Bottles were screw capped tightly
- Covered with thin film
- Incubated in the plant growth room for the induction of root.

Standardization of charcoal

- Media containing different percentage of charcoal was taken and inoculated with the proliferated plants in multiplication step.
- Best result was obtained in the media containing 2% of charcoal.

Standardization of charcoal % for rooting

- Rooting was done at different illumination period (8,10, and 12 hrs)

- Optimum result was obtained at 12 hours.

Primary hardening

- Exagar plants were taken and transplanted inside the pro trays containing the soil and coco beads which was present in the ratio 2:1
- Plant was supplemented with nutrients and fungicide to nourish the plant.

Rooting media standardization

S.No	Media composition	Rooting
1.	MS+1ml NAA	70%
2.	MS+1ml IAA	50%
3.	MS+1ml IBA	40%
4.	MS+ 2gm CHARCOAL	95%

Charcoal percentage standardization for rooting

Media composition	%	Result
MS + charcoal	0.5	0
MS + charcoal	1	60
MS + charcoal	2	95

DISCUSSION

In the present work surface sterilization was done, media standardization for multiplication and rooting, standardization of photoperiod for multiplication and rooting was done. Initiation of explant was done to select the aseptically well initiated meristem for further micropropagation technique. Initially surface sterilization and standardization of mercuric chloride was done. Optimum growth was obtained at 5minutes treatment. Well initiated explant was inoculated into the medium for multiplication. Standardization for multiplication was done from the third subculture of the proliferated plantlets because multiplication procedure alone takes 3 months. This is due to gestation period. Hence the multiplication process was started with the existing plants in the multiplication stage Dirt and the leaf parts were removed and they were inoculated into different composition of multiplication medium in which best results were obtained in the medium containing BAP + ascorbic acid + agar medium. In the standardization of photoperiod 8 hours of light and 6 hours of dark yielded the maximum multiplication of plantlets with adequate shoot elongation. In the rooting media addition of 2% of charcoal instead of auxin (IAA, NAA, IBA) showed successful ratio. Rooting was done at 12 hours of photoperiod which resulted in optimum growth. Rooted plants were taken from the bottles (exagar plants) and were washed thoroughly to remove the agar particles in their root. Exagar plants were inserted inside the pro trays containing soil and coco beads. It was exposed to 26 degree of temperature and 80-90 % of moisture. The following procedure is simply known as primary hardening

Conclusion

- The standardized multiplication media can be used to produce maximum number of plantlets in large scale.
- Usage of charcoal and agar other than rooting hormones and gelrite will reduce the cost of media.
- The yield of the plantlets with high quality and higher yielding will be obtained when compared to the conventional method.

Future work

- Primary harden plants must be transplanted for the secondary hardening.
- Then it must be transplanted to the nurseries from where it can be marketed.

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