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In-vitro shoot induction of Vigna radiata with 6-Benzene adenine

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ABSTRACT

The present project in vitro induction of shoot in Green gram (Vigna radiate L.Wilczek) was carried out with explants , seeds of Vigna radiata. Explants was tested against different concentration of BA (6-Benzene adenine) on MS media. Observation was recorded after two weeks in terms of elongation of shoots from seeds of Green gram . The effect of different concentration of cytokinin was examined for development of shoots from seeds. BA at 0.5 mg/lit was found to be best treatment for development (elongation) shoots from seeds. From this work the optimum concentration for shoot induction one should used 0.5mg/ lit BA along with MS medium while comparing with the literature.

Key Words: In-vitro,6-Benzene adenine, M.S., cytokinine,. Vigna radiata

INTRODUCTION

Mung beans, often known as green grams, are grown all over the world and are one of the most significant pulse crops in India (Vigna radiata(L.) . The crop is frequently used as fodder in addition to its direct usage as protein-rich edible seeds and sprouts high in vitamins and amino acids . Yet, a variety of stressors in various regions of the nation significantly lower grain output and quality. Though yield losses are significant when exposed to various stress conditions, conventional research have not provided much information regarding its degree of stress tolerance, despite its significant economic value. However, by creating transgenic green gram plants, we can use genetic engineering techniques to produce any form of stress tolerance.

Yet by creating transgenic green gram plants with various resistant genes, we can use genetic engineering techniques to create any form of stress tolerance (Yadav et al., 2012). Genetic transformation techniques, however, need the first development of an in vitro regeneration process. In an attempt to increase the rate of multiplication, micropropagation is a substitute for the traditional technique of vegetative propagation. To achieve high clonal fidelity and efficiency, cotyledonary node multiplication is one of the explants that receives the majority of attention in green gram in vitro micropropagation (Khatun et al., 2008).

MATERIAL AND METHOD

The present investigation entitle "In vitro induction of shoot in green gram (Vigna radiata L.)was conducted in the Department of Biotechnology, B.N.N Collage, Bhiwandi. The experiment material comprises of *Vigna radiata*. The seed of green gram were obtained from nearby local market.

Glasswares: All the glass where used of good quality borosilicate glass (regesistant to heat.All the required glasswares were first washed with tab water. Allow this glass in solution of detergent for 2 hours. Then again washed with the water, allow then for 24 hours socking in dilute nitric acid. On following they were thoroughly washed with tap water to remove traces of nitric acid and rinse with double distilled water. Dry in oven at for 2 hours. The sterilization of glassware was recommended by autoclaving. All the dried beakers and petriplates were rapped in brown paper. Also forceps, were rapped in brown paper. All these were first autoclaved at 121°C for 15 minutes/151b pressure During inoculation forcep again sterilized by dipping in absolute alcohol and holding on the flame alternatively. Media Preparation--. The media was prepared with double distilled water,according to the composition given by Murashinge and skoog (1962).

After autoclaving place the test tubes in a tray, so that butts were obtained. Check the contamination for 24 hours and then proper explant was inoperated.

- **Preparation and inoculation of explants-**

All the aseptic operations were performed in a laminar air flow cabinet.

A. Explant prepration and surface sterilization-

1. Seed were washed thoroughly with double distilled water then seeds ware washed with 70% alcohol for 5-7 seconds followed by washing with double distilled water.
2. Then seeds were surface sterilized with 0.1% mercuric chloride solution for 1-2 min.washed Thoroughly in sterile distilled water 3-4 times for 5-10 min each.

B. Inoculation-

After the explant (seed) sterilization they were inoculated in MS medium different Concentration of BA under aseptic condition the culture were grown in light using 16 hours Light photo period at 25 ± 2◦C with the relative humidity 60% along with incoulation of explant on plane MS agar

C. Induction of shoots in Green gram

For induction shoots in test plant, the composition of basal medium The procedure for sterilizations of glasswere s, media was described earlier seed was Culture on MS basal medium supplemented with different concentration of BA.

D. Different concentration of BA-

The concentration of BA are given in table the culture were incubated at 25±2°C under 16 hours light.

RESULT AND DISCUSSION

The present investigation were carried out with *Vigna radiata* (Green gram) to standardize the medium and explants of Green gram for induction of shoot . Seeds were prepared for dissection in aseptic condition. and seed was inoculated on MS media with different concentration of BA for induction of shoots, observation was recorded daily for shoots and on 5th day of inoculation initiation of shoot was observed and further for 2 weeks the observation was carried out. Table shows the different concentration of cytokinin on MS medium. It was found that BA (0.5mg/liter) proved the best treatment producing shoots, which is maximum among all the treatment.

NAME OF CYTOKININ	CONCENTRATION (mg/lit)	Observation	
		5 days	8 days
MS+BA	0	_	_
MS+BA	0.2	_	_
MS+BA	0.5	Initiation of shoot	Elongation of shoot
MS	--	_	_

FIG-1 : Initiation of shoot 5TH DAY OBSERVATION

FIG -2: Elongation of shoot8TH DAY OBSERVATION

The first treatment was without BA, with shows the no induction of shoots. the second treatment was BA (0.5mg/lit) shoots initiated after 5 days and elongation of shoots was observed after 8 days. It means the concentration of cytokinin (BA) increases from 0.5mg/lit, shoot initiated and also elongated. From this table it can be concludate that optimum concentration of BA should be 0.5mg/lit with MS basal media. From this present work ,it can be concludate that for initiation and elongation of shoots , the MS media should be supplemented with BA (0.5mg/lit).

Green gram is a plant species that belongs to the **Fabaceae** family and it is botanically known as **Vigna Radiata L. Wilczek** . This small crop is native to India, and it is widely cultivated in many countries. Large amounts of a Green gram are cultivated in India, China, Southeast Asia.

The present investigation also supports by earlier work done by Himabindu Y. et al., studies on in vitro induction of shoot in green gram obtained multiple shoot by using cotyledonary node as explant on MS medium and Gamborg`s medium supplemented with 0.5 mg/lit BA medium.

Aparna Priyadarshini Patra et al., studies on invtro induction of shoot in green gram obtained multiple shoot by using cotyledonary node as explant on MS medium supplemented with BAP (2.0 mg/1) in combination with 1.0 mg/1 kinetin. M.K. khatun et al., obtainted multiple shoot by using cotyledonary node as explant on MS medium supplemented with combination of BAP (1.0 and 5.0 mg/lit).

S.K.Yadav et al., studies on in vitro induction of shoot in green gram obtained multiple shoot by using cotyledonary node as explant on MS medium and Gamborg`s medium supplemented with (2.0 mg/L) BAP.

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