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Hyacinth Pea (Phaseolus Vulgaris) In-Vitro Cultivation Of Shoot By Kinetin Action

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ABSTRACT :

The present work in vitro induction of shoot hyacinth pea (Phaseolus Vulgaris) was carried out with explants, seed of Phaseolus vulgaris.Explant was tested against different concentration of Kinetin on MS media. Observation was recorded after two weeks in terms of elongation of shoot from seeds of phaseolus vulgaris.

The effect of different concentration of Kinetin was examined for development of shoot from seed. Kinetin at 3mg/liter was found to be best treatment for development of [elongation] shoot from seed.

Key words: Kinetin,Hyacinth pea,elongation seed,MS media etc.

Introduction :

Hyacinth Pea **[Phaseolus Vulgaris]** has shown several agronomic, environmental economic advantages contributing to further improve the diets and incomes of peasant farming across Africa, Asia and South America hyacinth pea has its origin on the southern Africa region but has spread and is now cultivated in more than 100 countries. The environmental advantage of hyacinth pea arises from its ability to grow in semi-arid regions with low input requirements. Due to its recognized nutritional value high protein and low fat content, which is related to the prevention of diverse

World population has been growing explosively from 1.7 billion people at the beginning of 20th Century to now more than 5 billion and by beginning of new millennium close to 6.5 billion people is to populate our planet. Situation is getting worst in Asia particularly in countries like India and Pakistan where growth rate of population is much higher (Abbas, 1989).

Gamborg et.al [1968] reported an efficient protocol for plantlet regeneration from the cell suspension culture of hyacinth pea through somatic embryogenesis primary leaf derived, embryogeniccalli initiation in MMS | MS salts, Murashige and Skoog 1962] with B [vitamins] Medium. Fast growing embryonic cell suspension were established in 0.5 mg 2, 4-D, which resulted in the highest recovery of early stage of somatic embryos in liquid MS media.

MATERIAL AND METHOD :

The present investigation entitled "In vitro induction of shoot in Hyacinth pea (Phaseolus Vulgaris) performed in the Department of Biotechnology, BNN College. Bhiwandi. The experimental material comprised of hyacinth pea. The seeds of hyacinth pea were obtained from a nearby local market.

Treatments :

Isolation and sterilization of seed of both the leguminous crop were excised. Surface sterilization of the explants was carried out in the following steps: i. Explants were first washed on a running tap water. ii. They were treated with house hold detergent for five minutes. iii. They were washed again on a running tap water to remove all traces of detergent. iv. They were then washed with double distilled water in the laminar flow hood. v. They were further sterilized by dipping into 70% ethanol for 2 minutes. vi. They were then again washed three times with triple distilled water in the laminar flow hood to remove all traces of ethanol. vii. Seeds were treated with 0.1% of HgCl2 for 5 minutes. viii. They were finally rinsed 3 times with sterile triple distilled water to remove all traces of HgCl2 .

Washing and sterilization of glassware's-

All the required glassware's were first washed with tap water. Allow these Glassware's in solution of detergent for 2 hours. Then again washed with tap water. Allow them for 24 hours soaking in dilute nitric acid. On following day they were thoroughly washed with tap water to remove traces of nitric acid rinse with double distilled water. Dry in oven at 80°C for 2 hours. The sterilization of glasswares was recommended by autoclaving. All the Dried beakers and petri plates were wrapped in brown paper. Also,

Media preparation-

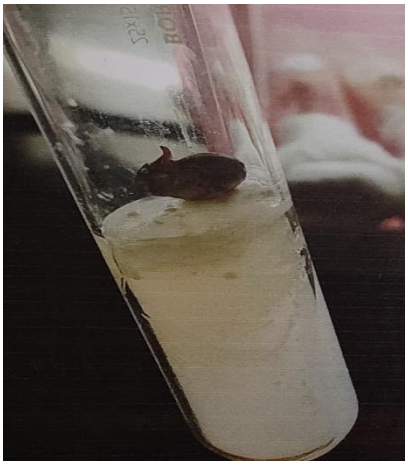
The media was prepared with double distilled water, according to the composition given by Murashige and Skoog (1962). _To prepare 1 lit of media take 50 ml of stock solution 1,5 ml each of stock solutions 2. 4. Desired quantities of different growth hormone were added accordingly The media was dissolved in double distilled water. Growth hormone with different concentration and 30% sucrose were added. The pH of the medium was adjusted as 5 8 agar was added at concentration of 10% and then the medium were steamed to melt the agar. The medium was distributed as 20 ml in Each tube. These tubes were then sterilized at 121°C for 15min/15 lb pressure in autoclave.After autoclaving, place the test tubes in a tray, so that butts were obtained Check the contamination for 24 hours and then proper explants was inoculated

RESULT AND DISCUSSION :

The present investigation was carried out with Hyacinth Pea [Phaseolus Vulgaris] to standardize the medium and explants of Hyacinth Pea for induction of shoot in Hyacinth Pea. The seeds were prepared for dissection in aseptic condition. And seed was inoculated on MS media with different concentration of Kinetin for induction of shoot; observation was recorded only for shoots after 7 days of inoculation to three weeks: Table shows the different | concentration of Cytokine on MS medium. It was found that Kinetin (3mg/lit). proved the best treatment producing shoot, which is maximum among all the treatment.

Table -Induction of shoot in the MS medium supplemented with KINETIN-

NAME OF CYTOKININ	CONCENTRATION	OBSERVATION	
		5 DAYS	9 DAYS
MS+KINETIN	0	----	----
MS+KINETIN	1	-----	----
MS+KINETIN	2	------	-----
MS+KINETIN	3	INITITION OF SHOOT	ELONGATION OF SHOOT



Effect after 5 day induction

MS+Kinetin

Effect after 9 day induction

MS+Kinetin

In vitro plant regeneration of Phaseolus has been reported by organogenesis (Malik and Saxena, 1991; Malik and Saxena, 1992; Ahmed et al. 2002) or through somatic embryogenesis (Zambre et al.1998; Schryer et al. 2005). Although several protocols have been described in the literature for bean regeneration,development of an optimal in vitro culture system still remains a major challenge since this and other species from the Phaseolusgenus, are recalcitrant for in vitro regeneration (Veltcheva et al. 2005).

The first treatment was without Kinetin, which shows no induction of shoot. The second treatment was with Kinetin [1mg/lit], which shows no induction of shoot As the concentration of Kinetin was increased [3mg/lit] shoot were initiated after 5 days and elongation of shoot was observed after 9 days. It means the concentration of cytokinin [Kinetin] increases from to 2 mg/lit, shoot initiated and also elongated. From this table it can be concluded that optimum concentration of Kinetin should be 3 mg/lit with MS basal media. From this present work, it can be concluded that for initiation and elongation of shoot, the MS media should be supplemented with kinetin 3mg/l

REFERENCE :

1. Abbas, S.T. 1989. Induction /screening of salt tolerance in Pakistani rice varieties Basmati-370, I R-6 and KS -282 through tissue culture. Ph.D. Thesis, University of the Punjab, Lahore.

2. Murashige, T and F. Skoog. 1962. A revised medium for rapid growth and bioassay with tobacco Tissue Culture. Physical Plant., 15: 473-478.

3. MALIK, Kamal A. and SAXENA, Praveen K.Regeneration in Phaseolus vulgaris L. Promotive role of N 6-benzylaminopurine in cultures from juvenile leaves.Planta, April 1991, vol. 184, no. 1, p. 148-150.

4. MALIK, Kamal A. and SAXENA, Praveen K.Regeneration in Phaseolus vulgaris L.: High-frequency induction of direct shoot formation in intact seedlings by N 6-benzylaminopurine and thidiazuron. Planta, February 1992, vol. 186, no. 3, p. 384-389

5. AHMED, E.E.; BISZTRAY, G.Y.D. and VELICH, I. Plant regeneration from seedling explants of common bean (Phaseolus vulgaris L.). Acta Biologica Szegediensis, 2002, vol. 46, no. 3-4, p. 27-28.

6. ZAMBRE, M.A.; DE CLERCQ, J.; VRANOVÁ, E.; VAN MONTAGU, M.; ANGENON, G. and DILLEN, W. Plant regeneration from embryo-derived callus in Phaseolus vulgaris L. (common bean) and P. acutifolius A. Gray (terapy bean). Plant Cell Reports, May 1998, vol 17, no. 8,p. 626-630.

7. SCHRYER, P.A.; LU, Q.; VANDERBERG, A. and BETT,K.E. Rapid regeneration of Phaseolus angustissimus and P.vulgaris from very young zygotic embryos. Plant Cell Tissue and Organ Culture, October 2005, vol. 83, no. 1, p.67-74.

8. VELTCHEVA, Margarita and SVETLEVA, Diana. In vitroregeneration of Phaseolus vulgaris L. via organogenesis from petiole explants. Journal Central European Agriculture, April 2005, vol. 6, no. 1, p. 53-58.