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Growth enhancement of Mat bean using In-vitro induction method by Naphthalene acetic acid

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Abstract
The present project in vitro induction of shoots in mat bean (Vigna aconotifolia) was carried out with explants, seeds of Vigna aconitifolia.Explants was tested against different concentration of Naphthalene acetic acid (NAA) on MS media. Observation was recorded after two weeks in terms of elongation of shoots from seeds of mat bean.
Moth bean produced nutritious seeds and green pod, leafy forage for hay or pasture and soil building "living mulch" to complaint orchard crops and to protect and improve fallow land. The pods, when young, are eaten as a table vegetable, the tiny beans they contain are high in protein and other nutrients and are a valued pulse for dry region
The effect of different concentration of auxin was examined for development of shoots from seeds. NAA at 1mg/lit was found to be best treatment for development (elongation) shoots from seeds.
From this work I would like to suggest the optimum concentration for shoots induction one should used 1mg/lit NAA along with MS media.
Key Words: Mat bean, Explant, *Vigna aconitifolia,NAA,MS media*

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I. Introduction

Vigna aconitifolia is a drought-resistant legume, commonly grown in arid and semi-arid regions of India.It is commonly called mat bean, moth bean, matki, Turkish gram or dew bean. The pods, sprouts and protein rich seeds of this crop are commonly consumed in India

Vigna aconitifolia, commonly known as moth bean or matki bean, belongs to Fabaceae, the third largest plant family. Moth bean is one of the most important crops among all the pulse crops of the world (Chopra and Swamy, 1975). It has a high level of tolerance to drought and heat among all Asian Vigna species. It can survive at temperatures up to 40-45 °C in its harsh natural habitat as it has evolved a few morphological and physiological features. It is a herbaceous, short-day crop having a deep and penetrating root system. Moth bean is an important source of proteins. Minerals, and vitamins and is also used in medicines and cosmetics. Therefore, it has been identified as one of the best food sources for the future. Moth bean, with its high tolerance to heat and drought, could be an excellent source of genes responsible for stress tolerance. There are no direct advanced technologies available that facilitate the crop production under extreme conditions. However, the development of stress-tolerant varieties and their inclusion in crop

improvement and breeding programs might be an optimistic approach. The genotypes RMO 40 and Jadia were considered as heat tolerant in earlier studies (Gurjar et al., 2014; Sharma et al., 2014; Harsh et al., 2016), although RMO 40 was reported to be drought susceptible by Sachdeva et al. (2016). Therefore, the genotypes with improved resistance and better performance under particular stress conditions need to be selected and studied for potentially important traits for use in breeding programs of moth bean.

The progress various fields of plant tissue culture has extensively been reviewed by several workers (Altmann et al., 1990; Bagde et al., 1998; Bajaj, 1986; Bhojwani and White, 1983; Nag and Badia, 1997.Avenido et al., 1991 in vitro propagation interspecific hybrids from a cross between vigna aconitifolia was successful when coytylebonary nobes of F1 hybrid seedling were cultured on an MS medium with 1.0mgL of Naphthalene acetic acid (NAA).

Anju and Jaiwal et al., 1992,1994 studies on in vitro cultures of mat bean showed that multiple shoots were successfully regeneration from the shoot tips and cotyledonary node of the seedling on MS medium with 0.5mgL and 1.0mgL of NAA. Cheg et al., 1992 and Jaiwal et al., 1992 obtained plant regeneration from cotylenonary node explants was observed on MS medium supplemented with 0.5mgL of NAA.Singh et al., 1994 obtained multiple shoots from shoot tip on MS media supplemented with1mg/L NAA, 0.5 auxin

II. MATERIALS AND METHODS

Plant Material and Seed Collection Cultivar of moth bean (Vigna aconitifolia) from local market of Bhiwandi
Explant Preparation Seeds were washed thoroughly with 5% solution of liquid detergent (Teepol) followed by rinsing with sterile distilled water several times. After washing, the seeds were treated with 0.1% HgCl2 for 2-3 minutes and then rinsed with sterile distilled water 3-4 times. Paper bridges were prepared with blotting paper strips taken in test tubes with their ends soaked in distilled water. These test tubes were autoclaved at 1.05 kg/cm2 pressure for 15-20 minutes. Surface sterilized seeds were aseptically inoculated on the paper bridges. Leaves, hypocotyls and roots were excised from these aseptically grown 5-7- days-old seedlings taken as explants.
Inoculation Inoculation of explant on MS media (Murashige and Skoog's 1962) was carried out in a sterile laminar airflow cabinet (horizontal type). The working bench was thoroughly cleaned with cotton swab dipped in rectified spirit (alcohol). Irradiation with ultraviolet light for 45 minutes was done prior to use by keeping sterilized surgical instruments like forceps, spatula, scalpel, culture media, sterile distilled water, autoclaved Petri dishes, spirit lamp in laminar air flow.
Different concentration of NAA –The concentration of NAA are given 0.4 and 1mg/L The culture were incubated at 25+ 2°C under 16 hours light. After 5 days initiation of shoots were formed and after 10 days elongated shoots were developed.

Leaves, hypocotyls and roots excised from 5-7-days-old in vitro grown seedlings were transferred to the culture media with the help of forceps in the vicinity of spirit lamp. After inoculation, culture vessels were labeled and kept in culture chamber. The cultures were maintained under aseptic uniform conditions of temperature 26° ± 2°C, relative humidity 55% and diffused light (3500 lux). For callus induction, different explants were inoculated on the medium containing various concentrations of growth regulator under study. Initially cultures were examined everyday upto 10 days and finally growth responses were recorded weekly. Desired callus obtained during primary culture was subsequently transferred to suitable fresh medium after 6-8 weeks of culture.along with MS media..

III. RESULT AND DISCUSSION

The present investigation were carried out with vigna aconitifolia (Mat bean) to standardize the medium and explants of Mat bean for induction of shoots in vigna aconitifolia.
Seeds were prepared for dissection in aceptic condition. And seed was inoculated on MS media with different concentration of NAA for induction of shoots, observation was recorded only for shoots after 5 days of inoculation to 2 weeks.
Table no.2 shows the different concentration of a on MS medium. It was found that NAA (1 mg/liter) proved the best treatment producing shoots, which is maximum among all the treatment.

Table.- Induction of shoots in the MS medium supplemented with NAA-

NAME OFAUXIN	CONCENTRATION (mg/lit)	Observation	
		5 days	10 days
MS + NAA	0	-	-
MS + NAA	0.4	-	-
MS + NAA	1	Initiationof shoots	Elongation ofshoots

The first treatment was without NAA, with shows the no induction of shoots. The second treatment was NAA (0.4mg/lit) with shows the no induction of shoots. As the concentration of NAA increase (1mg/lit) shoots initiated after 5 days and elongation of shoots was observed after 10 day. It means the concentration of auxin (NAA) increases from 0 - 1mg/lit, shoots initiated and also elongated. From this table it can be concluded that optimum concentration of NAA should be 1 mg/lit with MS basal media. From this present work, it can be concluded that for initiation and elongation of shoots, the MS media should be supplemented with NAA (1 mg/lit).

In support of the present investigation liturature of Shekhawat and Galston (1982) established tissue culture of this plant from leaf and stem explants on Gamborg's B5 (Gamborg et al., 1968) medium supplemented with 2,4-D and kinetin.

Advancements in the field of agricultural biotechnology have generated a considerable optimism for the future growth and development of agriculture to meet the increasing needs of the world in the coming decades

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