

IN VITRO REGENERATION OF BRAHMI (*BACOPA MONNEIRI* (L.) PENN.) - A THREATENED MEDICINAL PLANT

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ABSTRACT

In vitro regeneration of memory enhancing plant *Bacopa monneiri* was observed by using nodal explant. In present study, shoot initiation was best seen in MS media in 3mg/l BAP concentration. Combination of BAP and IAA (1mg/l+3mg/l concentration) was best suited for root initiation. The rooted plantlets were hardened and successfully established in soil.

Key Words: *Bacopa monneiri* (L) Penn., brahami, BAP, *in vitro* culture

INTRODUCTION

A rich heritage of knowledge on preventive and curative medicine is available in Indian books such Atharva Veda, Charkha, Sushruta etc. Over 25,000 plants are used in Ayurveda system to cure and relief many physical and mental diseases. In India the plant is used for all sorts of skin problems- eczema, psoriasis, abscess, ulcerations and diseases of the nervous system (Shakoor *et al.* 1994). *Bacopa monnieri* (L) Penn. is popularly known as brahami. It is a perennial, creeping herb whose habitat includes wetlands and muddy shores. *In vitro* techniques such as tissue and organ culture offer plant breeders new openings for clonally propagation, genetic manipulation and production of inbred lines supplementing the routine vegetative production rare plant species. Using the *in vitro* method, a million – fold increase per year in clonal multiplication over conventional methods is possible. This study highlights the effect of different plant growth regulators on shoot proliferation in water hyssop with special reference to *B. monnieri*.

The demand of *Bacopa* is met from natural population generally, which can lead to put heavy strain on existing natural population and hence resulting in slow depletion of this already threatened herb. Tissue culture techniques can be used to attain rapid multiplication of elite clones and germplasm conservation of *B. monneiri*.

MATERIALS AND METHODS

The explants *Bacopa monnieri* were collected from A-minor (A water tributary of Gang Canal) adjoining Sriganaganagar, Rajasthan, India. The twigs with node explants were washed in running tap water and then washed again thoroughly by adding a few drops of teepol (5%) for 20 minutes, surface sterilized with 0.1% (w/v) mercuric chloride (HgCl₂) for 3-5 minutes. Finally the explants were repeatedly washed for 3 to 5 times in distilled water (Dubey, 1999) to remove the impurities and stubborn particles attached to the explants. Nodal segments about 0.5-0.8 cm were prepared aseptically and were implanted

vertically on MS medium fortified with specific concentrations of growth regulators (BAP and IAA) singly or in combination adding 30 g/l sucrose and 0.8% agar. The hormones used for experiment were taken from stock solutions, which were previously prepared and kept under cold condition in refrigerator (Doods and Roberts, 1985). The pH of the medium was adjusted to 5.7 with 0.1 NaOH before autoclaving at 15 lbs and 121°C for 18 min. The cultures were incubated at a constant temperature of 26±1°C with 16 h photoperiod (3000 lux).

RESULTS AND DISCUSSION

Nodal explants were incubated on MS medium fortified with different concentrations of BAP (1, 2, 3, 4 mg/l) IAA combination (Table 1; Fig. 1, 2). The shoot initiation was observed after one week of inoculation in all sets. Among the set 90% of shoot regeneration was observed in 3mg/l of BAP concentration. It was also observed that root formation was best at 40% at combination of BAP and IAA at 1mg/l and 3mg/l concentration respectively. One of main function of exogenous cytokinin in tissue culture is induction of adventitious shoots. Results of this study indicate that large scale propagation of *B. monneri* by tissue culture is feasible & several plantlets can be regenerated from one nodal explant. Present study indicate that an effective cytokinin for shoot induction as well as shoot propagation in *B. monneri*. Generally BAP is employed for regeneration of shoot. Several studies also showed that media supplemented with NAA & BAP have also useful for production of shoots (Mc Pheeters *et al.*, 1980, Janson & Borman 1980). In present MS supplemented IAA in combination with BAP was useful for regeneration of shoots. The present study provides an effective high frequency micro propagation protocol.

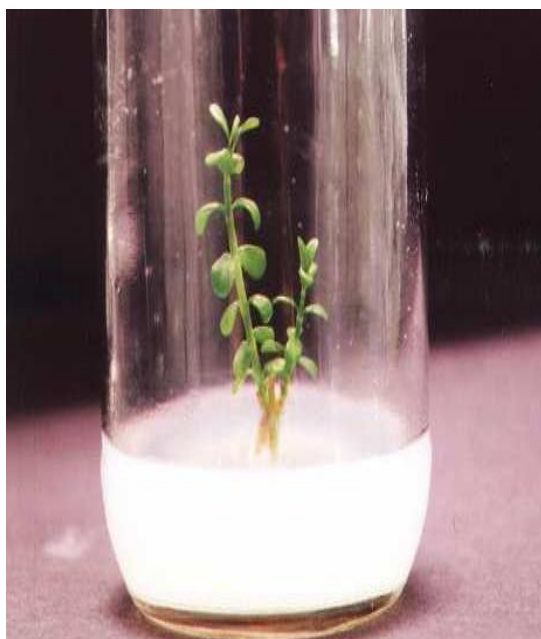


Fig. 1: Effect of BAP on shoot formation nodal explants



Fig. 2: Effect of BAP and IAA on root of formation of nodal explants

Table 1: Effects of different concentrations of cytokinin and auxin in MS medium on *in vitro* shoot proliferation from nodal segments of *Bacopa*.

Hormone concentration (mg/l)	% of explant showing shoot proliferation	% of explant showing root proliferation
BAP (1 mg/l)	50%	-
BAP (2 mg/l)	60%	-
BAP (3 mg/l)	90%	-
BAP (4 mg/l)	60%	-
BAP + IAA (1 mg/l + 1 mg/l)	50%	10%
BAP + IAA (1 mg/l + 2 mg/l)	60%	15%
BAP + IAA (1 mg/l + 3 mg/l)	90%	40%
BAP + IAA (1 mg/l + 4 mg/l)	70%	20%

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