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INVESTIGATION OF GROWTH FACTORS EFFECT ON IN *Vitro* CULTURING OF *Solanum melongena*

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ABSTRACT

Brinjal (*Solanum melongena*) is the second most important vegetable crop grown after potato in Asian countries and in various tropical and temperate parts of the world. It also plays a vital role in the National economy as a cash crop. Its tissues possessing a high morphogenetic potential that is useful for developmental studies as well as for establishing biotechnological approaches to produce improved varieties, such as transgenic plants. Here, stem and leaf explants were inoculated on MS medium with different hormones and different ratio for optimization of in *vitro* regeneration. Callus was initiated within a period of 10-12 days of culture and a mass of callus was formed in vivo and in *vitro* explants. In BAP: NAA 2.0mg/L:0.5mg/L, the callus induction started from the 12th day of inoculation. After 14 days, white hairy growth appears from light greenish callus which indicate the formation of root like structures. Callus induction was obtained from another hormone combination 2mg/L:0.5mg/L NAA: BAP .The Kinetin (2 to 3 mg/l) and BAP (2 to 3 mg/l) alone used for multiple shoot initiation. The shoot was initiated within a period of 4-5 days of culture from nodal explants. Even we have been used above 3 mg/L Concentration of NAA and IBA, Concentration of 2 - 3 mg/L NAA and IBA have been showed the optimal shoot initiation.

KEYWORDS : Benzylaminopurine (BAP), Indole-3-acetic acid (IAA), Indole-3-butyric acid (IBA), Kinetin (KIN), 2, 4-dichlorophenoxyacetic acid (2,4-D), Murashige and Skoog medium (MS), Naphthylacetic acid (NAA)

Eggplant tissues possessing a high morphogenetic potential that is useful for developmental studies as well as for establishing biotechnological approaches to produce improved varieties, such as transgenic plants (Scoccianti et al, 2000). In vitro culture techniques are widely used for the improvement of various crops such as Bt (*Bacillus thuringiensis*) transgenic brinjal. Callus culture can induce genetic and epigenetic changes in the regenerated plants. Eggplant has been divided into three main types egg-shaped (*S.melongena* var. *esculentum*); long slender shaped (*S.melongena* var. *serpentium*) and dwarf type (*S.melongena* var. *depressum*) (Kalloo, 1993).

In *vitro* culturing mainly depends on the culture medium and hormones. There are different medium used in the plant tissue culture such as MS medium (Murashige and Skoog, 1962), White's medium (White, 1963), LS medium (Linsmaier and Skoog, 1965), B5 medium (Gamborg et al., 1968), Nitsch medium (Nitsch, 1969) and Woody plant medium (Lloyd and McCown, 1981) which are permanently applicable for the plant tissue culture. Intra and inter specific Solanum hybrids were multiplied in *vitro* through somatic embryogenesis and adventitious organogenesis was reported (Mohammed Ali et al., 1991). Induction of callus and plantlet formation from the culture of isolated microspores of eggplant was experimented (Miyoshi, 1996). The shoot of eggplant was regenerated by inducing the plant with thidiazuron (Magioli et al., 1998).

MATERIALS AND METHODS

MS basal Media (Sucrose 30 g/L, myo-inositol 100 mg/L and pH 5.6 to 5.8) supplemented with different growth regulators at different concentrations were used depending on the purpose of the individual experiment. Media and apparatus were rendered sterile by autoclaving at 15 lbs/inch^2 (121°C) for 15 minutes. Surface sterilized (1% Sodium hypochlorite) stem and leaf explants have been used for the inoculation containing MS medium with different growth supplements. BAP : NAA combination have been used for callus induction. BAP alone used for shoot initiation. Kinetin has also been used for shoot initiation. The Kinetin:IBA combination have been used for the micropropagation of node. NAA have also been used for the rooting of *Solanum melongena*. All culture have been maintained at 25 ± 2^{0}C, uniform light (1000 lux) provided by fluorescent tubes (72000K) over a light/dark cycles of 16/8 hours.

RESULTS AND DISCUSSION

Concentration of IBA at 2mg/L showed a normal and elongated single rooting from shoot explants. When

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Figure 1 : Rooting After 8 Days of Inoculation - IBA 2.0 mg/l

Figure 2 : Callus Induction at NAA:BAP 2.0:0.5 mg/l

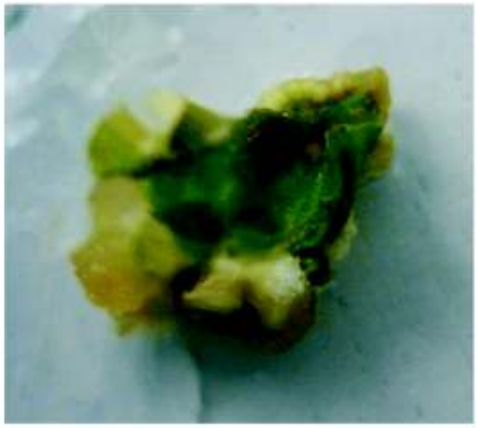
IBA concentration was increasing above 2.0mg/L or concentration was decreasing below 2.0mg/L no rooting was observed. Among the concentrations Kinetin : IBA combination, 2.0:0.5 mg/L concentration have been showed a normal and elongated single root observed after 7 - 9 days of inoculation(Figure 1).

Callus developed from the leaf explants of *Solanum melongena* on MS medium supplemented with growth regulators. Cytokinin in combination with auxins was found effective in callogenesis. Interestingly, explants cultured on MS medium in combination with BAP at 2.0 mg/L and NAA at 0.5 mg/L induced calli of *Solanum melongena*. Due to the effect of hormones Cytokinin and Auxin, the cells were dedifferentiated into calli then it differentiated into root cells. Among Combination of BAP: NAA and 2, 4-D: BAP, the BAP: NAA combination had effective cell growth and cell elongation during the calli induction on *Solanum melongena*. Although the hormone combination is greatest way to initiate the callus in short period of time, the optimum concentration of hormones also important in in *vitro* studies (Figure 2).

As *Solanum melongena* is an important economical crop plant, study of in *vitro* propagation support to produce improved varieties of *Solanum melongena*. In *vitro* propagation through axillary bud proliferation has been the safest and faithful strategy to maintain genetic integrity of the developing progenies. In the present study, it was observed that MS medium with BAP at 2.0 mg/L was the most efficient for shoot induction of *Solanum melongena* (Figure 3). Due to the effect of hormone BAP, the cells were elongated to initiate shootlet. Among BAP and Kinetin, the BAP has the more efficient growth of cells and elongation than Kinetin. Shoot regeneration has been reported among various plants of Solanaceae. As in the present study, delayed shoot bud regeneration was observed in BAP at 3.0 - 4.0 mg/L and it shows stunted growth. Hence, we can conclude that the optimum concentration of hormones differ to different specious of plants due to the genetic specificity of each individual to initiate the shoot in *vitro* condition.

Callus induction after 6 days of inoculation - BAP:NAA 2.0:0.5 mg/l

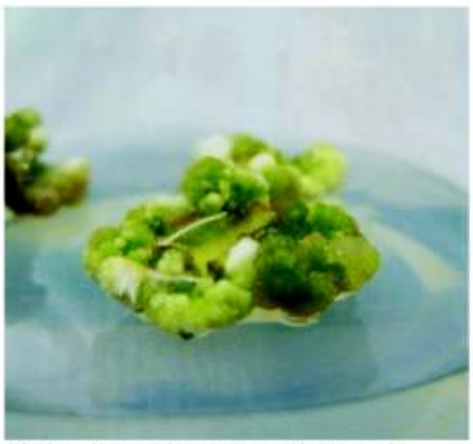
Callus induction after 12 days of inoculation - BAP:NAA 2.0:0.5 mg/l



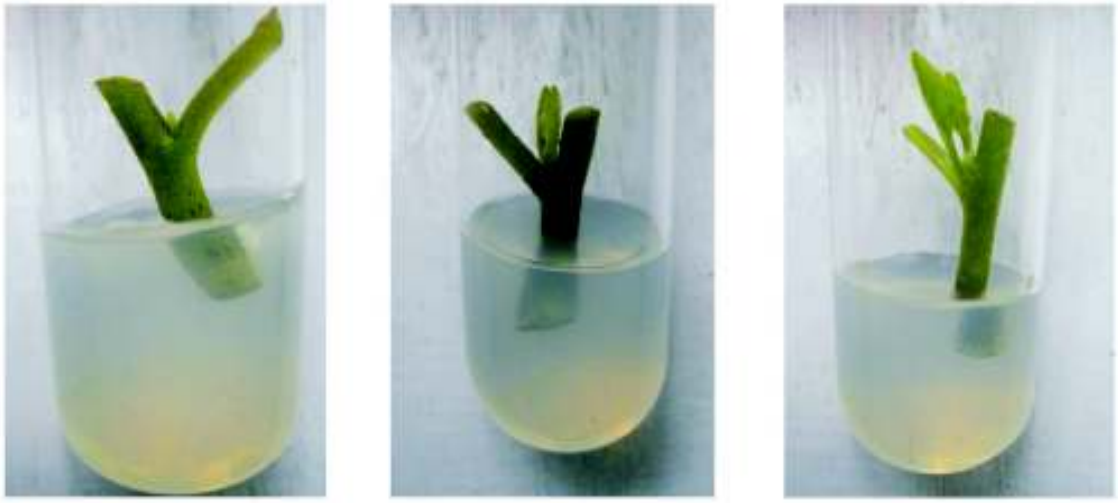
Callus induction after 15 days of inoculation - BAP:NAA 2.0:0.5 mg/l

Callus induction after 18 days of inoculation - BAP:NAA 2.0:0.5 mg/l

Callus induction after 20 days of inoculation - BAP:NAA 2.0:0.5 mg/l



Callus induction after 25 days of inoculation - BAP:NAA 2.0:0.5 mg/l



Shoot initiation after 5-8 days of inoculation -BAP 2.0 mg/l

Shoot initiation after 10-15 days of inoculation -BAP 2.0 mg/l

Shoot initiation after 8-15 days of inoculation -Kinetin 2.0 mg/l

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