ORIGINAL RESEARCH PAPER

Botany

TISSUE CULTURE STUDIES CALLUS TREATMENT ON STEM NODE EXPLANTS OF CITRULLAS VALGARIS, L.

KEY WORDS:

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ABSTRACT

The plant tissue culture methods also provide base for the improvement of crop to induce somaclonal variations, *In Vitro* mutations, genetic transformation of medicinally important genes and development of somatic hybrids plant regeneration protocol is required. Embryo genic callus induction and plant let proliferation of Solanum nigrum Venkateshwarlu M (2017). Shoot regeneration better than other cytokinins (Hussain *et al* 2007). Phyto chemical analysis of *Solanum Surattense* young leaves evauated for the presence of bioactive compounds using various polarity solvents petroleum (Venkateshwarlu *et al* 2018). The present study established reliable and reproducible protocol for rapid multiple shoot induction from node explants of *Citrullas Valgaris,* using different concentration and combination of cytokinins. Murashige and Skoog (1962) medium supplemented with 0.5 to NAA 2.0 mg/l BAP was found to be optimum to induce shoots directly from the stem node explants. Since very scarce information is available about micro propagation of this important medicinal plant, an attempt was made to develop a reproducible protocol for multiple shoot induction form stem node explants of one the tissue culture. Several workers in past have micro propogated some of the important Asclepiadaceae members such as *Cerogia bibosa* (Britto *et a*l., 2003), holostemma adakodien (Martin, 2002-2003). Significant increase in the number of shoots per explants was found ion M.S. medium supplemented with NAA, BAP and 14 mg/l adenine sulphate . All the tested combinations have little effect on increasing the number of shoots. Nodal explants derived shoot cultures were sub cultured to M.S. medium fortified with same concentration of hormone for shoot elongation. The percentage of explants exhibiting shoot induction was found to be between 50-60 i. Most of the concentrations of Benzyl amino purine tested except M.S medium supplemented with 0.5-2.0 mg/l benzyl amino purine. Stem segments are used as important explants for genetic transformation system, described in many plants species (Rastogia and Dwivedi, 2006)

INTRODUCTION:

The best results were obtained when stem node explants were initially cultured with (4.0mgl *l* BAP, 1.0mg/l NAA, 3.0mg/l) developed callus after 15-20 days of culture. *In Vitro* techniques using micro propagation and tissue culture offer a great possibility to overcome their problem. Micro Propagation using stem node explants. The auxin 2,4-D has been dermined as potent callus inducing phyto hormones in studies with many plant species. Various in the callus forming ability of different concentration of BAP, NAA and 24-D Table-I, Plate-I were placed in the medium to compare their growth responses. Among the tested NAA, Indeed the hight yield of callus followed by 2,4-D similarly. The callus so produced was green in colony and soft in texture presence of 2,4-D has been shown to be essential for callus formation. They observed that the maximum callus growth on MS medium containing 2,4-D in contrast to our present findings. The combination of Auxins and Cytokinins promote cellular differentiation and also organogenesis. In the medium concentration of BAP was increased up to 20-30 mg/l the multiple number of shoots Direct *In Vitro* plant propagation was achieved in pusa ruby cucutivar of Tamato (*Solanum lycoperiscan L* Mill)T. Ugender, Venkateshwarlu M (2018).

MATERIAL AND METHODS:

In the present investigation we present the result of our efforts to develop a protocol for plant regeneration through stem node explants in C*itrullus Valgaris L* a medicinally important plant. Stem node explants of different sizes were cultured with the induction medium consisting of MS Salts and Vitamins 6% Sucrose supplemented with BAP, 1.0mg/l to 4.0mg/l NAA (1.0mgl *l* to 3.5mg/l) and 2,4-D (1.5mgl *l* to 3.5mg/l). PH 5.7-5.8 The percentage of explants responding was evaluated after 4-6 weeks of cultures. The cultures were transferred to fresh medium after an internal of 6 weeks.

The nodal raised from control seeds could produce only callus on MS with different supplements which was regenerated into a single shoot. Well filled undamaged and uniform sized seeds were handpicked from the seed lot and equilibrated to the moisture content of 12 percent. For each dose of physical mutagen and random sample of 100 seeds were treated in *Citrillus Valgaris.* Explants were excised aseptically and were inoculated on the MS based medium supplemented by kinetin or BAP at concentrations ranging from 0.5 to 5 mg/l. Cultures were incubated under 10 h fluorescent light at 25 ± 2°C temperature

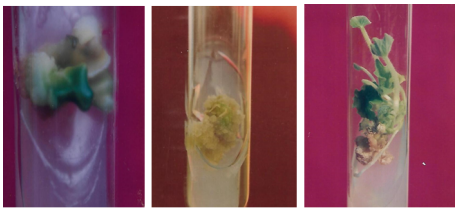
RESULTS AND DISCUSSION:

The maximum number of shoots on the explants were observed at 4.0 mg/l BAP or 5.0 mg/l kinetin, but at higher levels of BAP or kinetin, the formation of callus had taken place and the number of shoots per explant was reduced. The isolated *in vitro* raised shoots of 1-2 cm long, rooted profusely on MS medium with BAP (2 mg/l) + NAA (1 mg/l) within 15 days resulting in the formation of complete plantlets.

Table-1 . Tissue culture studies on stem node explants of *Citrillus Valgaris.*

% of Growth regulators (mg/l)	*Stem Node*	
	Mean number of shoots per shoot tip	*% of callus production*
MS + 1.0 BAP + 1.0 NAA+Kn2,4-D	10.3± 2.2	*44*
MS + 2.0 BAP + 1.5 NAA+Kn2,4-D	9.3 ± 2.4	*42*
MS + 3.0 BAP + 2.0 NAA+Kn2,4-D	6.2 ± 1.6	*30*
MS + 4.0 BAP + 3.0 NAA+Kn2,4-D	1.4 ± 0.2	*35*
MS + 5.0 BAP + 3.5 NAA+Kn2,4-D	6.6 ± 1.3	*25*
MS + 2.0 BAP + 1.0 L-glutamic acid	12.4 ± 1.0	*20*
MS + 3.0 BAP + 1.0 L-glutamic acid	15.6 ± 0.5	*18*
MS + 4.0 BAP + 1.0 L-glutamic acid	*14.4 ± 0.4*	*15*

Plate 1. Tissue Culture studies on stem node explants of Citrullus Valgaris L



CONCLUSION:

The stem node explant transferred to a fresh medium containing the some concentration of growth regulators, again resulted in the formation of multiple shoots.

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