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**ORIGINAL ARTICLE** 



# Direct Organogenesis from nodal explant of *Solanum* elaeagnifolium Cav.

**S. Balavivekananthan, T. Francis Xavier, S. R. Senthil Kumar and R. Sabitha** PG and Research Department of Botany, St. Joseph's College (Autonomous) Affiliated to Bharathidasan University, Tiruchirappalli, Tamil Nadu – 620 002.

Email: vivek02ayr@gmail.com

#### ABSTRACT

In this present study reveal that the suitable protocol was devised for an in vitro micropropagation of Solanum eleagnifolium L. belongs to solanaceae member. It has been highly used for many medicinal values. The 3 cm lengths of nodal explants were selected for the shoot development. The selected explants were culture on murashige and skoog (MS) medium supplemented with different concentration of growth hormones Cytokininsviz BAP and kinetin (1.0, 2.0, 3.0, 4.0 and 5.0 mg/l) is used for shoot proliferation and elongation. The best shooting was obtained in 3.0 mg/l, since it gave the highest percentage of shoot regeneration 95% and a number of explants responded (19/20). Whereas the auxins viz IAA and IBA combinations at different concentrations (0.5 + 1.0 mg/l, 1.0 + 1.5 mg/l, 1.5 + 2.0 mg/l, 2.0 + 2.5 mg/l, 2.5 + 3.0 mg/l) for root induction. Among various concentrations of IAA+IBA combinations tested, 1.0+1.5 mg/l was the more effective concentration for rooting of shoots in Solanum elaeagnifolium. The plantlets with well-developed root systems were gradually acclimatized to greenhouse using green manure with soil and take out to polyhouse. **Keywords:** Auxin, Cytokinin, Plant growth hormones, Shoot induction, Solanum elaeagnifolium CAV.

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# INTRODUCTION

In the plant Kingdom *solanaceae* is one of the largest families about 33 species out of 8 genera. This family is indigenous to Egypt, distribution in different localities [12]. Many of the medicinal plants are from the Solanaceae family [6]. *Solanum elaeagnifolium* is a perennial herb and also the common weed of the western North America [10]. It is also called as Silver leaf nightshade which classified as a toxic or poisonous plant both to the cattle and the humans. However, some birds feed on the fruits. This plant is rich in solanine, a poisonous glycoalkaloid that causes gastrointestinal, neurological, and coronary problems including emesis, stomach pains, dizziness, headaches, and arrhythmia [4]. Glycoalkaloids from members of the nightshade family have been shown to be effective in variety of medical applications, including limiting growth of certain cancer cells and treating herpes complex viruses. Plant tissue culture is a great device in the large cultivation and propagation of rare as well as medicinal plants [2][6][11]. In recent years, there has been a great interest among breeders in biotechnological methods including *in vitro* culture, which can accelerate and intensify the breeding process [15]. The present investigation of this study is to developing an initiation and induction of shooting and rooting of *Solanum elaeangifolium* which can possess the production and propagation of the medicinal plant by an alternate and cost effect approach.

### MATERIALS AND METHODS

### **Plant Material**

The plant material Solanum eleagnifolium Cav. is a medicinal plant belongs to solanaceae family. It was collected from pithalaipatty, dindigul district of Tamil nadu. The collected plant material was authenticated by Dr. S. Soosai Raj, Taxonomist, Department of Botany, St. Joseph's College (Autonomous), Tiruchirappalli.

# Method

The present investigation has been made to propagate plants from node, inter node, leaf, and shoot tip explants of *solanumelaeagnifolium* Cav. growing in the field for the in vitro experiments.

# Sterilization

Different plant parts such as shoot tip, nodal, internodal, and leaf were used as explants and they also contained the source of contamination. The explants were thoroughly washed with tap water for 15-20 minutes remove the soil particles and other extraneous fine particles and rinsed with 1% of teepol solution for 2 minutes. The explants were surface sterilized with 1% of bavistin for 2 minutes. Then they were rinsed in distilled water thrice and then taken into the laminar air flow chamber, they are surface sterilized with 0.1% of HgCl<sub>2</sub> for 2 minutes. They were again washed 5 times using sterilized water.

#### **Media preparation**

The nutrient media were used for the present investigation, they are Murashige and Skoog, 1962 medium along with the various hormone compositions were used [9]. The chemicals used for the experiment include the macronutrients, micronutrients, vitamins, amino acids and hormones were obtained from Himedia laboratories. The sterilized explants were inoculated on the ms medium in laminar air flow chamber. Inoculated cultures were maintained at  $25 \pm 2^{\circ}$ C with a photoperiod of 16 hours light and 8 hours dark per day of fluorescent light (3000 lux) for all treatments. Subcultures were made once in 15-20 days.

The rooted plantlets were removed from the culture tubes and washed in sterilized distilled water. Then they were transplanted into cups containing sterilized vermiculite and soil (1:1). The plants need 95-100% humidity and therefore they were covered with plastic bags with perforation or holes. After 15 days, the plantlets in the cups were transferred to a shadow for about 30 days and then transfer to the field.

## **RESULTS AND DISCUSSION**

In this present study two different cytokinins viz. BAP and Kinetin were used. Nodal explants (1.0 cm length) of Solanum elaeagnifolium were cultured on MS medium supplemented with different concentrations of BAP and kinetin individually (1.0, 2.0, 3.0, 4.0 and 5.0 mg/l)). After 30 days of incubation shoot initiation was observed (Fig.3). The results indicate that, of the various concentrations of BAP alone supplied to MS medium significantly influenced the shoot proliferation. The percentage of shooting ranged from 55% to 95%. The best shooting treatment was 3.0 mg/l, since it gave the highest percentage of shoot regeneration 95%) and a number of explants responded (19/20) (Table -1and Figure - 1). In the present study BAP alone 3.0mg/l was found to be ideal for multiple shoot induction (Fig.3). The same response was observed in Moreinga pteryosperma [8] and Citrus sincensis [13] whereas the various concentrations of kinetin supplemented with MS medium significantly influenced the shoot proliferation and elongation. The percentage of shooting ranged from 60% to 90%. The best shooting treatment was 4.0 mg/l, since it gave the highest percentage of shoot regeneration 90%) and a number of explants responded (18/20) (Table -1 and Figure - 1) the culture medium devoid of growth regulators failed to stimulate bud break in all the nodal explants. The result indicates that the presence of cytokinin in the medium is required by the explants to respond which is in conformity with the result of Loh and Rao (1989); Amin and Jaiswal (1987) [1][7].

# Induction of Rooting on Regenerated shoots

The regenerated shoots from nodal explants were excised from *Solanum elaeagnifolium* and inoculated on MS medium supplemented with the combinations of IAA and IBA at different concentrations (0.5 + 1.0 mg/l, 1.0 + 1.5 mg/l, 1.5 +2.0 mg/l, 2.0 +2.5 mg/l, 2.5 +3.0 mg/l) for root induction (Fig.4). Among various concentrations of IAA+IBA combinations tested, 1.0+1.0 mg/l was the more effective concentration for rooting of shoots in *Solanum elaeagnifolium* (Table 2 and Figure - 2) Since it gave the highest percentage of rooting (86%) and higher number of explants were responded (6/7) But increasing concentration of IAA+IBA combinations showed a decreased trend in rooting. These results are in consonance with that of Anand *et al.*, (1997) and Usha and Swamy, (1998), who worked on *Kampfrenia rotunda* and *Artemisia annua* respectively [3][14]. The results indicated that IAA was found be suitable for root induction. The present study results also supported by the studies made by Jameel and Bahrany. (2001) where IAA +IBA combinations significantly influenced root proliferation in *Citrus aurantifolia* [5]. **Hardening:** 

# At the final stage of this present study the in vitro rooted plants were takeout from the culture tubes and transferred to the polythene cups filled with manure and the soil (1:1) then the plants get adjust to outer environment (Fig.4). Among all the transferred plants 75% were able to survive under field condition.

S.No	PGR	No of Explant Inoculated	No of Responded	% of Respoded	No of shoot per Explant
	BAP (mg/l)				
1	1	20	15	75	2±0.53
2	2	20	13	65	1.92±0.61
3	3	20	19	95	2.10±0.56
4	4	20	18	90	2.05±0.63
5	5	20	11	55	1.90±0.7
	PGR				
	KIN (mg/l)				
1	1	20	14	70	2±0.53
2	2	20	13	65	1.92±0.47
3	3	20	17	85	2.05±0.62
4	4	20	18	90	2.05±0.55
5	5	20	12	60	1.83±0.15

Table – 1: Effect of cytokinin (BAP & KIN) (mg/L) from nodal explant of *Solanum elaeagnifolium* 

Figure - 1: Effect of cytokinin (BAP & KIN) (mg/L) from nodal explants of *Solanum elaeagnifolium* Cav.



 Table -2: Root formation from nodal explants of Solanum elaeagnifolium Cav.

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S. No.	Plant Growth Regulators (PGRs)		No. of explants Inoculated / Response	% of root induction	No. of roots / shoot Mean ± SD
	IAA (mg/L)	IBA (mg/L)	Nodal	Nodal	Nodal
1.	0.5	1.0	3/7	43	8.0 ± 2.64
2.	1.0	1.5	6/7	86	$12.16 \pm 3.76$
3.	1.5	2.0	5/7	71	$4.4 \pm 1.14$
4.	2.0	2.5	4/7	57	4.25 ± 1.25
5.	2.5	3.0	3/7	43	$5.33 \pm 1.15$

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Figure - 3: The effect of different concentration of BAP on shoot induction from nodal explant of Solanum elaeagnifolium Cav.



d) – Multiplication of Shoot

a & b) – Initiation of Shoot,

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# Figure - 4: The effect of different concentration of IAA and IBA on root induction from nodal explant of *Solanum elaeagnifolium* Cav.



a & b) - Initiation of root, c) - Multiple roots d) - Hardening

#### CONCLUSION

The suitable protocol has been developed for *solanum eleagnifolium*. A high significant shoot multiplication was noted in nodal explants. Maximum percentage of shoot proliferation were reported in explants grow on ms medium supplemented with BAP (3mg/L) and kinetin (4mg/L) respectively. This protocol can be used to mass scale production.

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