



## MICROPROPAGATION OF *SOLANUM TRILOBATUM* FROM SHOOT TIP EXPLANTS

Dr. H. David Raja\*, K. Senthilarasu and Dr. D.I. Arockiasamy

Assistant Professor, GCIC, Department of Botany, St. Joseph's College, Tiruchirappalli – 600002, Tamil Nadu, India.

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**\*Correspondence for  
Author**

**Dr. H. David Raja**  
Assistant Professor,  
GCIC, Department of  
Botany, St. Joseph's  
College, Tiruchirappalli –  
600002, Tamil Nadu,  
India.

### ABSTRACT

*Solanum trilobatum* is a medicinally important perennial herb of the family Solanaceae. An efficient micropropagation protocol was established for the regeneration of *Solanum trilobatum* using shoot tip explants. Maximum multiple shoots (12 shoots/explant and 80% of response) were obtained from the shoot tip culture on MS medium supplemented with BAP (2.5mg/l). The shoots were rooted on MS basal medium supplemented with IBA (3.0mg/l). *In vitro* plantlets were transferred to pots containing a mixture of vermiculite and red soil (1: 1) for acclimation for a period of three - four weeks. The well-acclimatized plantlets were transferred to the field condition. The transplantation survival rate was 85 – 90% after 30 days.

**KEYWORDS:** Micropropagation, *Solanum trilobatum*, Shoot tip, Multiple shoots.

### ABBREVIATION

MS: Murashige and Skoog (1962) medium; BAP: 6-benzylaminopurine; TDZ: Thiodiazuron; PGR: Plant growth regulator.

### INTRODUCTION

In recent years, Medicinal plants have attracted global interest as they constitute a rich treasure improve of cultural information and are source of natural products, which provides health security to millions to in rural communities. They help to generate additional employment and income offer opportunities for processing enterprises; contribute to foreign exchange and support biodiversity and conservation objectives.

In recent years there has been renewed interest in natural medicines that are obtained from plant parts or plants extracts. Nearly 40% or more of the pharmaceuticals currently used in Western countries are derived or at least partially derived from natural sources.<sup>[1]</sup> Indiscriminate exploitation coupled with lack of attention to the development of cultivation practices has resulted in considerable depletion of the wild stock of many medicinal herbs. Preservation of germplasm collections and micro propagation of economically important plants are of utmost important.

*Solanum trilobatum* L. (Solanaceae) is one of the important perennial medicinal herb found in some of the warmer parts of the tropical and subtropical regions. Popularly called 'thuduvalai' by the local tribes, villagers and herbalists, this ethno botanical herb is known to have unique medicinal properties. Preparations made from the leaves and stems of the plant are used in herbal medicine for asthma, chronic febrile affections and difficult parturition. In addition, *S. trilobatum* seeds are known to be difficult to germinate. Increasing human and livestock populations have already affect either status of wild plants, particularly those used in herbal medicine. In this study, we report successful and efficient plant regeneration from callus of *S. trilobatum*.

## MATERIALS AND METHODS

Shoot tip explants of *S. trilobatum* were collected from wild population of Tiruchirappalli regions without removing the plant from natural habitat. The shoot tip explants were first washed with running tap water for half an hour to remove the soil particles and other extraneous fine particles. Explants were surface sterilized with Bavistin 5% for 15 minutes. Then they were rinsed in distilled water twice or thrice and were then taken to the laminar airflow chamber where wash twice or thrice using sterile distilled water. After that, shoot-tip explants were surface sterilized with 0.1% HgCl<sub>2</sub> for 4 minutes. They were again washed twice or thrice with sterile distilled water.

The surface sterilized shoots were excised to obtain shoot tip explants. Explants were cultured individually on MS medium containing different concentrations (1.0 - 3.0 mg/l) of BAP and TDZ (1.0 - 3.0 mg/l). Well-elongated shoots were carefully excised and rooted on MS basal medium supplemented with IBA (1.0 - 2.5 mg/l). Both proliferation and rooting media contained 3% sucrose and gelled with 0.8% agar (Hi-Media, India). The pH was adjusted to  $5.7 \pm 0.1$ . All the cultures were maintained in a growth room with a 16 h

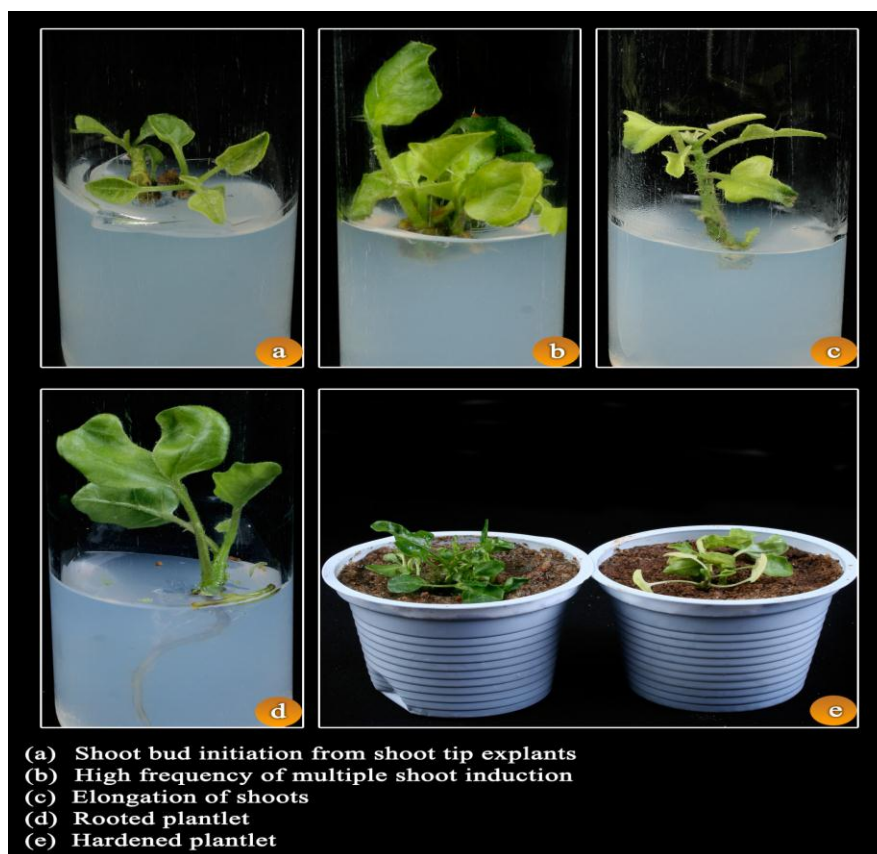
photoperiod (cool, white fluorescent light -30 $\mu$ mol m/s) and the temperature maintained at 25 $\pm$ 2 $^{\circ}$ C.

### STATISTICAL ANALYSIS

Each treatment consisted of 10 replicates and the experiment was repeated thrice. Observations were recorded on the percentage of response (number of cultures responding for shoot proliferation and root induction), the number of shoots per explants and mean shoot length, respectively.

### RESULTS AND DISCUSSION

Micro propagation was carried out with various concentrations of BAP and TDZ. The response of shoot tip to various concentrations of these hormones was expressed in Table-1. The various concentrations BAP (1.0 to 3.0 mg/l) and TDZ (1.0 to 3.0mg/l) were used. Maximum response of explants as well as maximum number of shoots was observed at 2.5 mg/l BAP (80% and number of shoots 12). Considering the TDZ the optimum hormone concentration was 2 mg/l in which the response was 45 % and the average number of micro shoots was 3. However, the response in higher dosage was decreased in multiple shoot formation. A comparison of efficiency of hormone indicates that BAP is certainly superior to TDZ because the response of explants was 80 %. The number of average micro shoots was 12 at 2.5 mg/l BAP. Hence, BAP is ideal in the micro propagation of *Solanum trilobatum* from shoot tip. The various stages of micro propagation using shoot tip explants were expressed in figure – 1a,b &c. Similar results were also observed in Sanatomb, K., and Sharma, J. reported that high frequency of shoots were produced on *capsicum frutescens* L. using MS medium with 8.8- 44.4  $\mu$ M BAP.<sup>[2]</sup> Similar studies were carried out with BAP 0.5 mg/l in Banana.<sup>[3]</sup> The combination of BAP and Kin was ideally suitable in plants like *Ocimum sanctum*.<sup>[4]</sup> *Hypericum patulum*.<sup>[5]</sup> *Malus domestica*.<sup>[6]</sup>



**Figure - 1**

**Table – 1: Effect of BAP and TDZ on micropropagation of *Solanum trilobatum* from shoot tip explants.**

PGR concentration mg/l	No. of Tubes Cultured	No. of Tubes Response	% of Response	No. Shoots /Explants Mean $\pm$ SD
<b>BAP</b>				
1.0	20	4	20	1.20 $\pm$ 0.80
1.5	20	8	40	3.00 $\pm$ 0.80
2.0	20	10	50	4.80 $\pm$ 0.63
2.5	20	14	70	11.30 $\pm$ 0.88
3.0	20	11	55	6.70 $\pm$ 0.93
<b>TDZ</b>				
1.0	20	5	25	1.60 $\pm$ 1.20
2.0	20	9	45	3.10 $\pm$ 0.32
3.0	20	7	35	1.60 $\pm$ 0.50
PGR concentration mg/l	No. of Tubes Cultured	No. of Tubes Response	% of Response	No. Shoots /Explants Mean $\pm$ SD

The well-elongated shoots were excised and root induction was carried out with different concentrations of IBA. The MS medium supplemented with IBA (2.0 mg/l) was most effective on rooting, resulting in maximum rooting percentage (85%). A well established root system was formed in 3 to 4 weeks (Fig-1d). Maximum number of roots  $4.85 \pm 0.28$  were achieved with 2.0 mg/l IBA and mean root length was  $3.8 \pm 0.10$ cm. Similar results were also obtained from *Plumbago zeylanica*.<sup>[7]</sup> and *Abrus precatorius*.<sup>[8]</sup> After the root development, the plantlets were removed from the culture tubes and washed in running tap water. After that, root system was treated with 5% of Bavistin to avoiding the fungal contamination. Well-rooted plantlets were transferred to the poly-cups with sterilized red soil and vermiculate with half strength of MS salts solution for a week (Fig-1e). About 85 – 90% of the regenerated plantlets could tolerate and survive under field conditions.

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