

## *In Vitro* studies in *Solanum virginianum* L.

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### Article Info

Received: 19-07-2020,

Revised: 29-08-2020,

Accepted: 05-09-2020

**Keywords:** *In Vitro*,  
*Solanum virginianum*,  
IAA, BAP.

### Abstract

The present investigation aimed to develop standard protocol for *in vitro* propagation of *Solanum virginianum* L. It is one of the important medicinal plant belongs to family Solanaceae. Leaf and stem node explants were tried on MS medium along with various concentration of growth hormone. The regeneration of multiple shooting was achieved on MS medium along with 0.5 mg/L of IAA in combination of various concentrations of BAP and KIN. The maximum percentage of regeneration and maximum length of shoot were recorded on 0.5 mg/L of IAA in combination of 2.0 mg/L of BAP and KIN using stem node explant however 2.0 mg/L and 3.0 mg/L of BAP and KIN using leaf explant respectively.

### INTRODUCTION

Medicinal plant *Solanum virginianum* L is belong to the family solanaceae, commonly known as yellow berried night shade. It is coined by different vernacular name like in Marathi called as bhui ringani, Sanskrit kantkari etc.

**Description and distribution of plant:** It is frequently distributed in Asia (Saudi Arabia, Yemen, Afghanistan, Iran, China, Bangladesh, India, Nepal, Pakistan, Sri Lanka, Myanmar, Thailand, Vietnam, Indonesia, Malaysia are native countries) and is adventives in Egypt. In India it is recorded tropical, subtropical and all four geographical regions. Frequently it has been considered as weed plant but in Ayurveda and folklore medicine since time immemorial there are major reports in literature about its other potentials (Madhavi *et al.*, 2014). The morphologically *Solanum virginianum* is prickly diffuse bright green perennial herb, somewhat woody at the base while stem is zigzag, branches are numerous. The younger ones clothed with dense stellate tomentum, prickles are compressed, straight, yellow, glabrous and shining, often exceeding 1.3 cm. Leaves are usually

5-10 in numbers and 2.5-5.7 cm in length, ovate or elliptic, sinuate, obtuse or sub-acute, stellate hairy on both sides. Sometimes become nearly glabrous in age, armed on the midrib and often on the nerves with long yellow sharp prickles, base usually rounded and unequal-sided. The petiole 1.3-2.5 cm long, stellately hairy. The berries are green with white strips when young but yellow when mature and many seeded. They are 1.3-2 cm in diameter, yellow, or white with green veins, surrounded by the enlarged calyx. Seeds are 2.5 mm in diameter and glabrous. Calyx is nearly 1.3 cm long, densely hairy and prickly; tube short, globules. Lobes are 11 mm long, linear-lanceolate, acute and hairy outside. Filaments are 1.5 mm long, glabrous; anthers 8 mm long, oblong lanceolate, opening by small pores. Ovary is ovoid, glabrous; style glabrous (Sachin *et al.*, 2010).

### Medicinal and chemical properties:

In Ayurveda, plant is described as pungent, bitter, digestive, alternative astringent. Stems, flowers, fruits are bitter and contains carminative properties. Root decoction used as febrifuge, effective diuretic and expectorant.

Charaka and Sushruta used the extract of entire plant and fruits in internal prescription for bronchial asthma, tympanitis, misperistalsis, piles and dysuria and for rejuvenation. The whole plant is used traditionally for curing various ailments (Atul *et al.*, 2013). Decoction of the plant is used in gonorrhoea; paste of leaves is applied to relieve pains, useful in the treatment of catarrhal fever, coughs, asthma and chest. *Solanum virginianum* is a well-known medicinal plant in traditional medicinal system. Recent scientific studies have emphasized the possible use of *Solanum virginianum* in modern medicine (Reddy and Reddy, 2014). Chemically Okram and Thokchom (2010) reported it is a valuable source of alkaloids, sterols, saponins, flavonoids and their glycosides and also carbohydrates, fatty acids, amino acids (Gnana *et al.*, 2013).

## MATERIALS AND METHODS

### Preparation of explant and sterilization:

The explant like leaf, stem node and mature fruits were collected from young healthy plantlets of *Solanum virginianum*, from road sides of caves and campus of Dr. Babasaheb Ambedkar Marathwada University, Aurangabad. All these explant were washed with running tap water for 5 minutes, followed by 70% ethanol for 1 minute and finally with distilled water for 3 minutes. Surface sterilization of explant was carried out by washing with sterile distilled water for 3 minutes followed by various concentration of mercuric chloride (HgCl<sub>2</sub>), leaf explant sterilized with 0.1% whereas stem node with 0.2% of HgCl<sub>2</sub>. It was followed by two subsequent rinses with sterilized double distilled water in laminar airflow. All these explant were cuts in to small pieces and inoculated on suitable media.

### Culture media and Culture condition:

All experiments of investigation were tried on MS media (Murashige and Skoog, 1962) supplemented with various concentration of growth regulators. MS medium was fortified with 3 % sucrose and clorigar for solidification respectively and pH was adjusted to 5.8. The media were steam sterilized in an autoclave under 15 psi and 121°C. After the inoculation culture bottles were transfers to culture room under a 16 h photoperiod supplied by cool white fluorescent cool tubes light and temperature at 25± 2°C. At least 5 replicates raised for each treatment and data were recorded in table.

## RESULTS AND DISCUSSION

Standard protocol for surface sterilization of explants was standardized by trial and error method. Surface sterilization of leaf and stem node explant were tried with 0.1-0.3% of HgCl<sub>2</sub> for 3 and 5 minutes duration. The maximum microbe's free cultures and high regeneration percentage were recorded at 0.1% for leaf and 0.2% of HgCl<sub>2</sub> for stem node explant during the present study. The hormones free MS medium was found ineffective to induced callus or regeneration of shoot using both explants leaves and stem node. Shoot regeneration was achieved from both explant from BAP and KIN in combination of 0.5 mg/L of IAA. Lower concentration of BAP was found effective to induced shoot regeneration however higher concentration revealed poor result of shoot regeneration similar kind of result was also recorded. The maximum shoot induction percentage along with shoot length was recorded from 0.5 mg/L of IAA in combination of 2.0 mg/L of BAP and 3.0 mg/L of KIN with 86.66% regeneration along with 1.62±0.203 and 1.35±0.181cm of shoot length using leaf explant respectively.

Stem node also revealed induction of shoot, maximum percentage of shoot regeneration was achieved on 2.0 mg/L: of BAP and KIN with 80.00% of shoot regeneration along with shoot length 1.36±0.215 and 1.30±0.191cm respectively. Similar kinds of results were reported by Sanjay *et al.*, (2013). Standard protocol for shoot regeneration was also develop by Ramar *et al.*, (2011), revealed that growth hormone BAP incorporated with MS media exhibits rapid multi[pl]ication of *solanum virginianum*.

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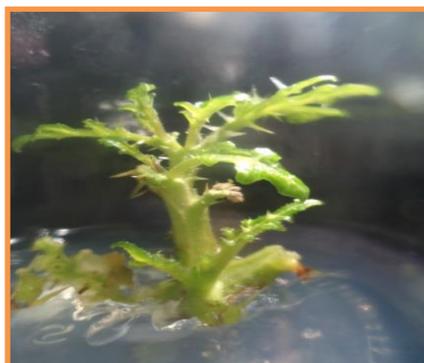
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**Table 1: Effects of different concentration of PGR's on shoot multiplication of *Solanum Virginianum* L.**

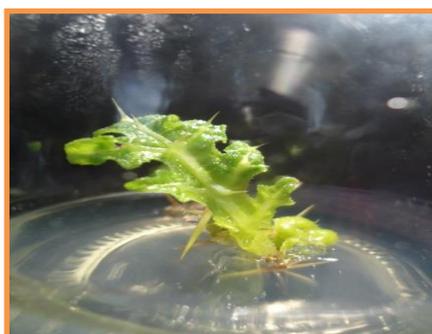
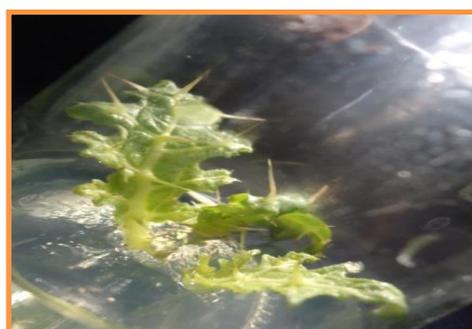
Explant	Growth hormones (mg/L)			Shoot length	% of shoot formation
	IAA	BAP	KIN		
Leaf segment	0.5	1.0		0.77±0.211	53.33%
		2.0		<b>1.62±0.203</b>	<b>86.66%</b>
		3.0		0.84±0.198	60%
		4.0		0.84±0.217	53.33%
		5.0		1.06±0.194	73.33%
	0.5		1.0	0.84±0.176	66.66%
	0.5		2.0	0.92±0.191	66.66%
	0.5		3.0	<b>1.35±0.181</b>	<b>86.66%</b>
	0.5		4.0	0.71±0.160	60.00%
	0.5		5.0	0.68±0.178	53.33%
Stem node	0.5	1.0		0.36±0.126	40%
		2.0		<b>1.36±0.215</b>	<b>80%</b>
		3.0		0.72±0.153	66.66%
		4.0		0.79±0.163	60%
		5.0		1.00±0.178	66.66%
	0.5		1.0	0.74±0.400	53.33%
	0.5		2.0	<b>1.30±0.191</b>	<b>80%</b>
	0.5		3.0	0.83±0.167	66.66%
	0.5		4.0	0.60±0.156	53.33%
	0.5		5.0	0.58±0.152	53.33%

After 24 days mean ± SE of 5 replicates

**Photo plate 1: Effect of IAA and BAP on Multiplication**



**Photo plate 2. Effect of IAA and BAP in vitro growth**



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#### How to cite this article

**Haribabu Narra, 2020.** *In Vitro* studies in *Solanum virgianianum* L. . *Bioscience Discovery*, **11**(4):258-261.

**Google Scholar citation:** <https://scholar.google.co.in/citations?user=vPzEyC8AAAAJ&hl=en>

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