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In vitro micropropagation of *Sida acuta* (Burm.F) An important medicinal plant through tissue culture technique

[*1]Mushraf Qureshi and [2]Mukta Shrivastava

[1,2]*Department of Botany Govt. M.L.B Girls P.G. College Bhopal (M.P) (India)*

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[*]**Corresponding Author**
Email: musharaf.qureshi9[at]gmail.com

ABSTRACT

The plant *Sida acuta* Burm. F. belongs to family*Malvaceae* commonly known as wire weed or (Broom weed)is an annual or perennial medicinal herb or under shrub, mostly distributed in tropical and sub tropical regions of the world.Due to the vast medicinal importance of *Sida acuta.*Its natural population has been declined very fast due to indiscriminate and illegal collections and destruction of its habitats a result there is need to conserve the important medicinal plant species. Its *in vitro* protocol techniques are currently unavailable to help growers to meet the demand of the plant for cultivation and pharmaceutical industry. Thus the present study has been designed to develop a reliable and reproducible protocol of this important plant which could be used for mass multiplication of this plant species to meet the increasing requirement of the pharmaceutical industry as well as for the conservation of germplasm. The present study deals with rapid and efficient protocol development for *in vitro* propagation of *Sida acuta* . Shoot induction on Murashige and Skoog medium supplemented with various auxins and cytokinins individually and in various combinations has been achieved by using axillary and apical meristems.MS medium fortified with BAP and NAA was found to be effective individually.

1. Introduction

Sida acuta Burm. F., a small, erect, much branched, perennial shrub or herb; ranging from 30 to 100 cm in height, with a strong taproot; stem and branches flattened at the extremities, fibrous, almost woody at times. The weed is frequently found in pastures, wastelands, cultivated lands, roadsides, lawns, and in planted forests. Once the plant becomes established, it is very competitive, holding and denying sites to other plants. It does appear to do best in disturbed habitats1**.** The genus comprises of about 200 species in the world, out of which 12 are occurring in India. The plant has a variety of traditional uses. In traditional medicine, The leaf juice is used for vomiting and gastric disorders 2, the hot water extract of the dried entire plant is administered orally as a febrifuge, an abortifacient and a diuretic 3, the plant is often assumed to treat diseases such as fever, headache, skin diseases, diarrhoea, and dysentery. It is used as astringent, cooling, tonic, and febrifuge, stomachic4, anti-inflammatory and hepatoprotective 5. The total aqueous extract of the whole plant showed significant activity against carbon tetrachloride, paracetamol and rifampicin induced hepatotoxicities in experimental albino rats. Phytochemically, these species contain a group of alkaloids like ephedrine and its isomers, vasicinone, vasicine and vasicinol .

Due to these medicinal values, the plant is being over-exploited in recent years. In addition, the efficiency of reproduction is also found to be less due to its low seed germination and viability and lack of vegetative propagation methods. Thus the present study has been designed to develop a reliable and reproducible protocol of this important endangered plant which could be used for mass multiplication of this plant species to meet the increasing requirement of the pharmaceutical industry as well as for the conservation of germplasm.

2. Materials and Methods

Nodal segments (1.0-1.5cm) were excised from the plants growing in green house of Botany Department OF GOVT. M.L.B College Bhopal. All the explants were washed with liquid detergent under running tap water to remove dust particles. The explants were then treated with 0.1% (w/v) mercuric chloride for 4-6 minutes under aseptic conditions. After this these explants were then thoroughly washed 5-6 times with sterilized double distilled water to remove the traces of $HgCl_2$ (mercuric chloride). The nodal segments were inoculated on MS medium supplemented with various concentrations of BAP (0.5-4.0 mg/l) alone or with combination with auxins (0.5-1.0 mg/l) NAA, in various combinations for shoot induction and regeneration .The cultures were incubated at a temperature of 25±2ºC and a photoperiod of 16hrs light (intensity of 2000 lux) and 8hrs of dark. Visual observations like callus induction, growth of callus, number of days taken for bud break, percentage of bud break and number of shoots regenerated per explants were recorded regularly.

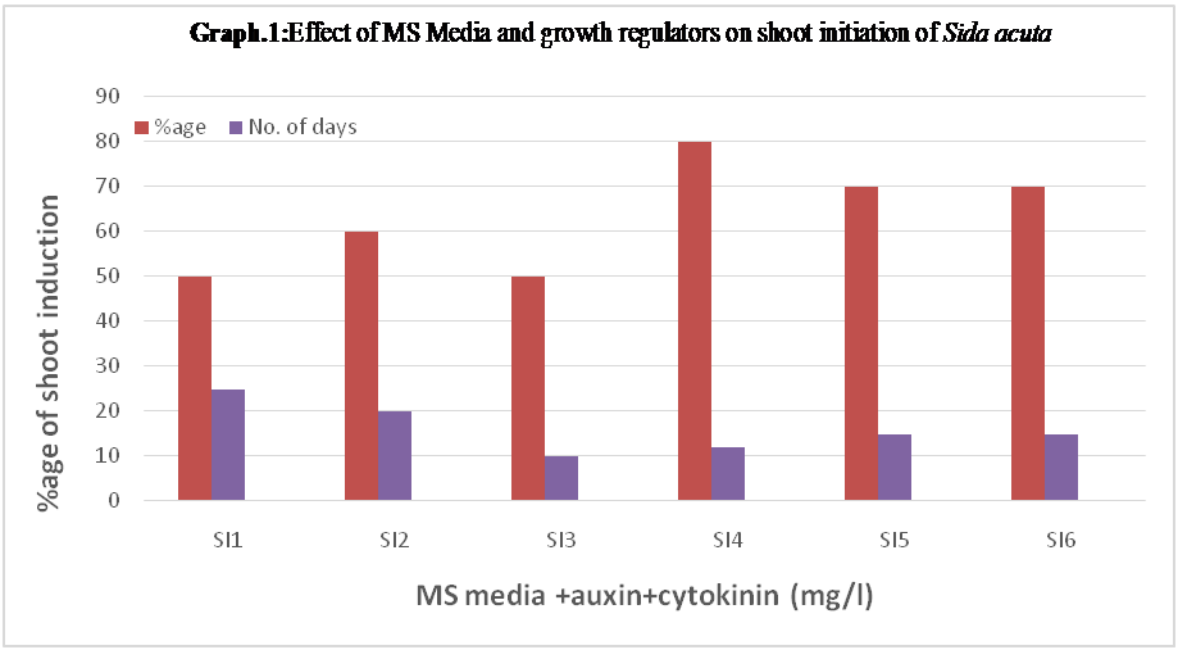
3. Observation and results

A combined effect of different cytokinins BAP and auxins (NAA) in various combinations was also studied. The medium with BAP (0.5 mg/l) + NAA (0.2 mg/l) showed maximum Eighty (80%) bud break after 12-15 days of inoculation. Supplementation of NAA with BAP did not make much difference. In case of BAP supplemented media, the medium with BAP (0.5 mg/l) showed seventy (70%) percent bud break after15- 20 days of inoculation with 1-2 shoots per explant**(Table-1).** The combination of NAA with cytokinins (BAP) promoted shoot formation in various plant species as observed by **6**78**.** The medium supplemented with BAP (0.5 mg/l) + NAA (0.2 mg/l) supported maximum number of shoots

(3.0) per explant and responded best among all media tried in combination.

Table.1.Effect of MS medium with growth regulators on Shoot induction of *Sida acuta*

Media Code.	MS+Auxin+cytokinin (mg/l).	%age of bud break.	No. of days required.	Mean No. of shoots initiated ± SE.	Mean shoot length in cm±SE.
SI_1	0.1 BAP.	50	25	2.3±0.1	1.9 ± 0.05
SI_2	0.5 BAP.	60	20	4.9± 0.15	3.03 ± 0.12
SI_3	1.0 BAP	50	10	4.2±0.05	2.9± 0.17
SI_4	0.5 BAP+0.5NAA	80	12	3.8±0.24	2.6 ±0.12
SI_5	0.5 BAP+0.5 IBA	70	15	5.2±0.26	3.5± 0.25
SI_6	1.0 BAP+0.5NAA	70	15	3.4±0.17	2.26 ± 0.03



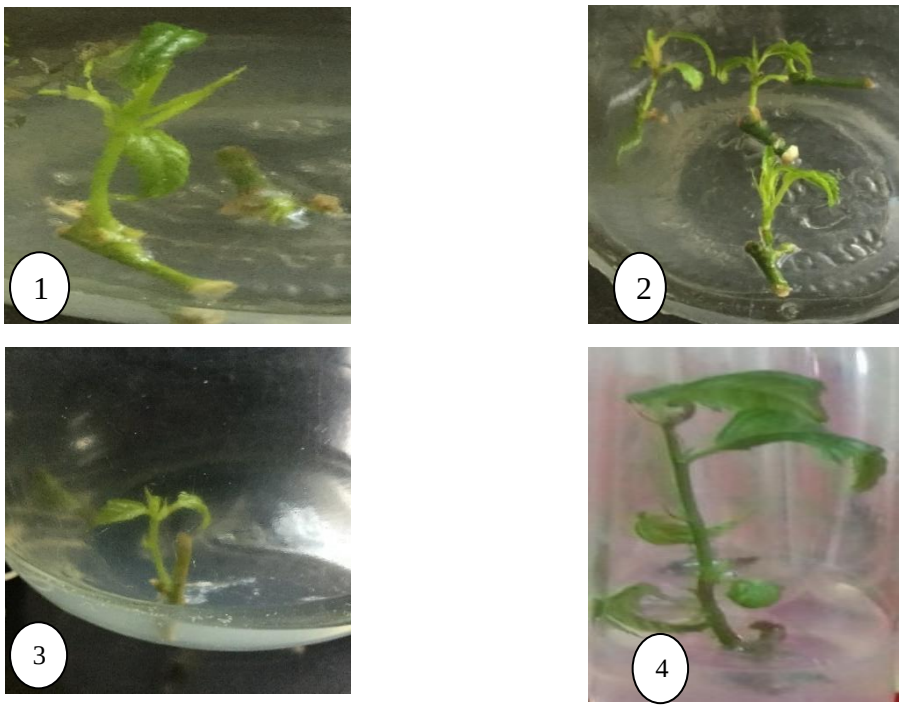


Figure (1-3) Shoot induction in 0.5 mg/l BAP+0.2 mg/l NAA, after12- 15 days.

4. Conclusion

The plant holds great promise as a commonly available medicinal plant and it is indeed no surprise that the plant is referred to in the Indian traditional circles. From the available literature on various aspects of the plant -traditional to biochemical and ethnobotanical to pharmacological and micro propagation however there many gaps which need to be filled by concurrent researchers in different disciplines. One must make the best use of the naturally available resources which provide valuable raw material for advanced research. The present study deals with rapid and efficient protocol development for *in vitro* propagation of ***Sida acuta*** . Shoot induction on Murashige and Skoog medium supplemented with various auxins and cytokinins individually and in various combinations has been achieved by using axillary and apical meristems.MS medium fortified with 0.5- 2.0 mg/l BAP and 0.2 mg/l NAA was found to be effective individually.

Thus the present study has been designed to develop a reliable and reproducible protocol of this important medicinal plant which could be used for mass multiplication of ***Sida acuta*** to meet the increasing requirement of the pharmaceutical industry as well as for the conservation of germplasm.

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