

## RAPID *IN VITRO* PLANTLET FORMATION FROM SHOOT TIPS IN SOYBEAN (*GLYCINE MAX* L. MERR.) CV. PUSA-16

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The *in vitro* regeneration from shoot tips of *Glycine max* (L. Merr.) cv. Pusa-16, obtained rapidly, when explants excised from 7-10 days old seedling were placed on medium, containing MS salts, B<sub>5</sub> vitamins supplemented with benzyladenine, BA (0.2, 1.0 mg/1) and Indolebutyric acid (0.5 mg/1). The regenerated plants were transferred to the field.

**Keywords:** *Glycine max*; *In vitro* regeneration; Plantlet; Shoot tip.

Plant regeneration from seed explants has been reported for a number of legumes, including Soybean<sup>1-4</sup>. Immature leaves were taken as source to induce regeneration in many plants like cultivated peanut (*Arachis hypogaea*), perennial peanut (*A. glabrata*)<sup>5</sup> and *Lathyrus sativus*<sup>6</sup>.

There are many reports on adventitious shoot regeneration<sup>7,8</sup>. Plant regeneration is based on proper manipulation of the cultural environment and appropriate selection of genotype and tissue. We report here on the regeneration of Soybean from shoottips of *G. max* cv. Pusa-16.

Seeds of Soybean (*Glycine max* (L.) Merr. cv. Pusa-16) were procured from Rajasthan State Seeds Corporation Ltd. Seeds were surface sterilized by 0.1% mercuric chloride for 3min and washed thrice with sterilized distilled water. Seeds were germinated aseptically on half strength Murashige and Skoog's (MS) Medium<sup>9</sup> containing 0.8% agar-agar, 1.5% sucrose, lacking hormones. pH of the medium was adjusted to 5.8 before autoclaving. Seedlings were incubated under continuous illumination (2500 Lux) at 25 ± 2°C. Various explants e. g. hypocotyls, cotyledons, shoottips, leaves were excised from 7-10 days old seedling and transferred to basal medium of Murashige and Skoog (MS) with B<sub>5</sub> vitamins<sup>10</sup>

supplemented with different concentration of Benzyladenine (BA), Indolebutyric acid (IBA). The medium was autoclaved at 15-20 lbs pressure for 20 min. The cultures were maintained at temperature of 25 ± 2°C under continuous fluorescent light.

In *Glycine Max* cv. Pusa-16 successful induction of shoot buds via callus formation was observed in all the shoot tip explants. Callus tissue was obtained within 2 weeks of inoculation of explant in Murashige and Skoog<sup>9</sup> basal medium and B<sub>5</sub> vitamin<sup>10</sup> supplemented with BA (1.0mg/1) and IBA (0.5 mg/1). Shoot bud differentiation was obtained from this callus tissue. Root initiation was also observed simultaneously. Thus the callus, shoot, roots and entire plant formation from shoot tip cultures in *G. max* c.v. pusa-16 was obtained within 3-5 weeks of initiating cultures. The maximum plantlet formation per responding explant was at 0.2 mg/1 and 1.0 mg/1 of BA and 0.5 mg/1 of IBA.

The use of a single hormone or in combination for the plantlet formation is in contrast to the previous work by Wright *et al.*<sup>4</sup> on *G. max*; multiple shoots were obtained on half strength MS basal medium with BA; while rooting was obtained on B<sub>5</sub> medium lacking phytohormones. The present study was undertaken for the purpose of obtaining rapid regeneration in Soybean without any intervening.

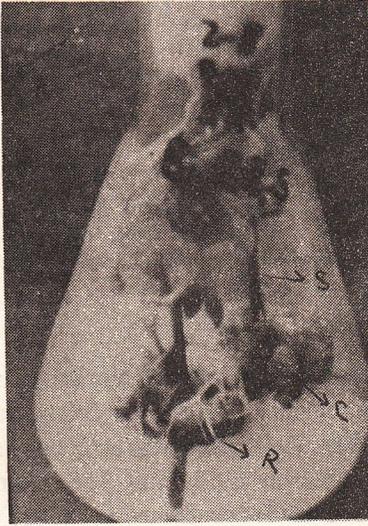


Fig 1. Plantlet formation in Soybean. C, Callusing on MS Salt and B<sub>5</sub> vitamins supplemented with BA (1.0 mg/l) and IBA (0.5 mg/l), R, root initiation, S, shoot.

## References

1. Grant JE 1984, *Plant Cell Tissue Organ Cult.* 3 169
2. Hildebrand DF, Phillips GE and Collins GB 1985, Soybean In: Bajaj YPS (ed) *Biotechnology of plant improvement*, vol. 2 282. Berlin, Heidelberg : Springer-Verlag
3. Hymowitz T, Chalmers ML, Constanze SH and Saam MM 1986, *Plant Cell Rep.* 3 192
4. Wright MS, Rocler SM, Hinchee MA and Carnes MG 1986, *Plant Cell Rep.* 5 150
5. McKently Att., Moore GA and Gardner FP 1991, *Crop Science* 31 833
6. Roy RK, Singh B, Mehta SL, Barat GK, Gupta N, Kirti PB and Chopra VL 1991, *Ind. J. Exp. Biol.* 29 327
7. Hammat N, Doney MR and Nelson RS 1986, *Physiol. Plant* 68 125
8. Surya Prakash, Chowdhury JB, Jain RK and Chowdhury VK 1992, *Ind. J. Exp. Biol.* 80 1149
9. Murashige T and Skoog F 1962, *Physiol. Plant.* 15 473
10. Gamborg OL, Miller RA and Ojima K 1968 *Exp. Cell. Res.* 50 151