Tropical Agricultural Research Vol. 4 1992

# *In vitro* Propagation of *Katurumurunga* (*Sesbania grandiflora* (Linn) Poir) through Callus Culture

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*ABSTRACT. Hypocotyl explants obtained from in vitro grown seedlings of Katurumurunga (Sesbania grandiflora Linn) were cultured in MS medium supplemented with BAP and 2, 4-D for callus induction. The medium supplemented with 1 mg/l BAP and 2.5 mg/l 2, 4-D was found to be the best of the conditions tested for establishment of callus. Shoot formation was observed following transfer of these calli to MS medium containing 1 mg/l gibberellin acid. Root induction of these shoots was achieved in half strength MS medium containing IBA (2 mg/l) and Kinetin (0.005 mg/l).*

## INTRODUCTION

Kathurumurunga (*Sesbania grandiflora*) is a nitrogen fixing plant possibly native to Indonesia (Dassanayake and Fosberg, 1991) and cultivated in many South Asian countries. Leaves, flowers and young pods of the plant are used as a food which is of high protein, mineral and vitamin content. Katurumurunga also provides animal feed, fertilizer and pulpwood. The plant is of medicinal value and is used for treatment of rheumatic swellings and eye ailments (Jayaweera, 1981). It is propagated by seeds but seedling establishment is difficult due to poor germinability of seeds.

Propagation of many woody plants by *in vitro* techniques has been reported (Lakshmi *et al.*, 1979; Bayliss and Dunn, 1979). Micropropagation of many tropical woody legumes has also been

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reported (Sharma and Chandra, 1987). Propagation of *Acacia auriculiformis*, a multipurpose tree legume has been achieved by induction of plantlet regeneration from callus obtained from hypocotyl segments (Rao and Prasad, 1991). The present study was undertaken to investigate the feasibility of *in vitro* propagation of *Sesbania grandiflora*.

## MATERIALS AND METHODS

Seeds, directly collected from a plant in a home garden in Dangolla were scarified mechanically, surface sterilized with 12% (w/v) sodium hypochlorite and cultured in MS (Murashige and Skoog, 1962) medium without any growth regulators. The cultures were incubated in light (2.9 $Wm^{-2}$) at 26 – 28 °C with a 8h photoperiod.

When the seedlings were 30 days old, hypocotyl segments (one cm long) were transferred into MS media modified with 0, 2.5 and 5 mg/l of 2, 4 – D and 0, 1 and 2 mg/l of BAP (Table 1). The callus formation was observed after 20 days in culture. After 22 days in culture the calli were transferred into MS media modified with 1 mg/l of gibberellic acid and 0, 1 and 2 mg/l of BAP for plant regeneration.

Regenerated shoots were separated after 65 days in culture and were kept for rooting in half strength MS medium supplemented with 2 mg/l IBA and 0.005 mg/l Kinetin. Plantlets were acclimatized in sterilized and non sterilized top soil, sand and a mixture (1.1) of top soil and sand. The potted plants were covered with polythene bags to maintain relative humidity.

## RESULTS AND DISCUSSION

BAP alone was not useful for callus induction in hypocotyl explants. 2, 4 – D favoured callus initiation and cell proliferation was greatly enhanced in the presence of both BAP and 2, 4 – D. Similar growth response for these two chemicals have been observed with cultured tissues of Nadun (Abeyratne *et al.*, 1990) but the cytokinin and auxin requirements generally vary with the plant species. For good callus production from shoot segments and shoot tips of sandal wood, the presence of Kinetin with 2, 4 – D is required (Lakshmi *et al.*, 1979). For other species like Barley 2, 4 – D alone in the MS medium produces

Table 1. Growth response of cultured hypocotyl explants of Katurumurunga in relation to BAP and 2, 4-D in MS medium.

| BAP mg/l | 2, 4-D mg/l | Growth response |
|---|---|---|
| - | - | Explant degenerated |
| 1 | - | No growth but explant survived |
| 2 | - | No growth but explant survived |
| - | 2.5 | Fair callus formation |
| - | 5 | Poor callus formation |
| 1 | 2.5 | Good callus formation |
| 2 | 2.5 | Poor callus formation |
| 1 | 5 | Fair callus formation |
| - | 5 | Poor callus formation |

good callus from mature embryos (Bayliss and Dunn, 1979). In *Acacia auriculiformis* callus formation from hypocotyl segments was observed with BA, BAP and NAA in MS medium (Rao and Prasad, 1991). Callus tissues established from *Aegle Marmeles* produced shoots when cultured in MS medium containing kinetin (1 mg/l) but BAP (0.5 mg/l) was found to be most potent in shoot regeneration (Arya and Shekhawat 1987). These shoots produced more shoots following transfer to hormone free half MS medium. Subsequently, root induction of these shoots was achieved with Whites medium containing 0.5 – 2.5 mg/l IBA (Arya and Shsekhawat, 1987). However, in the present study shoot formation was observed after transferring the callus to MS medium containing gibberellic acid (1 mg/l). BAP in the presence of gibberellic acid (1 mg/l) did not favour shoot formation and at 2 mg/l (BAP) shoot formation was not observed. Half strength MS medium containing IBA (2 mg/l) and kinetin (0.005 mg/l) was good for induction of roots in these shoots. The plantlets produced could be successfully established in the soil.

## CONCLUSION

*In vitro* propagation of *Sesbania grandiflora* through callus culture is feasible. Its commercial use may now be investigated. Application of the technique with appropriate modifications to commercially promising species of the same family could be undertaken.

## ACKNOWLEDGEMENTS

The authors thank Mr. H.N P. Wijeyagunasekera, Head of the Department of Agricultural Biology for providing an opportunity to do the research study. The help from the technical staff of the department is also acknowledged with gratitude.

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