

Callogenesis in *Tragia involucrata* L.-A potent tribal medicinal plant

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SUMMARY

Tragia involucrata L. is a well known medicinal plant used in tribal medicine of India. In this paper *in vitro* studies have been reported. Calli were initiated from leaf and stem segments on Murashige and Skoog's (MS) medium supplemented with 2, 4-D or Kinetin. Explant browning a major hurdle in the establishment of cultures was minimized by adding ascorbic acid (2mg/l) to the MS basal medium. The explants induce proliferated mass of callus on MS medium supplemented with 2, 4-D and Kn. Luxuriant mass of callus was achieved by sub culturing the calli on MS supplemented with BAP (3.0 and 5.0 mg/l) alone or in combinations with NAA (0.5, 1.0 and 1.5 mg/l). By subculturing the callus on fresh medium at 15 days interval, browning of the callus was eliminated simultaneously the initiation of somatic embryos. But calli failed to induce somatic embryo genesis even addition of coconut water (10.20 and 25%) while sub culturing, it produce only whitish green, friable, soft callus. Rhizogenesis was achieved by subculturing calli on MS medium containing IAA (2.5 and 5.0 mg/l) and IBA (2.0 and 4.0mg/l) alone.

Key words : Leaf culture, Callus induction, Sub culture, Rhizogenesis

Tragia involucrata (Euphorbiaceae) is a perennial evergreen twiner with hispid, stinging bristles and widely used in traditional system of medicine for a variety of diseases. The roots have been reported to possess diaphoretic and alterative actions and the infusion is given when the extremities are cold during fever, skin infection and also for pains in the legs and arms. Root paste is applied for removal of guinea worms and it forms basis of an external application in leprosy. A decoction of the root (1 in 10) was found to be useful in relieving bronchitis and the attendant fever (Kirtikar and Basu, 1975; Chatterji and Pakrashi, 1994 and Chopra *et al.*, 1956). The decoction of the leaves is being used by tribal people of western ghats of Tamilnadu (Kalrayan hills) for the treatment of skin infection, pain swelling, children scabies and eczema (Perumalsamy *et al.*, 2006c).

Callus formation is the fundamental stage for many tissue culture techniques such as organogenesis, somatic embryogenesis and protoplast culture. However callus formation can be especially difficult to attain in some species. We are not aware of any method that was available in the literature on the tissue culture studies in *T. involucrata*. Phytochemically, the air-dried powder of alcoholic and ether fraction of root contains beta sitosterol and beta-sitosterol-beta-D-glycoside (Srinivasan, 1985).

Leaf and root extracts show multiple pharmacological effects including antibacterial (Perumalsamy *et al.*, 2006b), wound healing (Perumalsamy *et al.*, 2006c), psychopharmacological (Dhara *et al.*, 2002) and a significant analgesic and anti-inflammatory activities (Dhara *et al.*, 2000; Perumalsamy *et al.*, 2006a).

MATERIALS AND METHODS

Leaf and stem parts of the plant were used as explants source. The explants were collected from in and around university campus and the explants were excised into 1.0-1.5cm length and washed thoroughly under running tap water for 30 min to remove the adhered surface particle. Then, treated with a liquid detergent laboline (5% v/v) for 30 min followed washing under running tap water. The explants were surface sterilized with 0.1% (v/v) aqueous mercuric chloride for 10 min and finally rinsed with sterilized, cooled distilled water aseptically and were carefully inoculated onto the Murashige and Skoog's (1962) basal media supplemented with 3% sucrose and solidified with bacteriological grade agar (0.8%) and various concentrations of growth hormones. The media pH was adjusted to 5.8 with 1N NaOH or 1N HCl before autoclaving at 121°C for 20 min at 15 lbs pressure. The cultures were incubated and maintained at 25±2°C under 16/8 hr photoperiod of 2000lux light intensity provided by white, fluorescent tubes with 60-80% relative humidity.

Callus induction:

MS medium containing 2, 4-D (1.0, 2.0 and 5.0mg/l) and kinetin (1.0, 2.0 and 5.0mg/l) were tested for callus induction from the explants. MS lacking growth

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