

IN VITRO ORGANOGENESIS IN *PLUMERIA ACUTIFOLIA* (APOCYANACEAE)

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Zygotic embryo axis, cotyledon segments of *Plumeria acutifolia* were cultured on Murashige and Skoog's basal medium supplemented with adjuvants (2, 4-D, IBA, KN, BAP, CW). Profused rooting in cotyledon and normal shooting in embryo axis explants were noted. Both explants showed considerable variation for organogenesis under tested combinations.

Keywords : *Plumeria acutifolia*; *in vitro*; Organogenesis.

Much of the earlier *in vitro* work on latex producing plants has been carried out in view of the presence of considerable amounts of low molecular weight hydrocarbons that could be used as a substitute for fossil liquid fuel (San Pietro, 1980; Tideman and Hawker, 1982). *In vitro* propagation enables production of large number of these promising plants.

Apart from Euphorbiaceae and Asclepiadaceae, Apocyanaceae is one of the latex yielding family and known for useful alkaloids, and hence tissue culture work has been carried out in some species (Harris *et al.*, 1964; Mitra *et al.*, 1965; Lee *et al.*, 1972; Carew and Bainbridge, 1976; Dhruva *et al.*, 1977; Ramavat

et al., 1978; Abou-Mandour *et al.*, 1979; Heble *et al.*, 1983).

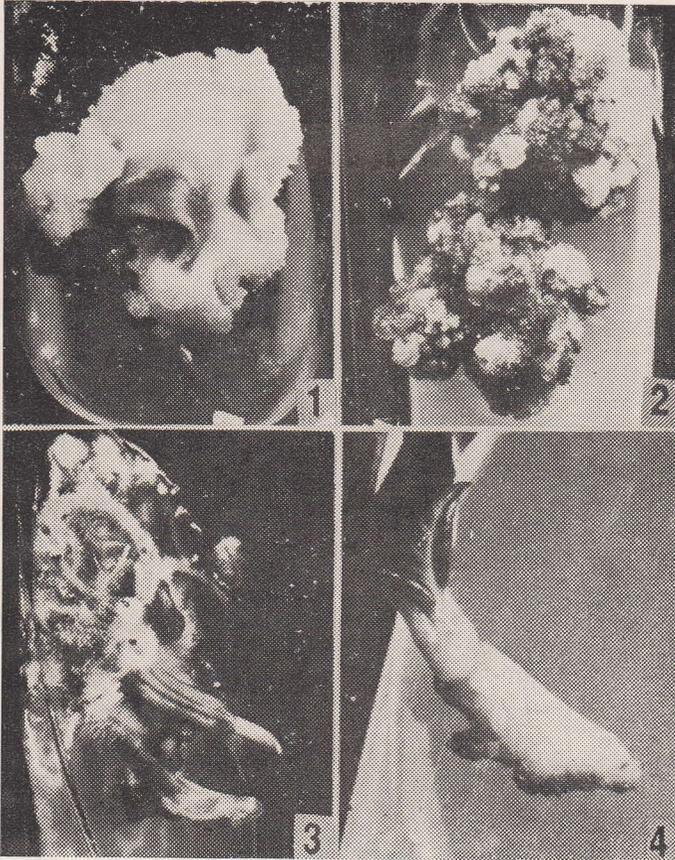
Earlier work in family Apocyanaceae is concentrated on secondary metabolism as in *Catharanthus roseus* and *Rawolfia serpentina* (Sagishima *et al.*, 1989; Ruyter *et al.*, 1989). Earlier stem, shoot apex and internode explants were studied for morphogenesis in *Plumeria rubra* (see Tideman and Hawker, 1982). Datta *et al.*, (1975) studied the effect of acidity levels during *in vitro* xylem differentiation in *Plumeria* species.

Plumeria acutifolia the temple tree is generally grown for its fragrant large flowers and is almost always self incompatible. In Java the roots are medicinally used as

Table 1 : Effect of medium constituents on morphogenesis in tissue cultures of *Plumeria acutifolia*

Medium composition	Morphogenetic response	
	Zygotic embryo axis	Cotyledon segments
MS basal medium	—	—
MS+KN 2 mg/l	Greening of explants	Greening of explants
MS+CW 10%	Greening of explants	Greening of explants
MS+2, 4-D 2 mg/l	Callus induction	Callus+root formation
MS+2, 4-D 2 mg/l CW 10%	Brown callus	Green callus
MS+2, 4-D 2 mg/l+IBA 1 mg/l+KN 2 mg/l	Shooting	Callus+roots
MS+CW 10%+IBA 1 mg/l+BAP 1 mg/l	Shooting	Greening of explants
MS+KN 1 mg/l+BAP 0.5 mg/l+IBA 2 mg/l Ascorbic acid 0.1%	Pigment formation	Pigment formation

KN : Kinetin; CW : Coconut Water; BAP : Benzyl amino purine; IBA : Indole Butyric acid; 2, 4-D : 2, 4 Dichlorophenoxy acetic acid.



Morphogenesis in tissue cultures of *pluneria acutifolia*

Fig. 1 & 2 Callus formation from the cultured cotyledon segment and zygotic embryo axis on MS+2, 4-D 2 mg/l.

Fig. 3 profuse rooting from cut ends of cotyledon segments on MS+2, 4-D 2 mg/l+KN 2 mg/l+IBA 1 mg/l in one week.

Fig. 4 Shoot induction from embryo axis cultured on MS+IBA 1 mg/l +BAP 1 mg/l+ CW 10%.

cathartic (Lindley, 1981). Present communication deals with morphogenetic studies using embryo axis and cotyledon explants of a rare fully self compatible fruiting *Plumeria acutifolia* plant collected by one of us (BB) from Mandavalipakam, Madras, Tamilnadu.

Fresh fruits were surface sterilized with tap water and detergent (Teepol) and washed in 70% ethanol (10 sec.), 0.1% mercuric chloride (3 min.). The zygotic embryos and cotyledon segments from excised seeds were cultured separately on Murashige and Skoog's basal medium (Murashige and Skoog, 1962) fortified with different adjuvants (Table 1). The medium was adjusted to 5.8 pH and autoclaved prior to inoculation. The cultures were incubated at $25 \pm 2^\circ\text{C}$ under 2000 lux.

Explants cultured on basal medium showed no response. Medium supplemented with any of the cytokinins (KN, CW, BAP) showed greening of explants. Auxins 2, 4-D in the medium induced brown callus from embryo axis and white callus+roots from cotyledon segments in a week (Figs. 1 & 2).

MS basal medium+IBA+KN+2, 4-D induced shooting in embryo axis cultures and rooting in cotyledon cultures (Fig 3). MS+IBA+BAP

+CW also induced shooting in embryo axis explants but rooting was not observed in cotyledon cultures (Fig 4; Table 1). Further elongation of shoot was also not observed. Since commencement of experiments rooting dominated over shooting. The developed roots were white, long and arise from the entire cut edge of cotyledon explants.

One auxin (IBA) and one cytokinin (BAP or KN) was necessary to induce shooting in embryo explants. Because single phytohormone showed little or no response on shooting, 2, 4-D in the medium callus and roots but did not inhibit shooting in embryo axis culture. When Ascorbic acid was supplemented to MS+IBA+BAP+KN the explants exude light greenish yellow pigment resulting in cessation or morphogenetic response (Table 1).

The above results suggest that organogenesis in both types of explants is different together with their nutrient requirements. Further work is required to induce regeneration in this self compatible rare plant hence the present experiments are significant. Further organogenesis on other medicinal Apocyanaceae and *Plumeria* will enable better understanding of the morphogenetic events in recalcitrant latex producing plants.

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