

Short communication

Development of protocorm-like bodies and shoots in *Dendrobium* cv. Sonia following gamma irradiation

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Abstract

This paper reports a preliminary attempt to evolve mutant varieties of *Dendrobium* with desirable traits. *Dendrobium* cv. Sonia was subcultured on half-strength MS medium supplemented with BA 1.0 mg L⁻¹ and NAA 1.0 mg L⁻¹ to induce production of protocorm-like bodies (PLBs). The PLBs were exposed to different doses of gamma rays (20 Gy and 30 Gy) and re-irradiated from a cobalt 60 source. The highest number of shoots was observed at 20 Gy (3.4). A comparison of three levels of BA with ½ MS media on the irradiated protocorms showed more PLB production at 0.5 mg L⁻¹ while shoot production was higher at 1.5 mg L⁻¹.

Keywords: *In vitro* mutagenesis, Additive effects.

Dendrobium cv. Sonia is a commercial orchid cultivar widely grown in Kerala. To evolve *Dendrobium* varieties with desirable flower colour, shape, and size, induced mutation by gamma irradiation was attempted. This follows the work of Thammasiri (1996) and Angamnuasiri (2001), who reported the development of mutant *Dendrobium* hybrids in Thailand through gamma radiation. We irradiated the protocorm-like bodies (PLBs) of *Dendrobium* cv. Sonia with gamma rays and monitored the development of PLBs and shoots. To verify whether irradiation led to changes in the physiological activities of the cells, PLB multiplication and shoot production under varying BA levels were also assessed.

The experiment was conducted during 2003–'06. Freshly formed shoots (excised and soaked in 0.1% w/v of Labolene for 30 min. followed by thorough rinsing in running tap water) were used as the source of explants. They were surface sterilized with 0.08% HgCl₂ for 10 min. and rinsed thoroughly with sterile double-distilled water. The explants were inoculated on Vacin and Went

medium containing 3% sucrose and 0.7% agar (Bacteriological grade, Hi media Laboratories, Mumbai) in culture tubes (25 x 150 mm) plugged with absorbent cotton. The pH of the medium was adjusted to 5.7 prior to autoclaving at 108 Kpa and 121°C for 15 min. The cultures were maintained at 25±2°C and 16 h photoperiod (40 µmol m⁻² s⁻¹) provided by cool white fluorescent tubes. They were then subcultured on half strength MS medium supplemented with BA (1.0 mg L⁻¹) and NAA (1.0 mg L⁻¹) to induce production of protocorm-like bodies (PLBs). The PLBs were then multiplied in basal medium supplemented with BA (1.0 mg L⁻¹) and 7.5% coconut water. Pilot experiments were set up exposing 10 PLBs in six replicates to different doses of gamma rays from a cobalt 60 source at a dose rate of 10 Gy per min. at the Radiotracer Laboratory, Vellanikkara, Thrissur. The irradiation doses were fixed at 20 Gy and 30 Gy of gamma rays, as the PLBs showed the highest rate of multiplication and shoot development at these levels. To evaluate the additive effects, if any, the PLBs irradiated with 20 Gy and 30 Gy of gamma rays were re-

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irradiated with 20 Gy and 30 Gy of gamma rays one week later. Effect of three different levels of BA viz. 0.5 mg L⁻¹, 1.0 mg L⁻¹, and 1.5 mg L⁻¹ on the development of PLBs and shoots was also assessed on the irradiated protocorms. The data were analysed using ANOVA for completely randomized design.

There was no significant difference among the irradiation treatments in terms of the number of PLBs produced (Table 1). The multiplication rate of PLBs ranged from 27.4 at 30+20 Gy to 49.4 at 20 Gy of gamma rays. Browning of PLBs was observed in the additive

Furthermore, higher numbers of PLBs were produced at BA 0.5 mg L⁻¹ and more shoots at BA (1.5 mg L⁻¹), implying that at the lowest level of BA tried, irradiation did not alter the physiological response of tissues to cytokinin.

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Table 1. Effects of gamma irradiation and graded doses of BA on multiplication of protocorm-like bodies (PLBs) and development of shoots in *Dendrobium* cv. Sonia.

Treatments	Additive effects of gamma rays		Concentration of BA					
			½ MS+BA 0.5 mg L ⁻¹		½ MS+BA 1.0 mg L ⁻¹		½ MS+BA 1.5 mg L ⁻¹	
	PLBs	shoots	PLBs	Shoots	PLBs	Shoots	PLBs	Shoots
20 Gy	49.4	3.4	22.0	22.6	27.4	17.4	36.2	16.8
30 Gy	37.4	2.0	37.8	18.6	29.8	13.8	28.4	16.4
20+20 Gy	31.2	0.4	40.8	9.0	36.0	11.2	30.8	5.6
20+30 Gy	29.2	1.2	41.2	11.2	32.8	9.0	40.6	8.0
30+20 Gy	27.4	1.0	27.2	19.2	38.0	24.4*	52.8*	28.0*
30+30 Gy	32.0	0.2	60.0*	25.6	57.8*	28.2*	58.4*	29.2*
Control	36.2	9.8*	26.0	21.2	34.6	16.2	17.2	15.6
CD (0.05)	NS	3.41	17.32	NS	17.54	10.15	14.5	10.43

Values after '+' sign under treatments indicate re-irradiation doses; NS= not significant; *significant at 5%.

treatments. At 20+20 Gy of gamma rays, the size of PLBs was also reduced. Shoot formation in all the treatments was lower than the control (9.8). This is consistent with the findings of Adrain (1999) who showed that regeneration of plantlets from PLBs after gamma irradiation declined with higher doses in *Mokara* Chark Kuan and *Dendrobium* Jacky. A comparison of the data in Table 1 also indicates that at all the three levels of BA, more PLBs and shoots were produced in the additive treatment of 30+30 Gy followed by 30+20 Gy.

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