

In Vitro Rooting of Banana Musa (AA) 'Kadali' through in Vitro Male Bud Culture

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Abstract: Male buds of banana can be used as a potential explant for in vitro shoot regeneration. The present investigation was undertaken to study the effect of different concentrations of sucrose and IBA on rooting in Kadali variety of banana. The shoots which are developed on Murashige and Skoog medium (full MS and half MS) was inoculated on same medium supplemented with combinations of sucrose (1.5 % and 3.0%) with IBA (1.0, 2.0, 3.0 and 4.0 mgL⁻¹). With regard to different treatments, results revealed that banana cv. Kadali recorded minimum number of days for root initiation and produced higher number of roots in full MS medium with 3 per cent sucrose and IBA 1.0 mgL⁻¹. The longest root was observed in the combination of half MS with 1.5 per cent sucrose and IBA 3.0 mgL⁻¹ and 3.0 per cent sucrose and IBA 1.0 mgL⁻¹ under full MS. Well rooted plants were hardened after potting in pro trays containing 1:1 (v/v) sterilized coco peat and vermiculite. After one month of planting out, 90 per cent of the plants were found to survive.

Key words: Kadali, Musa, Male bud, MS media, Tissue culture

In Kerala, Kadali is one of the banana varieties in great demand as it is used for offering in temples, and the fruits are bestowed with a special flavour besides possessing medicinal properties. The major problem faced by the banana farmers growing Kadali is the non availability of good quality planting material. Normally suckers and tissue culture plants (raised through shoot tip culture) are used for planting, the availability of which are limited. Further, contamination is a problem in shoot tip culture. In addition to suckers, male buds can be used as a potential explant for raising tissue culture plants as this part goes unutilized during the harvesting of banana bunches. Therefore, *in vitro* male bud culture forms a good alternative since the male buds are less contaminated and more number of explants can be obtained from one male bud. *In vitro* propagation of different cultivar required different culture media for shoot proliferation and root differentiation. However, there is still lack of information on *in vitro* rooting of banana. Now a days, the plant growth regulators are widely used

in modern agriculture to promote rooting. Hence the present investigation was undertaken to standardize different concentrations of sucrose and IBA in full MS and half MS media for *in vitro* rooting of male buds of Kadali.

The present study was carried out at the Plant Tissue Culture Laboratory of Banana Research Station, Kannara during 2017-2018. Healthy male flower buds were collected 20-25 days after the emergence of the bunch from the field. The bracts associated with hands of male buds were removed in a stepwise manner upto 35-45 according to the size of male bud without making any injury until it became too small in non-sterile conditions. Male buds reduced to a size of 4-5cm length were washed four times thoroughly in tap water. The explants were then surface sterilized with 0.1 per cent (w/v) mercuric chloride for 4 minutes followed by three rinses in autoclaved double distilled water in the laminar air flow cabinet. Protective bracts about 45-50 corresponding to the male flowers were removed by using sterilized blade and forceps and male

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flowers having size 0.5-1.0 cm were taken out and inoculated in full MS and half MS (Murashige and Skoog) medium supplemented with different combinations of naphthalene acetic acid (NAA) and benzyl adenine (BA) (Murashige and Skoog, 1962). The experiment was laid out in CRD. Each treatment was replicated thrice with 6 cultures for each replication. Cultures were incubated at 27°C in an air conditioned culture room with a 16 hour photoperiod of light intensity 2000 lux supplied by cool day light fluorescent tubes. After culturing for 30 days on full MS and half MS media, the explants were transferred to fresh media every three weeks until the formation of shoot buds. Axillary shoots of 1.0-1.5 cm length from established culture were used as explant for shoot multiplication in full MS and half MS media containing different combinations of naphthalene acetic acid (NAA) and Benzyl adenine (BA) for further multiplication. The multiple induced shoots were separated into clump sizes of two to three shoots per clump and subcultured at intervals of 30 days. For induction of roots, the shoots were separated and transferred on full MS and half MS media supplemented with sucrose and IBA after 90 days of culturing. Observations for number of days taken for root initiation, number of roots and length of longest root (cm) were recorded on six cultures per treatment after four weeks of culturing.

Table 1
Effect of growth regulators on number of days taken for root initiation, number of roots and length of longest root.

Treatments	Number of days taken for root initiation	Number of roots	Length of longest root (cm)
Half MS+ sucrose 1.5 + IBA 1.0	5.33	3.00	5.66
Half MS+ sucrose 1.5 + IBA 2.0	5.66	3.16	4.63
Half MS+ sucrose 1.5 + IBA 3.0	6.49	3.66	9.99
Half MS+ sucrose 1.5 + IBA 4.0	7.33	3.66	4.24
Half MS+ sucrose 3.0 + IBA 1.0	5.16	4.99	8.33
Half MS+ sucrose 3.0 + IBA 2.0	6.99	4.49	6.64
Half MS+ sucrose 3.0 + IBA 3.0	6.66	3.49	4.74
Half MS+ sucrose 4.0 + IBA 4.0	6.6	3.49	4.78
Full MS+ sucrose 1.5 + IBA 1.0	5.16	1.83	2.66

Full MS+ sucrose 1.5 + IBA 2.0	6.99	2.83	4.19
Full MS+ sucrose 1.5 + IBA 3.0	6.99	2.49	5.93
Full MS+ sucrose 1.5 + IBA 4.0	6.99	4.50	2.33
Full MS+ sucrose 3.0 + IBA 1.0	4.66	8.33	8.99
Full MS+ sucrose 3.0 + IBA 2.0	6.16	5.83	4.33
Full MS+ sucrose 3.0 + IBA 3.0	6.66	3.50	2.49
Full MS+ sucrose 4.0 + IBA 4.0	7.00	4.49	1.58
CD (0.05)	0.68	1.07	1.47

*Data were collected after 30 days of rooting. Values represent mean of three replications, with six explants per replication.

Spontaneous root formation rarely occur in some cultures under *in vitro* conditions. Moreover, shoots of majority of the species did not develop root system during multiplication stage of cultures. Rooting can be achieved either by subculturing to a medium lacking cytokinin, with or without a rooting hormone or by treating the shoots as conventional cuttings after removal from the sterile culture (Yeoman, 1986). Table 1 reveals that among various treatments minimum number of days taken for root initiation (4.66) was noticed in treatment combination of 3 per cent sucrose with IBA 1.0 mgL⁻¹ in full MS medium which was on par with the same media supplements in half MS (5.16) and also combination of sucrose at 1.5 per cent with IBA 1.0 mgL⁻¹ under full MS (5.16). Highest number of roots (8.33) were recorded in culture containing 3 per cent sucrose with 1.0 mgL⁻¹ IBA under full MS and was superior to all the other treatment combinations. A study conducted by Hussein (2012) on the effect of sucrose in MS medium in banana cultures reported an increase in plantlet fresh weight at sucrose 3.0 mgL⁻¹. IBA at 1 µM or even at 10-50 µM were also found to be effective in rooting of *Musa* cultivars (Dore Swamy *et al.*, 1983; Mante and Tepper, 1983; Banerjee and De Langhe, 1985; Vuylsteke and De Langhe, 1985). MS medium supplemented with different concentrations of IBA is best for good rooting of banana (Raut and Lokhande, 1989; Khanam *et al.*, 1996). The results were in confirmity with a study, where different concentrations of IBA were used in rooting of male bud culture of banana cv. Sabri. The best rooting was noticed in medium supplemented with IBA 1.0 mgL⁻¹ (Sultan *et al.*, 2011). The longest root was observed in the combination of half MS with 1.5 per cent sucrose

and IBA 3.0 mgL⁻¹ which was on par with 3.0 per cent sucrose and IBA 1.0 mgL⁻¹ under full MS. Cultures from male flowers were found to respond well to rooting hormones like IBA and NAA (Sultan *et al.*, 2011). Induction of rooting in *in vitro* culture of banana cultivar Sabri (AAB) was observed in 1.0 mgL⁻¹ of IBA (5 roots per shoot) and NAA (4 roots per shoot). When all the above observations were pooled together, the combination of full MS with sucrose 3 per cent and IBA 1.0 mgL⁻¹ was found to be the best medium for *in vitro* rooting.

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