



RESEARCH ARTICLE

STUDIES ON SHOOT AND ROOT INDUCTION OF EIGHT COMMERCIAL BANANA VARIETIES OF WEST BENGAL BY MACRO-PROPAGATION TECHNIQUE

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ABSTRACT

For mass multiplication of healthy planting materials, shoot and root were successfully induced by macro-propagation technique from eight commercial banana varieties grown in West Bengal, viz., Grand Naine (AAA), Robusta (AAA), Martaman (AAB), Champa (AAB), Kanthali clone-1 (ABB), Bagda Kanthali (ABB), Baish Chhara (ABB) and Behula (ABB). Corms prepared by decapitation and pairing of healthy suckers (4-5 months old) were planted in growth media bed (containing 2 kg sawdust + 30g *Trichoderma viride*/*Bacillus subtilis* + 30g VAM per corm) and treated weekly with 25 ppm 6-benzylaminopurine (BAP) solution. However, each variety was significantly different from others with respect to induction time and number of primary, secondary and tertiary shoots induced and the number and length of primary and secondary roots induced. The time required for induction of primary shoot varied from 19.75 days in Grand Naine to 28.25 days in Bagda variety. The number of induced primary shoot was recorded higher (3.23 to 3.40/corm) in varieties under AAA group, compared to the AAB (2.70-2.90 shoots/corm) and ABB groups (2.20-2.50 shoots/corm). Similar trends were also recorded for induction of secondary and tertiary shoots. The number of tertiary shoots produced per corm was maximum (24.23) in Grand Naine and minimum (9.61) in Kanthali variety. The tertiary shoots were separated from corms at 2-3 leaves stage and planted in sterile sand bed by basal treatment with rooting hormone (IBA) @ 5 ppm. Two weeks after treatment, primary roots were induced, which was higher (2.20-2.65/plantlet) in the varieties under AAA group, compared with 1.23 to 2.29/plantlet in AAB and ABB groups. These plants were planted in polythene packets containing hardening mixture and hardened for 60 days under shade net house. After hardening, the number of primary and secondary roots were recorded maximum (10.80 and 35.50/plant, respectively) in Grand Naine and minimum (9.20 and 31.60/plant, respectively) in Baish Chhara variety.

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INTRODUCTION

Banana is an important commercial fruit crop in West Bengal, with annual production of 1077.80 thousand tones and productivity of 24.10 t/ha from 44.70 thousand hectare cultivation (Anon, 2014). The commercial varieties grown in West Bengal are Grand Naine, Robusta, Martaman, Champa, Kanthali, Bagda, Baish Chhara, Behula, etc. The commercial cultivation of banana in any region is greatly hindered by biotic and abiotic factors and for many cases, selection of healthy planting materials, free from insect pests and diseases is very important (Singh, 2008, Uma *et al.*, 2008, Kasyoka *et al.*, 2010). To meet this demand, tissue culture banana plant has been introduced for commercial adoption, which appears to be very low due to high costs, capital requirements and

skills involved (Kasyoka *et al.*, 2011). In India, only 4.0% of the demand is met by tissue culture plant, while the rest 96% of planting material requirement is being catered as conventional suckers (Uma *et al.*, 2008). The macropropagation technology has been reported to be an alternative, farmer's friendly, cost effective technology for producing healthy planting materials of banana (Njau *et al.*, 2011, Njukwe *et al.*, 2012, Dayarani *et al.*, 2013, Debnath *et al.*, 2015). With this background, an experiment was carried out at the research units of ICAR-AICRP on Fruits and RKVY project on Banana Fibre, Bidhan Chandra Krishi Viswavidyalaya (SAU), Mondouri, Nadia, West Bengal, India during 2015-16 to study the induction of shoot and root of eight commercial banana varieties grown in West Bengal by macropropagation technique.

MATERIALS AND METHODS

The experiment was carried out at the research units of ICAR-AICRP on Fruits and RKVY project on Banana Fibre, Bidhan

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Chandra Krishi Viswavidyalaya (SAU), Mondouri, Nadia, West Bengal, India during 2015-16. The technical procedures of macropropagation, as described by Uma *et al.* (2008), Anon (2015) and Debnath *et al.* (2015) were followed for this experiment on eight commercial banana varieties, viz., Grand Nain (AAA), Robusta (AAA), Martaman (AAB), Champa (AAB), Kanthali clone-1 (ABB), Bagda Kanthali (ABB), Baish Chhara (ABB) and Behula (ABB). The experiment was laid out in a Randomized Block Design (RBD) with 4 replications, 8 treatments (varieties) and 10 corm per replication. Healthy suckers were collected from the well-maintained healthy germplasm block. Suckers were cleaned, paired, treated with protective chemicals, decapitated (removal of apical bud) and planted in growth media beds containing 2 kg sawdust + 30g *Trichoderma viride*/Bacillus subtilis + 30g VAM per corm. Application of 4 ml BAP solution (25 ppm) per corm per week and light moistening of growth media beds were done for activation of lateral buds and induction of primary shoots. The same technique, i.e., decapitation and BAP treatment were applied on primary shoots to induce secondary and on secondary shoots to induce tertiary shoots. The tertiary shoots were separated from corms and planted in sterile sand bed by basal treatment with rooting hormone @ 5 ppm. These plantlets were planted in polythene packets containing hardening mixture and hardened for 60 days. Observations were recorded on induction time (days) for primary, secondary and tertiary shoots and its growth characters, viz., height, girth, leaf number, length and width of leaf. Regarding root induction, observations were recorded on number and length of primary and secondary roots at 15 days after separation of tertiary shoots followed by root hormone treatment and 60 days of hardening. The data were analyzed for statistical inference following the statistical method for Randomized Block Design (RBD) described by Gomez and Gomez (1983).

RESULTS AND DISCUSSION

Induction of primary, secondary and tertiary shoots

The data recorded on induction of primary, secondary and tertiary shoots have been presented in tables 1, 2 and 3. Significant variations were recorded on induction time and number of induced primary, secondary and tertiary shoots. It took 19.75 to 20.25 days for primary shoot induction in the varieties under AAA group, which increased to 25.25 to 25.50 days in AAB group and it was maximum (26.50 to 28.25 days) in ABB group. The varieties under the AAA genomic group i.e., Grand Nain and Robusta responded well to induction of primary shoots and produced 3.23 to 3.40 shoots per corm, compared to the AAB genomic group (2.70 to 2.90 shoots/corm) and ABB genomic group (2.20 to 2.50 shoots/corm). Induction of secondary shoot from primary shoot and tertiary shoot from secondary shoot were achieved by decortications (removal of apical bud) of primary shoot and secondary shoot at 2 to 3 leaves stage and application of 4 ml of 40 ppm BAP solution. Minimum time (27.00 to 27.50 days) was taken for secondary shoot induction in the varieties under AAA group which increased to (29.75 to 31.00 days) under AAB group and maximum (33.50 to 34.25 days) under ABB group. Induction of secondary shoot per corm was lowest at (4.42 per corm) in Baish Chhara variety and maximum in (8.15 per corm) in Grand Nain variety. Different varieties took different time for induction of tertiary shoot which varied from 28.00 days in Grand Nain to 35.50 days in

Bagda variety. Similarly, the tertiary shoot induced per corm varied between 9.61 tertiary shoots per corm in Kanthali variety to 24.23 tertiary shoots per corm in Grand Nain variety. The varieties under AAA group responded well to induction of tertiary shoots compared to AAB and ABB group. These results corroborated the findings by Debnath *et al.* (2015). According to Ortiz and Vuylsteke (1994), apical dominance, i.e., the inhibition of lateral bud growth due to growth substances released by the terminal bud, has been considered as a limiting factor for the perennial productivity of plantains (*Musa* spp., AAB group). By decapitation and decortications of the corms in present experiment, the apical or terminal buds were removed and hence, the inhibition of lateral buds was removed. Application of BAP (*6-Benzylaminopurine*), a synthetic cytokinin was done for activation of the lateral buds. The effectiveness of benzylaminopurine (BAP) for shoot multiplication was known in banana (Muhammad *et al.*, 2007). These two factors (removal of apical dominance and application of BAP) appeared responsible for activation of lateral buds and induction of primary, secondary and tertiary shoots in the present experiment. However, the differences in multiplication or production of shoots in different varieties may be due to the differences in the uptake rates of cytokinin by different genomes (Blakesly, 1991), variations in translocation rates to meristematic regions and type of metabolic processes take place (i.e., cytokinin may be degraded or conjugated with sugars or amino acids to form biologically inert compounds) (Kaminek, 1992).

Growth of primary, secondary and tertiary shoots

The growth of the primary, secondary and tertiary shoots showed significant variations only in height of plantlets of different varieties and non-significant variations in girth, leaf number and size (length & width) of leaf per plantlet (Tables 1, 2 & 3). The height of primary shoot varied from 12.15 cm in Grand Naine variety to 16.08 cm in Bagda variety, while the girth of primary shoot varied from 5.83 cm in Martaman variety to 6.69 cm in Champa variety. The height of the induced tertiary shoots varied from 6.49 cm in Grand Naine variety to 8.93 cm in Bagda variety. The girth of tertiary shoot was lowest (2.61 cm) in Robusta and maximum (3.55 cm) in Bagda variety. The Martaman variety recorded maximum (2.70) leaf number per tertiary shoot and minimum (2.15) leaf per tertiary shoot in Champa variety. The tertiary shoot of Bagda variety recorded maximum length (7.98 cm) and width (4.10 cm) of leaf, while it was minimum (6.10 and 3.18 cm) in Robusta variety. The observed growth of shoots of different varieties were supposed to be a reflection of varietal characters and also supported by the constituents of the culture media, viz., VAM and *Trichoderma viride* which are reported for beneficial effects on growth of banana plant (Sabarad *et al.*, 2004).

Induction of primary and secondary roots

The tertiary shoots were separated from the corm at 2-3 leaf-stages and its basal portion were treated with 5 ppm IBA solution and planted on sand bed for induction of roots. The data recorded on induction of primary and secondary roots have been presented in table 4. Significant variations were recorded on the number and length of primary and secondary roots induced at 15 days after separation and treatment with rooting hormone and at 60 days of hardening. The induction of primary root was maximum (2.65/plantlet) in Grand Naine, while it was minimum (1.23/plantlet) in Baish Chhara variety.

Table 1. Induction of primary shoot (PS)- time taken for induction; number, height and girth of induced PS; number, length and width of leaf per PS

Variety of banana	Induction time for PS (Days)	No. of PS induced per corm	Height of PS (cm)	Girth of PS (cm)	Leaf No. per PS	Length of leaf (cm)	Width of leaf (cm)
Grand Nain (AAA)	19.75	3.40	12.15	6.46	2.40	16.25	6.03
Robusta (AAA)	20.25	3.23	12.73	6.35	2.78	15.13	5.98
Martaman (AAB)	25.50	2.90	14.95	5.83	2.50	17.00	7.80
Champa (AAB)	25.25	2.70	15.75	6.69	2.55	17.35	7.03
Kanthali (ABB)	27.75	2.40	14.94	6.23	2.20	18.25	8.13
Bagda (ABB)	28.25	2.50	16.08	6.43	2.35	18.90	8.28
Baish Chhara (ABB)	26.50	2.20	14.85	6.37	2.55	17.63	7.20
Behula (ABB)	28.00	2.45	14.80	6.40	2.33	17.54	7.10
SEm (\pm)	2.263	0.274	0.721	0.536	0.262	1.235	0.795
C. D. at 5%	4.706	0.571	1.501	NS	NS	2.569	1.654

Table 2. Induction of secondary shoot (SS)- time taken for induction; number, height and girth of induced SS; number, length and width of leaf per SS

Variety of banana	Induction time for SS (Days)	No. of SS induced per PS	No. of SS induced per corm	Height of SS (cm)	Girth of SS (cm)	No. of Leaf per SS	Length of leaf (cm)	Width of leaf (cm)
Grand Nain (AAA)	27.00	2.40	8.15	10.80	5.43	2.83	11.90	3.98
Robusta (AAA)	27.50	2.30	7.40	10.83	4.90	2.48	11.50	3.93
Martaman (AAB)	29.75	2.20	6.42	11.98	5.03	2.48	13.88	4.23
Champa (AAB)	31.00	2.00	5.46	11.08	4.90	2.50	14.30	4.18
Kanthali (ABB)	31.75	2.10	5.09	13.44	5.47	2.13	14.48	5.30
Bagda (ABB)	34.25	2.00	4.92	14.43	5.45	2.25	14.78	5.60
Baish Chhara (ABB)	33.50	2.03	4.42	12.23	4.71	2.53	14.50	5.33
Behula (ABB)	33.25	2.05	4.94	13.93	4.80	2.50	14.30	5.13
SEm (\pm)	2.184	0.300	0.823	0.805	0.285	0.213	1.141	0.637
C. D. at 5%	4.543	0.624	1.711	1.674	NS	NS	NS	NS

Table 3. Induction of tertiary shoot (TS)- time taken for induction; number, height and girth of induced TS; number, length and width of leaf per TS

Variety of banana	Induction time for TS (Days)	No. of TS induced per SS	No. of TS induced per corm	Height of TS (cm)	Girth of TS (cm)	Leaf No. per TS	Length of leaf (cm)	Width of leaf (cm)
Grand Nain (AAA)	28.00	3.00	24.23	6.49	3.02	2.48	6.90	3.18
Robusta (AAA)	29.00	2.70	19.78	6.53	2.61	2.32	6.10	3.18
Martaman (AAB)	31.00	2.50	16.25	7.35	3.33	2.70	7.53	3.83
Champa (AAB)	31.50	2.30	12.89	7.23	3.13	2.15	7.65	3.80
Kanthali (ABB)	33.25	2.10	10.92	7.93	3.49	2.27	7.83	3.90
Bagda (ABB)	35.50	2.20	10.76	8.93	3.55	2.28	7.98	4.10
Baish Chhara (ABB)	34.50	2.20	9.61	8.01	3.45	2.23	7.88	3.95
Behula (ABB)	34.50	2.15	10.59	7.86	3.40	2.18	7.70	3.93
SEm (\pm)	2.340	0.261	2.577	0.541	0.284	0.200	0.580	0.313
C. D. at 5%	4.866	0.543	5.361	1.124	NS	NS	NS	NS

Table 4. Induction of primary root (PR) and secondary root (SR) per plant at 15 days after separation and treatment with rooting hormone and at 60 days of hardening

Variety of banana	15 days after separation and treatment with rooting hormone				60 days of hardening			
	No. of PR	Length of PR (cm)	No. of SR	Length of SR (cm)	No. of PR	Length of PR (cm)	No. of SR	Length of SR (cm)
Grand Nain (AAA)	2.65	1.70	7.11	4.36	10.80	19.40	35.50	21.60
Robusta (AAA)	2.20	1.48	6.90	4.05	10.40	18.40	34.80	21.90
Martaman (AAB)	2.29	1.56	6.98	4.43	10.50	18.70	33.20	23.10
Champa (AAB)	1.55	1.23	6.30	3.20	9.70	17.70	33.00	22.80
Kanthali (ABB)	1.83	1.28	6.38	3.46	9.30	17.90	32.10	24.90
Bagda (ABB)	2.00	1.35	6.73	3.89	9.60	18.00	32.40	25.30
Baish Chhara (ABB)	1.23	1.00	4.85	2.80	9.20	17.10	31.60	24.70
Behula (ABB)	1.30	1.20	5.78	3.03	9.20	17.20	31.80	24.40
SEm (\pm)	0.219	0.170	0.512	0.391	0.149	0.166	0.538	0.591
C. D. at 5%	0.456	0.353	1.064	0.812	0.311	0.346	1.120	1.230

The banana varieties under AAB and ABB groups recorded lower (1.23-2.29/plantlet) induction of primary roots, compared with the varieties under AAA group (2.20-2.65/plantlet). The length of induced primary root also showed significant variation due to different varieties, which varied from 1.00 cm in Baish Chhara to 1.70 cm in Grand Naine variety. Similar variations were recorded in the number and length of secondary roots due to different varieties. Length of induced secondary root was lowest (2.80 cm) in Baish Chhara and maximum (4.43 cm) in Martaman variety. Induction and promotion of rooting *in vitro* in banana have been reported due to auxins category of hormones (Hussein, 2012). In present experiment, eight varieties of banana also showed better response to IBA treatment for rooting of plantlets. After hardening for 60 days under shade net house (75% shade), the number of primary and secondary roots were recorded maximum (10.80 and 35.50/plantlet, respectively) in Grand Naine and minimum (9.20 and 31.60/plantlet, respectively) in Baish Chhara variety. The length of primary root after hardening varied from 17.10 cm in Baish Chhara to 19.40 cm in Grand Naine variety, while the length of secondary root varied from 21.60 cm in Grand Naine to 25.30 cm in Bagda variety. Satisfactory hardening and acclimatization of tissue culture plantlets of different banana varieties have also been recorded by Patel *et al.* (2015), using hardening mixture of clay + sand + FYM and 75% shade net house.

Conclusion

From the present study, it was concluded that all the major commercial banana varieties grown in West Bengal were responded to macropropagation technique for satisfactory induction of shoot and root.

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