Effect of biofertilizers on macro-propagation of banana

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ABSTRACT

Banana has strong apical dominance (harmone-mediated); when suppressed it gives rise to many side suckers. This principle was used in macro-propagation in stimulation of lateral buds development. Plantlet production is generally accomplished through decapitation methods in banana. In the present study attempts were made to enhance the efficacy of decortication in cvs Grand Naine and Shrimanti by using biofertilizers and plant hormones. The study was carried out with suckers weighing 1.0-1.5 kg and composted sawdust was used as initiation medium (substrate). The treatment comprising sawdust + VAM (30 g) + BAP 40 ppm (4 ml) + *Bacillus substillis* (30 g) produced the maximum number of primary buds (2.99) followed by sawdust + BAP 40 ppm (4 ml) + *B substillis* (30 g) with primary buds 2.59. In case of tertiary bud production, treatment comprising sawdust + BAP 40 ppm (4 ml) + *B substillis* (30 g) resulted in the highest number of buds (6.71) followed by sawdust + IBA (dipping in 0.25% solution) + *Azospirillum* (30 g) (6.12) and sawdust + VAM (30 g) + BAP 40 ppm (4 ml) + *B substillis* (30 g) (26.30) followed by sawdust + IBA (dipping in 0.25% solution) + *Azospirillum* (30 g) (23.07) and sawdust + VAM (30 g) + BAP 40 ppm (4 ml) + *B substillis* (30 g) (26.30) followed by sawdust + IBA (dipping in 0.25% solution) + *Azospirillum* (30 g) (23.07) and sawdust + VAM (30 g) + BAP 40 ppm (4 ml) + *B substillis* (30 g) (16.73) suggesting that treatment combinations of VAM, *B substilis* + BAP and IBA were effective for macro-propagation of cv Grand Naine and Shrimanti.

Keywords: Banana; macro-propagation; decortications; sawdust; BAP

INTRODUCTION

In most banana cultivars the emergence of new suckers is somewhat slow due to strong hormonemediated apical dominance exerted by the main plant. Micro-propagation (ie meristem/tissue culture) assures more rapid production of planting materials. These planting materials are healthy, vigorous and free from pests and diseases (Swennen 1990) but due to the large capital investment required for creation of tissue culture laboratory with sophisticated technique, skill and care to handle (Vuylsteke1998) the plantlets produced are fairly expensive which are not easily affordable by small and marginal farmers. Hence there is need for a cheap and easy technique that increases the sucker multiplication with minimum technical skill and is cost effective, easy and cheap. This technique is known as macro-propagation. Macro-propagation has been developed as a farmer friendly technology to cater to the growing needs of clean planting material (Uma et al 2010). In the present investigations the effect of plant hormones in improving the bud production

capacity of popular cultivars Grand Naine and Shrimanti has been studied. This technology can use whole suckers or sword suckers to produce planting material. Repression of apical meristem stimulates the regeneration of lateral meristem (Uma et al 2010).

Increased suckering rate can be achieved through complete/partial decapitation on a field-grown plant or through detached corm technique (Baiyeri and Aba 2007) and sawdust is the best substrate over others like rice hull, sand etc with higher water holding capacity (Baiyeri and Aba 2007). The attempts have been made to enhance the rate of plantlet production through macro-propagation by the addition of biofertilizers (VAM, *Trichoderma viride*, *Bacillus subtilis* and *Azospirillum*) and phyto-hormones (BAP and IBA) to the explant/substrate.

MATERIAL and METHODS

The present study was carried out for three years from 2012 to 2015 at AICRP on Banana, Banana

Research Station, Jalgaon, Maharashtra with six treatments [T_1 : Sawdust + VAM (30 g), T_2 : Sawdust + Trichoderma viride (30 g), T_3 : Sawdust + VAM (30 g) + Tviride (30 g), T_4 : Sawdust + IBA (dipping in 0.25% solution) + Azospirillum (30 g), T_5 : Sawdust + BAP 40 ppm (4 ml) + Bacillus substillis (30 g), T_6 : Sawdust + VAM (30 g) + BAP 40 ppm (4 ml) + B substillis (30 g)] in a completely randomized design with two replications with the objective of standardization of macro-propagation technique for banana for the production of quality planting material and also standardization of varietal response of two cultivars namely Grand Naine (V_1) and Shrimanti (V_2) through macro-propagation technique.

Under decortications, the field extracted suckers were detopped just above the juncture of the corm and the aerial shoot and under decapitation the apical meristem was removed to a depth of 2 cm leaving a cavity of 2 cm diameter in the rhizome and the rest of the corm was given 6-8 crosswise cuts. The decapitated corms were planted individually in the bed filled with sawdust leaving 5 cm from the top. Corms were buried 5 cm deep in the substrate and respective treatments were imposed and covered with sawdust up to a height of 2 cm.

Biofertilizers and plant growth hormones were used as additives. Indole butyric acid (IBA) in T₄ treatment was given by dipping the corm region of decorticated suckers in 0.25 per cent IBA solution for 20 min prior to planting. In the treatments T_5 and T_6 the apical meristem was removed to a depth of 2 cm near the crown region; the corms were given 4-6 transverse incisions to a depth of 2 mm and 4 ml of 40 ppm BAP was poured into the cavity left by the removal of the apical meristem. The same treatment was imposed during the primary and secondary decapitation stages. The suckers were fully covered by sawdust to prevent exposure to direct sunlight. Four different biofertilizers were in combination to improve the bud proliferation rate at primary and secondary decapitation stages. Thirty grams of each of the biofertilizers was mixed with the substrate before planting. After primary decortication the emerging shoots were allowed to grow for 25-30 days and when they attained three-leaf stage (height 15-20 cm, stem girth 2.5 cm), the secondary decapitation was imposed. The aerial portion of the plantlets was decapitated; juvenile meristem was removed and 4-6 horizontal incisions were given for the young rhizome and covered with sawdust.

The same procedure was repeated for secondary and tertiary decapitations. At the end of tertiary bud stage, the corms were removed from the substrate and washed carefully. Each plantlet was separated so as to retain at least 2-3 roots and separated plantlets were hardened in mixture of red soil, sand and farmyard manure (1:1:1) filled in polybags with drainage holes. Plantlets were watered sufficiently and maintained under shade for 45 days. Time taken for bud (primary buds) regeneration from the decorticated suckers was recorded. Total number of primary, secondary and tertiary buds formed at the end of 3rd month was recorded. Other parameters measured were plant height, pseudostem girth and number of leaves at hardening stage (Sajith et al 2014)

RESULTS and DISSCUSSION

There were significant differences among treatments for the time taken for bud initiation under both varieties. Significantly the least number of days for bud initiation (18.70) was recorded in cv Shrimanti (V_2) under treatment T_5 (16.79) and 16.47 days in the interaction of V_1T_5 ie Grand naine + sawdust + BAP (4 ml) + B substillis (30 g).

There was no significant difference between two varieties for number of days taken for bud initiation. Among the various treatments, T_s [Sawdust + BAP 40] ppm (4 ml) + Bacillus substillis (30 g)] and T₆ [Sawdust + VAM (30 g) + BAP 40 ppm (4 ml) + B substillis (30 graphs)g)] took minimum (16.79 and 17.47 respectively) and T_3 [Sawdust + VAM (30 g) + T viride (30 g)] took maximum (21.07) number of days for bud initiation. Under the interaction effect V₁T₅, V₁T₆, V₂T₅, V₂T₆ and V₂T₂ were at par in taking minimum number of days for bud initiation. Number of primary buds was more (2.33) in V₂ (Shrimanti) as compared to 2.02 in V₁ (Grand Naine). Among the treatments, T₆ resulted in significantly more number of primary buds (2.99) followed by T_5 (2.59). In case of interactions, V_2T_6 and V₁T₆ resulted in maximum number of primary buds (3.16 and 2.84 respectively). There was no significant difference between two varieties for number of secondary buds.

Among treatments T_5 resulted in maximum number of secondary buds (3.38) followed by T_4 [Sawdust + IBA (dipping in 0.25% solution) + *Azospirillum* (30 g)] (2.90). Under interactions, V_1T_5 and V_2T_5 resulted in maximum number of secondary

Bud initiation duration and number of primary, secondary and tertiary buds as affected by varieties, treatments and their interactions (pooled

Treatment		Days taken for bud initiation	initiation	Number	of primary bu	Number of primary buds at 30 days	Numbe	er of secon	Number of secondary buds	Numb	Number of tertiary buds	ary buds
	>	V_2	Mean	>	V_2	Mean	>	V 2	Mean	>	\mathbf{V}_{2}	Mean
T,	20.06	18.71	19.39	1.79	2.02	1.91	2.30	2.23	2.26	6.01	5.18	5.59
$\mathbf{T}_{2}^{^{\dagger}}$	19.93	17.93	18.93	1.39	1.84	1.62	2.66	2.61	2.63	4.90	4.80	4.85
$\mathbf{T}_{_{\mathbf{J}}}^{_{\mathbf{J}}}$	21.30	20.84	21.07	1.66	2.02	1.84	2. 4	2.49	2.47	6.02	5.65	5.84
$_{_{\scriptscriptstyle{1}}}^{^{\prime}}$ T	19.79	19.69	19.74	1.96	2.24	2.09	3.25	2.56	2.90	6.58	99.5	6.12
T,	16.47	17.09	16.79	2.48	2.70	2.59	3.69	3.08	3.38	29.9	6.75	6.71
T,	16.99	17.93	17.47	2.84	3.16	2.99	2.47	2.57	2.52	6.03	80.9	90.9
Mean	19.09	18.70	ı	2.02	2.33		2.80	2.59	•	6.04	5.69	ı
	Time tak	Time taken for bud initiation	initiation	Number	of primary bu	Number of primary buds at 30 days	Numbe	r of secon	Number of secondary buds	Numbe	Number of tertiary buds	ary buds
	\	Т	VxT	>	Т	VxT	>	Т	VxT	>	Т	VxT
${ m SE}^{\pm}$	0.47	0.27	99.0	80.0	0.05	0.12	0.18	0.10	0.25	0.36	0.21	0.51
$CD_{0.05}$	NS	0.79	1.93	0.24	0.14	0.34	NS	0.29	0.73	SN	0.61	1.48

 T_1 : Sawdust + VAM (30 g), T_2 : Sawdust + Trichoderma viride (30 g), T_3 : Sawdust + VAM (30 g) + T viride (30 g), T_4 : Sawdust + BAP 40 ppm (4 ml) + B substillis (30 g), T_5 : Sawdust + VAM (30 g) + BAP 40 ppm (4 ml) + B substillis (30 g), V_1 : Grand Naine, V_2 : Shrimanti

buds (3.69 and 3.08 respectively). The two varieties did not differ significantly wrt number of tertiary buds. Tertiary buds were found more (6.71) in T_5 followed by 6.12 in T_4 . Among interactions V_1T_1 , V_1T_3 , V_1T_4 , V_1T_5 , V_1T_6 , V_2T_3 , V_2T_4 , V_2T_5 and V_2T_6 resulted in more number of tertiary buds.

Treatments showed good response on primary bud formation per explant suggesting that biofertilizers like VAM, *B subtilis* and plant growth regulator like BAP promoted better auxillary bud regeneration. The role of BAP as a shoot promoting hormone is well known and its activity has been reported by Mundhara and Rashid (2002). Association of *B subtilis* with variety of plants and involvement in promoting plant growth (Cazorla et al 2007) by making nutrients more readily available to plants (Nagorska et al 2007) is also well known.

Significantly the highest number of secondary buds was recorded in Grand Naine ie $V_1(2.80)$ and $T_5(3.69)$ and in the interaction of V_1T_5 ie Grand Naine + sawdust + BAP 40 ppm (4 ml) + B substillis (30 g) (3.38) suggesting that B subtilis in combination with BAP or in combination with IBA increased the regeneration efficiency of secondary bud in cv Grand Naine. Similar results were also reported in wild banana, Musa laterita (Dayarani et al 2013) with coinoculation of B subtilis. Similar results have been recorded under the number of tertiary buds.

Significantly highest number of plantlets per sucker (19.89), (26.30) and (27.49) was recorded in variety Grand Naine V_1 , treatment T_5 and interaction V_1T_5 respectively.

The maximum number of leaves per plant was recorded in variety Grand Naine $V_1(3.81)$, treatment $T_5(4.75)$ and in interaction V_1T_5 (4.80. Maximum plantlet height was recorded in variety

Table 2. Growth parameters of plantlets as influenced by different treatments (pooled means)

Treatment	Number of	Number of plantlets established/sucker	shed/sucker	Numbe	Number of leaves/plant	s/plant	Plantle	Plantlet height (cm)	m)	Number o	Number of secondary roots/plantlet	oots/plantlet
	\mathbf{V}_1	V_2	Mean	\mathbf{V}_{1}	V_2	Mean	N N	V_2	Mean	\mathbf{V}_{1}	V_2	Mean
$T_{_{_{\! \! \! \! \! \! \! \! \! \! \! \! \! \! \! \! \! $	13.47	11.96	12.72	2.56	2.31	2.43	12.71	14.63	13.67	4.34	5.41	4.88
T_{j}^{i}	16.03	14.59	15.31	3.47	3.30	3.39	13.26	14.21	13.74	5.15	4.07	4.61
Ľ	16.94	16.09	16.52	3.56	3.78	3.67	11.67	13.07	12.37	6.22	4.45	5.34
L T	24.36	21.78	23.07	4.36	4.72	4.54	17.31	12.79	15.05	7.35	90.9	6.71
Ţ	27.49	25.11	26.30	4.80	4.69	4.75	16.05	16.70	16.38	8.76	6.94	7.85
Ĺ,	21.02	12.44	16.73	4.08	3.25	3.67	12.02	14.35	13.19	90.6	5.12	7.09
Mean	19.89	16.99	ı	3.81	3.68	1	13.84	14.29	1	6.81	5.34	ı
	Number of	Number of plantlets established/sucker	shed/sucker	Numbe	Number of leaves/plant	s/plant	Plantle	Plantlet height (cm)	m)	Number 6	Number of secondary roots/plantlet	oots/plantlet
	>	T	VxT	>	Т	VxT	>	Т	VxT	>	T	VXT
$\mathrm{SE}\pm$	0.29	0.17	0.41	0.11	90.0	0.16	0.49	0.28	69.0	0.72	0.41	1.01
$CD_{0.05}$	0.85	0.49	1.21	NS	0.19	0.47	1.43	0.82	2.02	NS	1.22	2.97

 T_1 : Sawdust + VAM (30 g), T_2 : Sawdust + $Trichoderma\ viride\ (30\ g)$, T_3 : Sawdust + VAM (30 g) + $T\ viride\ (30\ g)$, T_4 : Sawdust + IBA (dipping in 0.25% solution) + $Azospirillum\ (30\ g)$, Sawdust + BAP 40 ppm (4 ml) + Bacillus substillis (30 g), T₆: Sawdust + VAM (30 g) + BAP 40 ppm (4 ml) + B substillis (30 g); V₁: Grand Naine, V₂: Shrimanti Shrimanti V_2 (14.29 cm), treatment T_5 (16.38 cm) and interaction V_2T_5 (16.70). The maximum number of secondary roots per plant was also recorded in cv Grand Naine V_1 (6.81), treatment T_5 (7.85) and interaction V_1T_6 (9.06).

Sawdust was used as substrate in all the treatments because of its better water holding capacity as also observed by Baiyeri and Aba (2005).

The higher water holding capacity of sawdust could be attributed to its better water retention ability. The physical composition of the growing medium is reported to have a profound effect on the supply of water and air to the growing plant as well as known to affect anchorage, nutrient and water holding capacity of the medium (Baiyeri and Aba 2005).

Thus the macro-propagation along with biofertilizer treatment and growth promoters showed varying effects on the growth parameters during the hardening stage.

CONCLUSION

Macro-propagation offers the cheap alternative with tremendous potential for the production of quality planting material in banana. It is concluded that incorporation of additives like biofertilizers and growth hormones to the sawdust substrate not only enhanced the regeneration of primary, secondary and tertiary buds but also promoted the growth and development of plantlets thereby reducing the post-transplanting shock.

The macro-propagation technique optimized in the present study is user-friendly which requires minimum skill and expertise and is suitable for adoption by the farmers at the farm level. Further the cost per plant production was less in all the treatments tested making it a cost effective technique accessible to the small and marginal farmers without compromising the quality.

REFERENCES

- Baiyeri KP and Aba SC 2005. Response of *Musa* species to macro-propagation. I: Genetic and initiation media effects on number, quality and survival of plantlets at pre-nursery and early nursery stages. African journal of Biotechnology **4(3)**: 223-228.
- Baiyeri KP and Aba SC 2007. A review of protocols for macropropagation in *Musa* species. Fruit, Vegetable and Cereal Science and Biotechnology **1(2)**: 110-115.
- Cazorla FM, Romero D, Perez-Garcia A, Lugtenberg BJJ, de Vicente A and Bloemberg G 2007. Isolation and characterization of antagonistic *Bacillus subtilis* strains from the avocado rhizoplane displaying biocontrol activity. Journal of Applied Microbiology **103(5)**: 1950-1959.
- Dayarani M, Dhanarajan MS, Uma S and Durai P 2013. Macro-propagation for regeneration of wild bananas (*Musa* spp). Advanced BioTech **12(12)**: 16-18.
- Mundhara R and Rashid A 2002. Stimulation of shoot-bud regeneration on hypocotyl of *Linum* seedlings on a transient withdrawl of calcium: effect of calcium, cytokinin and thidiazuron. Plant Science **162(2)**: 211-214.

- Nagorska K, Bikowski M and Obuchowski M 2007. Multicellular behaviour and production of a wide variety of toxic substances support usage of *Bacillus subtilis* as a powerful biocontrol agent. Acta Biochimica Polonica **54(3):** 495-508.
- Sajith KP, Uma S, Saraswathi MS, Backiyarani S and Durai P 2014. Macro-propagation of banana: effect of biofertilizers and plant hormones. Indian Journal of Horticulture 71(3): 299-305.
- Swennen R 1990. Plantain cultivation under West African conditions: a reference manual. International Institute of Tropical Agriculture, Ibadan, Nigeria, 24p.
- Uma S, Sajith KP, Saraswathi MS and Durai P 2010.
 Macropropagation: a farmers' friendly technology.
 Technical Bulletin #18, National Research Centre for Banana, Trichy, Tamil Nadu, India, 17p.
- Vuylsteke DR 1998. Shoot-tip culture for the propagation, conservation and distribution of *Musa* germplasm. International Institute of Tropical Agriculture, Ibadan, Nigeria, 82p.