Effects of cytokinins and auxins on micropropagation of *Musa* spp. cv. Yangambi

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Abstract— *The present study was conducted at Banana Tissue Culture lab of Regional Plant Resource Centre, Bhubaneswar, Odisha to obtain a standardized micropropagation protocol supplemented with different concentrations and combinations of cytokinins and auxins for Musa cv. Yangambi Km-5 (AAA) variety. Data collected for in vitro culture consists of the following parameters: days for bud initiation, rate of shoot proliferation (%), growth value (gm) and number of multiple shoot buds during multiplication stage. Remarkable effects of Cytokinins and Auxins were observed in Yangambi Km-5. Out of various treatments, best concentration for multiple shoot in short period of time for Yangambi Km-5 was found in mediums 3 mg/l BAP + 0.5 mg/l IAA + 0.25 mg/l NAA + 100 mg/l ADS and 1.5 mg/l BAP + 1.5 mg/l KN + 1 mg/l IAA + 0.25 mg/l NAA + 100 mg/l ADS. Maximum growth value, rate of proliferation and number of shoot buds was obtained from explants culture medium 3 mg/l BAP + 0.5 mg/l IAA + 0.25 mg/l NAA + 100 mg/l ADS.*

Keywords— Micropropagation, growth regulators, Yangambi Km-5, Musa spp.

I. INTRODUCTION

Banana is an important and widely grown fruit crop in the tropical and subtropical regions of the world (Darvari *et al*., 2010; Rahman *et al*., 2013).Banana represents the world's second largest fruit crop and fourth most important global food crop with an annual production over 100 million metric tons (FAOSTAT, 2010).Banana plants are usually propagated by vegetative means by using suckers which grow from lateral buds originating from corms, and suckers are used for production of individual plants. This process is very slow as the rate of multiplication of suckers through conventional vegetative means has been found to express several negative impacts which include transmission of diseases, low production and poor preservation of original plant genetic material (Hussein, 2012). The non-professional cultivation practices, pest epidemic and viral diseases affect the yield and quality of banana crop (Wambugu *et al.*,2008).The problem of emerging diseases can be solved by propagating banana through tissue culture (Ali *et al.*,2011). Success in *in vitro* multiplication is based on the growth and differentiation of plant tissues, which is viable only by the addition of suitable growth regulators (Gaspar *et al*., 2003).

Musa variety Yangambi is a vigorous plant that remains productive on poor soils and which has become well known for its thick peel being resistant to black leaf streak disease, caused by *Mycosphaerella fijiensis*, which ultimately increases the self-life of this banana cultivar.The goals of this study are (i) to find out the best plant growth regulators for shoot proliferation and multiplication of banana explants of *Musa paradisiaca* cv. Yangambi Km-5 cultured on Murashige and Skoog's medium containing different concentration of growth regulators (Cytokinins and Auxins).

II. MATERIAL AND METHODS

2.1 Plant materials

The plant material (Suckers) for the present experiment was collected from the Yangambi mother block of RPRC, Bhubaneswar during the month of January 2016.Suckers were washed thoroughly under running tap water for 10-15 min. The suckers were then chopped off about 5-6 cm in length and 3-4 cm in diameter.

2.2 Sucker sterilization

For surface sterilization several steps were carried out. After processing suckers were washed in liquid detergent (Labolene) for 2-3minutes.Explants were then dipped in bavistin solution (1 %) for 30 minutes. After 30 minutes the suckers were washed with autoclaved double distilled water and transferred to Calcium hypochlorite (30 %) for 30 minutes. The suckers were washed in 70 % alcohol solution for 1 minute. Finally the suckers were washed 3- 4 times with autoclaved double distilled water to remove excess chemicals from the sucker surface.

2.3 Preparation of culture medium

The medium used for banana tissue culture was Murashige & Skoog Medium (MS) (Murashige and Skoog, 1962). The pH of the medium was adjusted 5.75 to 5.8 with 0.1N NaOH or 0.1N HCl. To one liter of medium 5.0gms of agar (Plant tissue Culture grade, Hi-Media, India) was added. All the media were autoclaved at 15 psi and 121°C for 20 minutes. The autoclaved molten media were then dispensed into sterilized culture vessel inside a laminar air flow cabinet. 6-Benzyl Amino Purine (**BAP**), Kinetin (**KN**), Indole -3-acetic acid (**IAA**), Naphathalene acetic acid (**NAA**) and Adenine Sulphate (**ADS**) hormones were used in the present experiment.

TABLE 1
AMOUNT OF CYTOKININS AND AUXINS USED IN INDUCTION MEDIUM AND MULTIPLICATION MEDIUM ALONG WITH M.S MEDIUM FOR IN VITRO APICAL CULTURE.

Sl. No.	Medium code	Cytokinins and Auxins (Mg L^{-1})				
		BAP	KN	IAA	NAA	ADS
I	a	2.0	-	0.25	0.25	100
II	b	2.0	-	0.75	0.25	100
III	c	3.0	-	0.50	0.25	100
IV	d	4.0	-	0.50	0.25	100
V	e	6.0	-	1.00	0.25	100
VI	f	1.5	1.5	1.00	0.25	100

III. RESULT AND DISCUSSION

In the present work, effects of different concentrations of BAP (1.5, 2.0, 3.0, 4.0 and 6.0 mg L^{-1}), Kinetin (1.5 mg L^{-1}) with IAA (0.25, 0.50, 0.75 and 1.00 mg L^{-1}) and NAA (0.25 mg L^{-1}) were studies for optimizing the protocol for effective multiple shoot proliferation of the exotic *Musa*YangambiKm-5.

TABLE 2
EFFECTS OF CYTOKININS AND AUXINS

Sl. No.	Medium code	No. of explants/ medium	Days for buds initiation	Rate of proliferation (%)	Shoot buds per explant	Initial fresh Wt. (gm)	Final fresh Wt. (gm)	Growth value (gm)
I	a	10	14 ±1.11	60	7 ±1.00	1.4 ± 0.05	2.3 ± 0.04	0.64
II	b	10	15 ±0.97	50	6 ±0.86	1.5 ± 0.06	2.5 ± 0.06	0.67
III	c	10	12 ±0.86	90	11 ±0.92	1.3 ± 0.06	2.8 ± 0.06	1.15
IV	d	10	14 ±0.67	70	8 ±0.70	1.4 ± 0.06	2.5 ± 0.07	0.78
V	e	10	13 ±0.78	60	8 ±0.78	1.4 ± 0.05	2.4 ± 0.05	0.71
VI	f	10	13 ±0.70	80	9 ±0.78	1.4 ± 0.06	26 ± 0.06	0.85

From the above data it was observed that the explants cultured on medium ‘c’ had high rate of proliferation (90 %) and growth value (1.15 gm). It was also found out that the explants cultured on medium ‘c’ had maximum number of shoot buds (11) with short days for proliferation (12). Nearly similar effects were observed in explants culture on medium ‘f’ having 9 numbers of shoot buds with 13 days for proliferation. But the rate of proliferation of explants was low in medium ‘f’ in comparison to medium ‘c’. Benzylaminopurine (BAP) combined with auxins (indole acetic acid and naphthalene acetic acid) exhibit synergistic effect and hence has also been used by number of researchers (Al-Amin *et al*., 2009; Sipen and Davey, 2012; Ngomuo *et al*., 2013).Other researchers reported nearly similar effect of BAP and IAA on shoot length (Rahaman *et al*., 2013; Ahmed *et al*., 2014).The results agree with the findings of Muhammad *et al.* (2007).Habiba *et al.* (2002)and Ahmed *et al.* (2014), also reported synergistic effect of BAP and IAA at nearly similar combination of 4.0 mg L^{-1} BAP and2.0 mg L^{-1} IAA.

FIGURE 1- A: EXPLANT CULTURED ON MEDIUM 'C', B: EXPLANT CULTURED ON MEDIUM 'F'.

IV. CONCLUSION

From the data obtained from different experiments conducted in the present study it was concluded that cytokinins (BAP and KN) and auxins (IAA and NAA) had various effect on explants growth and shoot proliferation when used at different concentration. BAP was found effective for culture of Yangambi Km-5 in combination with IAA, NAA and ADS. Maximum growth value, rate of proliferation and number of shoot buds was obtained from explants culture medium 3 mg/l BAP + 0.5 mg/l IAA + 0.25 mg/l NAA + 100 mg/l ADS. Nearly similar effects was observed in explants culture on medium 1.5 mg/l BAP + 1.5 mg/l KN + 1 mg/l IAA + 0.25 mg/l NAA + 100 mg/l ADS.

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