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EFFECT OF SUCROSE ON OPTIMAL ROOTING OF *MUSA* SPP. PLANTLETS IN-VITRO CONDITION

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ABSTRACT

To evaluate the effect of various concentrations of sucrose on *in vitro* rooting of regenerated banana micro shoots. Three selected *Musa* varieties GCTCV-215 (AAA), Yangambi (AAA) and FHIA-23 (AAAA) were cultured in half strength MS media supplemented with root promoting growth regulator 1.0 mg/L^{-1} indole butyric acid (IBA) and different concentrations of sucrose (0, 20, 40, 60 and 80 g/L^{-1}). Significant differences ($p \leq 0.05$) were noticed in all *Musa* clones. Results indicated that as compared to control out of different concentrations of sucrose all varieties, showed the maximum number of roots as well as highest root length in MS medium augmented with 40 % and 20 % g/L sucrose respectively. Appropriate result among the varieties, in terms of showing maximum roots was observed in GCTCV-215, followed by Yangambi. Effectual rooting on *in vitro* is helpful for the successful establishment of these *Musa* varieties in field condition.

Key words: *In vitro* rooting, sucrose, *Musa* genomes (AAA, AAAA)

INTRODUCTION

Monocotyledon family Musaceae represents genera *Musa* and *Ensete*. Bananas are the main species of *Musa*. In the least developed countries, people depend on it for nutritional, medicinal and energy purpose (Darvari *et al*., 2010; Anbazhagan *et al*., 2014). In recent time steady supply of banana crop is the principal requirement. At present time Pakistan is facing major down fall in banana production due to monoculturing of single cultivar Basrai so acclimatization of new genetic architecture capable with diseases resistance is the demand of time. Through tissue cultural techniques adequate supply of banana is achieved and number of researchers work on them (Ikram-ul- Haq and Dahot, 2007; Muhammad *et al*., 2007; Al-Amin *et al*., 2009; Iqbal *et al*.,2013). For banana improvement micropropagation has been extensively used tissue culture technique, which provides large population of healthy and genetically homogeneous planting material (Gubbuk and Pekmezcu, 2004; Jafari *et al*., 2011; Ahmed *et al*., 2014). Successful acclimatization of micropropagated plants in the field is based on the strong and vigorous rooting (Ahmed *et al*., 2014) so it is essential to emphasize on major nutrition required for the efficient root growth and development. Optimal growth rate of *in vitro* rooting generally based on different aspect related with the presence of exogenous hormone, carbon source and its concentration in the medium (Kitto and Young, 1981; Fuentes *et al*., 2000; Mello *et al*., 2001; Placide *et al.*, 2012). Initial growth of root primordia also required carbohydrates as structural material. (Yoo *et al*., 1996).In plant cell carbon source serve as sole energy, work as osmotic agent and shield the plant cell against stresses (Lipavska and Konradova 2004). Many researchers previously reported the use of different carbohydrate sources along with Indole butyric acid and Naphthalene acetic acid for *in vitro* rooting of *Musa* plantlets (Iqbal *et al*, 2013; Morfeine 2014; Waman *et al.,* 2014; Ahmed *et al.,* 2014). Frequently used auxin, Indole butyric acid (IBA) at 1.0 mg/ L^{-1} was accounted by number of researchers as most suitable for rooting (Roy *et al*, 2010; Anbazhagan *et al,* 2014; Mahdi *et al*., 2014). Besides their role as primary transportable molecule and it's easily uptake across cell membrane (Morfeine, 2014), sucrose is highly soluble in water without interfere other most of the biochemical mechanism (Smith, 1995). Therefore the present work was carried out to assess the consequence of sucrose in different concentration on *in vitro* rooting of *Musa* spp. GCTCV-215, Yangambi and FHIA-23.

MATERIAS AND METHODS

Suckers of three *Musa* varieties 'Yangambi' (AAA), GCTCV-215 (AAA) and FHIA-23 (AAAA) were used as experimental material available in NIA, Tando Jam.

Sterilization and culturing media: Suckers of selected *Musa* varieties were collected from healthy banana plants. After removing extra tissues, suckers

were dipped in 70% alcohol for one minute and then in 10% sodium hypochlorite (NaClO) solution for twenty (20) minutes. Then suckers were thoroughly washed with sterile distilled water to remove the residue of sodium hypochlorite. After that explants were trimmed to a size of 7-8mm approximately from the base and shoot apex and then cultured on MS full strength basal medium with 4.0mg/L^{-1} BAP and 0.5mg/L^{-1} IAA for regeneration and formation of *in vitro* micro shoots.

Rooting media: For rooting, *in vitro* micro shootlets of 3-6cm in length were separated from shoots cluster and then transferred to half strength MS basal medium solidified with gelrite and supplemented with different concentrations of sucrose along with 1.0 mg/L^{-1} indole butyric acid (IBA) (Table 1). For growth and development of root, cultures were maintained at 25 °C ± 2 °C under 16/8 hour photoperiod. Three replications were used for each treatment and experiment was designed as Completely Randomized (CRD). Date was recorded after one month of culturing including parameters, root number and root length.

Table-1: $MS^{1/2}$ strength rooting media with different concentrations of sucrose and 1.0mg/ L^{-1} IBA

(IBA mg/ L^{-1} + Sucrose g / L^{-1})		
1.0	+	20
1.0	+	40
1.0	+	60
1.0	+	80

Statistical analysis: Mean values per treatments based on the average of three replications were statistically analyzed by ANOVA using statistical software STATISTIX (8.1version).

RESULTS AND DISCUSSION

Effect of different concentrations of sucrose on average root per shoot: Effect of different concentrations of sucrose along with 1.0 mg/L^{-1} IBA was observed on the average roots per shoot. Analysis of variance (ANOVA) for average roots was shown significant ($p \leq 0.05$) differences (Table 4). Data for average roots per shoot is presented in Table 2. As compared to control all varieties, showed the highest number of roots in MS medium augmented with 40 g/L^{-1}sucrose followed by 20 g/L^{-1} sucrose. Whereas reduction in root number was observed at 60 and 80 g/L^{-1} sucrose. According to analyzed data, it was noticed that in all applied concentrations, the number of roots decreased with increased concentration of sucrose. At low quantity of sucrose almost all varieties showed hopeful response and produced maximum rooting. Effect of sucrose on rooting was reported by Ahmed *et al.,* (2014). They found best rooting in media containing 30 g/L^{-1}sucrose. In another work, more or less similar quantity 30 and 45 g/L^{-1}sucrose gave significant response on rooting (Waman *et al*., 2014; Morfeine, 2014). In term of maximum roots, among the varieties best result were observed in GCTCV-215 and Yangambi.

Table-2: Effect of MS½ Strength media with different concentrations of Sucrose along with 1.0 mg/ L^{-1} IBA on Mean root number

Genotypes	Shoot Per Treatment	Sucrose g/ L^{-1}					Mean
		Control	20	40	60	80	
GCTCV-215	10	6.00 h	8.00 cd	9.86 a	7.63 de	6.70 g	**7.64 a**
Km-5	10	4.56 ij	7.33 ef	8.73 b	6.83 fg	5.10 i	**6.51 b**
FHIA-23	10	3.13 k	7.23 e-g	8.30 bc	4.66 i	4.00 j	**5.46 c**
Mean		**4.56 e**	**7.52 b**	**8.96 a**	**6.37 c**	**5.26 d**	

Values with dissimilar letter(s) in a column are significantly different at $P \leq 0.05$.

Effect of different concentrations of sucrose on root length: Data for length of roots was found significant $p \leq 0.05$ (Table 4). As contrasted to control, all the varieties showed highest root length in concentration 40 g/L^{-1}sucrose (Table 3). Result indicated that there was decline in root length observed with higher concentration of sucrose. The highest root length was noticed in Yangambi (13.6 cm) than that of rest of the varieties. Overall results suggested that root length declines above 40 g/L^{-1} sucrose. According to present results, in all treatments concentration 40g/L^{-1}sucrose along with 1.0 mg/L^{-1} IBA was recommended as most suitable for selected *Musa* varieties in term of producing maximum roots length. In other finding researcher observed maximum root length at 30 g/L^{-1}sucrose (Hussein 2012).

Table-3: Effect of MS½ Strength media with different concentrations of Sucrose along with 1.0 mg/ L^{-1} IBA on root length (cm)

Genotypes	Shoot Per Treatment	Sugar g/ L^{-1}					Mean
		Control	20	40	60	80	
GCTCV-215	10	4.0 i	7.0 e	12.6 b	6.0 fg	5.0 h	**6.94 b**
Km-5	10	5.1 h	8.1 d	13.6 a	6.1 f	5.5 gh	**7.73 a**
FHIA-23	10	3.0 j	5.8 fg	9.6 c	4.0 i	3.8 i	**5.25 c**
Mean		**4.0 e**	**6.9 b**	**11.9 a**	**5.3 c**	**4.8 d**	

Values with dissimilar letter(s) in a column are significantly different at $P \leq 0.05$.

Table-4: Analysis Of Variance (ANOVA) for *in vitro* rooting

Source of variation	Mean square (MS) of shoot parameters		
	DF	Root number	Root length (cm)
Replications	2	0.2060	0.3176
Varieties	2	17.7207*	24.0916*
Treatments	4	27.8892*	90.5344*
V x Treat	8	0.8812*	0.8854*
Error	28	0.1424	0.1133
Total	44	--	--

The results are for the mean of 3 replicate (Significant at $P \leq 0.05$)

CONCLUSSION

Result of this work showed that the combined effect of 40 gm/L^{-1}sucrose with 1.0 mg/L^{-1} IBA were played positive role in term of producing sufficient root with significant length. All varieties perform well on different level of sucrose and gave their best at moderate concentration. This work helps to established suitable media for sufficient rooting of *Musa* plantlets during *in vitro* condition which may increase their survival potential in *ex vitro* condition.

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