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RESEARCH PAPER

In vitro technology for propagation of pineapple (*Ananas comosus*) cv. KEW

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Abstract : Pineapple fruit cultivation, production and multiplication is entirely dependent upon vegetative means *i.e.* crowns, suckers and slips. The availability of suckers, slips and crowns on large scale as planting material and poor suckering habit of the commercial varieties like 'Kew' are the major problems in the pineapple planting and production. Micro-propagation studies in Kew variety of pineapple were attempted to release standard protocol for *in vitro* clonal multiplication. Slips apical section used as explants. Surface sterilization with 0.1 per cent $HgCl_2$ for 8 minutes, followed by 0.2 per cent ridomil 15 minutes treatment for reducing contamination of cultures. MS medium was found better when supplemented with 1.5 ppm BAP + 0.5 ppm NAA and 2.0 ppm BAP + 0.25 ppm NAA resulted in 80.53 per cent shoot establishment, and 7.10 numbers of shoots induction occurred. MS medium with 2 ppm IBA and MS medium with 1.5 ppm IBA + 0.5 ppm NAA resulted in 92.66 per cent rooting and 5.61 numbers of roots induction occurred.

Key Words : *Ananas comosus*, Micropropagation, Growth media, Growth regulators, Culture establishment, Rooting

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INTRODUCTION

Pineapple is considered as an exotic desert, tropical fruit due to its attractive appearance, excellent flavor, fragrance and exquisite taste qualities. According to Bose *et al.* (1999) it symbolizes blamy tropical lands and lisurly life tropical areas. Pineapple is one of the choicest fruit throughout the world. It is good source of carotene (Vitamin A) and ascorbic acid (Vitamin C) and is fairly rich in vitamin B_1 and B_2. It also contains phosphorus and mineral like Ca, Mg, K, and Fe. Besides, it is also fairly good source of bromelin, a digestive enzyme. It provides adequate roughages to prevent constipation. Its fresh juice has a cooling and refreshing effect and especially in summer season it acts as an appetizer. The need of large scale planting material for commercial or industrial use is difficult to achieve by conventional techniques, particularly when uniform planting material is needed. Hence, alternative technologies for production of uniform planting material in large scale are required to be investigated.

One of the technologies that could be used for this purpose is the *in vitro* technique (Ika and Ika, 2003). It involves the combination of growth media and growth regulators for culture establishment, shoot multiplication and rooting of cultures. The micropropagation, an advanced avenue of biotechnological intervention has opened up opportunities to overcome these problems. The micro-propagation has many advantages over conventional vegetative method of pineapple propagation.

MATERIAL AND METHODS

Plant material:

The experimental material used for the study consisted of cv. Kew of pineapple and slip was used as explants in experiment.

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Aseptic technique:

Slips were washed with running water. The pretreatment of bavistin (2.0 g/litre solution) for 4-5 hours was given and then used as the source of explants throughout the investigation. Dissected explant were treated with 0.1 per cent $HgCl_2$ for 8 minuts before inoculation of cultures.

Culture media for shooting:

To optimize the culture media for shoot regeneration and multiplication, an experiment was conducted with emerging vegetative buds on two media with four levels of 6-BAP and four levels of 6-BAP and NAA combinations. Subsequently, the established cultures were sub cultured on fresh media at regular interval of 28 to 30 days.

Treats.	Medium	6-BAP (ppm)	NAA (ppm)
$T_{1=}$	MS medium +	0.5 ppm+	-
$T_{2=}$	MS medium +	1.5 ppm+	-
$T_{3=}$	MS medium+	1.5 ppm+	0.5 ppm
$T_{4=}$	MS medium+	2.0 ppm+	0.25 ppm
$T_{5=}$	½ MS medium	+0.5 ppm+	-
$T_{6=}$	½ MS medium	+1.5 ppm+	-
$T_{7=}$	½ MS medium	+0.5 ppm+	0.1ppm
$T_{8=}$	½ MS medium	+1.0 ppm+	0.1ppm

Culture media for rooting:

An experiment was conducted, consisting of two media with four levels of IBA and four levels of IBA and NAA combinations. Subsequently, shoot cultures were sub cultured on fresh medium at interval of 28 to 30 days.

Treats.	Medium	IBA (ppm)	NAA (ppm)
T_1=	MS media +	1.0 ppm+	-
T_2=	MS media +	1.5 ppm+	-
T_3=	MS media +	2.0 ppm+	-
T_4=	MS media+	1.5 ppm+	0.5 ppm
T_5=	½ MS media	+1.5 ppm+	-
T_6=	½ MS media	+1.0 ppm+	-
T_7=	½ MS media	+0.5 ppm+	-
T_8=	½ MS media	+1.0 ppm+	0.5 ppm

The experiments were carried out using Completely Randomized Design (CRD). The data were statistically analyzed according to Panse and Sukhatme (1995).

RESULTS AND DISCUSSION

The treatment MS medium with 1.5 ppm 6-BAP + 0.5 ppm NAA (80.53 %) was found to be better for the per cent shoot establishment. The results revealed that as regarding average number of shoots, the treatment MS medium with 2.0 ppm BAP + 0.25 ppm NAA was significantly better over rest treatments. The treatment MS medium with 0.5 ppm BAP (41.0 days) was found to be significantly superior over the rest of treatments in case of average number of minimum days required for shooting of cultures (Table 1).

Table 1 : Effect of combinations of media and growth regulators on per cent shoot establishment, average no. of shoots and average no. of days

Treatments	Per cent shoot establishment (after 3-4 week)	Average no. of shoots (after 3-4 week)	Average no. days
T_1	62.31(52.14)	1.46	47.36
T_2	71.06(57.43)	2.10	75.00
T_3	80.53(63.83)	3.20	80.33
T_4	73.52(59.03)	7.10	85.00
T_5	41.59(40.16)	2.40	41.00
T_6	62.00(51.94)	1.95	44.83
T_7	52.13(46.22)	3.55	65.36
T_8	42.82(40.87)	2.22	71.00
Mean	60.74	2.99	63.73
S.E. ±	0.73	0.31	0.26
C.D. (P=0.01)	3.04	1.31	2.60
F-test	Significant	Significant	Significant

Values in parenthesis indicate arc-sine values

On an average MS full and MS half strength medium supplemented with 0.5 ppm to 2.0 ppm 6-BAP with or without 0.1- 0.5 ppm NAA, the media combination was best for the shoot regeneration and multiplication of pineapple. Akbar *et al*. (2003) found that MS medium containing 1.5 ppm 6-BAP + 0.5 ppm NAA promoted tiny multiple shoots in pineapple and also reported the minimum 40 to 90 days required for shooting when explant cultured on ½ MS medium with 1.5 ppm 6-BAP. Khan *et al*. (2004) suggested ½ MS medium for *in vitro* clonal multiplication with 1.0 ppm to 1.5 ppm 6-BAP + 0.001 to 0.25 ppm NAA reported average

Table 2 : Effect of combinations of media and growth regulators on per cent root initiation, average no. of roots and average no. of days required for rooting.

Treatments	Per cent root initiation (after 3-4 week)	Average no. of root (after 3-4 week)	Average no. days
T_1	31.16(33.93)	4.5	8.00
T_2	90.00(71.56)	3.95	19.16
T_3	92.66(74.88)	3.66	12.06
T_4	50.00(45.00)	5.61	12.50
T_5	41.29(43.55)	2.06	14.00
T_6	0(0)	0	0
T_7	0(0)	0	0
T_8	26.80(20.33)	2.5	17.56
Mean	41.45	2.78	10.41
S.E. ±	1.14	0.28	0.25
C.D. (P=0.01)	4.73	1.16	1.04
F-test	Significant	Significant	Significant

Values in parenthesis indicate arc-sine values

of one to three shoots. Almeida *et al*. (2002) and Chandra (2000) reported micropropagation studies on MS medium in pineapple on other hand a significant decrease in shoot no. per explant was observed at lower that is less than 1.0 ppm 6-BAP. They reported an average 1.0 to 7.0 shoots on MS medium containing 2.0 ppm of 6-BAP with 0.5 ppm NAA.

Significantly, the highest per cent of root initiation was observed in MS medium with 2.0 ppm IBA (92.66 %) which was significant with rest treatments (Table 2). The highest average numbers of roots were formed on MS medium with 1.5 ppm IBA + 0.5 ppm NAA (5.61 average number of roots). Minimum average number of days required for rooting was observed on MS medium with 1.0 ppm IBA (8.0 average numbers of days).

According to, Akbar *et al*. (2003) MS medium containing 2.0 to 2.5 ppm IBA produced 4 to 5 roots within 8 to 12 days. Khan *et al*. (2004) reported MS medium containing 1.5 to 2.0 ppm IBA with or without NAA produced 4.0 roots within 7 to 8 days.

Conclusion:

From the present investigation, it can be concluded that, micropropagation offers simple and reliable method of the multiplication of pineapple. The pineapple plants produced through tissue culture are true to the type, uniform in growth, fruiting, quality and free from diseases. Murashige and Skoog medium showed the highest percentage of established cultures. The concentration of 2.0 mg/l BAP + 0.25 mg/l NAA has produced maximum number of shoots. 1.0 mg/l IBA induced early rooting.

REFERENCES

Akbar, A.M., Karmarkar, B.K. and Roy, S.K. (2003). Callus induction and high frequency plant regeneration of pineapple [*Ananas comosus* (L.) Merr.]. *Plant Tissue Cult.,* **13** (2): 109-116.

Almeida De, W.A.B., Santana, G. S., Rodrigues, A.P.M. and De Carvalho Costa, M.A.P. (2002). Optimization of a protocol for the micropropagation of pineapple. *Rev. Bras. Frutic, Jaboticabal sp*., **24** (2) : 296-300.

Chandra, R. (2000). Micropropagation of tropical and semitropical fruit crops. In: *Biotechnology of horticultural crops*. (Ed. Parthasarthy.) Naya Prakashan; Kolkata Publication; **3**: 127-128.

Ika, R.T. and Ika, M. (2003). *In vitro* culture of pineapple by organogenesis and somatic embryogenesis: Its utilization and Prospect. *Bull. Agro Bio*., **6** (1): 34-40.

Khan, S.V., Nasib, A. and Saeed, B.A. (2004). Employment of *in vitro* technology for large-scale multiplication of pineapple (*Ananas comosus*. Merr). *Pak. J. Bot.,* **36** (3): 611-615.

Panse, V.G. and Sukhatme, P.V. (1995). *Statistical methods for agricultural workers*. ICAR, New Delhi (INDIA).