Madras Agric. J., 98 (4-6): 118-120, June 2011
https://doi.org/10.29321/MAJ.10.100256

Short Note

In *vitro* Propagation of *Anthurium andreanum* cv. Temptation

R. Chitra*, M. Ganga, R. Arulmozhiyan and M. Jawaharlal
Department of Floriculture and Landscaping
Horticulture College & Research Institute
Tamil Nadu Agricultural University, Coimbatore - 641 003

Seeds of *Anthurium andreanum* cv. Temptation collected from the flower spadices were germinated on MS medium supplemented with 1.0 mg l^{-1} BAP. After two weeks, 100 % of the seeds germinated. Four weeks later, micro-cuttings from the *in vitro* germinated seedlings were subcultured on Murashige and Skoog's medium containing 0.5 mg l^{-1} BAP and 1.0 mg l^{-1} GA_3. On an average, 5.9 shoots per explant were obtained. The highest number of roots per shoot and the longest roots were obtained on media containing Nitsch's basal salts with 1.0 mgl^{-1} IBA. *Anthurium andreanum* plants regenerated by organogenesis were transferred to pots and 80 per cent of the plants acclimatized successfully.

Key words: Anthurium, seed, micropropagation, *in vitro*.

Anthurium popularly known as the 'tail flower' or the 'flamingo flower' is a tropical plant cultivated for its colourful spathe and attractive foliage. Anthuriums have been cultivated for many decades for their attractive cut flowers. Anthurium trade is next only to orchids among the tropical flowers. Currently, numerous cultivars with different flower sizes, shapes, colours and some with delicate fragrances are available for the consumer.

The *Anthurium* genus comprises about 1500 tropical species which are important ornamental plants and are normally propagated by seed (Dufour and Guerin, 2003). The traditional techniques of vegetative propagation such as the use of stem cuttings and suckers exist; they are tedious and not practical when carried out on a large scale. Vegetative propagation methods applied to these plants have not shown good results and tissue culture techniques appear as an alternative to increase the production (Pierik *et al.,* 1974, Chen *et al*. 1997). Tissue culture can also provide a source of clean material which has become increasingly important due to outbreak of bacterial and other diseases. Anthuriums are highly amenable for in vitro propagation using different parts as explants. Plant regeneration of *Anthurium andreanum* has been achieved through adventitious shoot formation from callus (Pierik *et al.* 1974; Pierik and Steegmans, 1976) and direct shoot regeneration from lamina explants (Martin *et al.* 2003). The production of *in vitro* plants directly from proliferating axillary buds (Kunisaki, 1980), adventitious buds (Cen *et al*., 1993), leaf or petiole organogenic callus culture (Kuehnle and Sugii, 1991) and from somatic embryos derived from in vitro grown leaf blade explants (Kuehnle *et al.,* 1992) has been reported. All workers found that there was great variation in the requirements of different genotypes. One such protocol for large-scale micropropagation of *Anthurium andreanum* is reported here.

Materials and Methods

Explants were obtained from plants of *A.andreanum* cv. Temptation germinated from seeds. The fruits were separated from spadices and sterilized for 15 minutes in 0.1 % mercuric chloride and then rinsed three times with sterile water for 10 minutes. Eighty seeds were collected and sterilized for 5 minutes in 0.1% mercuric chloride, and afterwards they were washed two times with sterile water for 10 minutes. After washing 4-5 times with sterile distilled water, the seeds were inoculated on basal medium consisting of Murashige and Skoog's (1962) salts and vitamins, 3% sucrose and 0.2% Gelrite. The media was buffered to a pH of 5.8 and dispensed in culture bottles before autoclaving at 121°C for 15 minutes. Basal MS medium was supplemented with various concentrations of BAP (0.5, 1.0, 1.5, 2.0 and 2.5 mg l^{-1}) for seed germination. One week later the radicle emerged and shoot developed under continuous light. After four weeks, micro-cuttings were subcultured on MS medium supplemented with BAP (0.5, 1.0, 1.5, 2.0 and 2.5 mg l^{-1}) and GA_3 (1.0 mg l^{-1}) and incubated under continuous fluorescent light (50 $\mu EM^{-2}S^{-1}$) at 25ºC. This process of subculturing was continued repeatedly every 30 days for three cycles. After three months, the micro-shoots were transferred to rooting medium consisting of Nitsch's and Nitsch's (1969) medium containing various concentration of IBA (0.25, 0.50, 0.75, 1.00 and 1.25

*Corresponding author email: chitra.varadharaj@gmail.com

Table 1. Effect of BAP on in vitro seed germination of *Anthurium andreanum* cv.Temptation

Culture Medium	Survival of explants (%)	Microbial contamination of explants (%)	Germination (%)
MS + BAP (0.5 mg l^{-1})	67	33	70
MS + BAP (1.0 mg l^{-1})	95	05	100
MS + BAP (1.5 mg l^{-1})	85	15	80
MS + BAP (2.0 mg l^{-1})	73	27	65
MS + BAP (2.5 mg l^{-1})	74	26	60

mg l^{-1}). After three months, the seven months old rooted plantlets were removed from the culture bottles. The roots were washed in tap water and the plants were transferred to pots containing a mixture of Cocopeat and Perlite (1:1). The plants were kept in hardening chambers with high relative humidity (70-80%) and low light intensity. One month later, when the plants had acclimatized well, they were transferred to the greenhouse.

Results and Discussion

Among the different concentrations of BAP used for *in vitro* seed germination, best response of radicle emergence was obtained on MS + 1.0 mg l^{-1} BAP (Table 1). Puchooa and Sookun (2003) had also reported that BAP is of essential significance for the development of shoots in Anthurium. However, high level of BAP (2.5 mg l^{-1}) suppressed the growth. Teresa Vargas *et al.*, (2004) had established an efficient regeneration system for *Anthurium andreanum* cv. Rubrun, seeds from *in vitro* seed germination on a medium supplemented with 2.2 mM BAP.

Table 2. Effect of BAP and GA_3 on multiple shoot induction from seed explants of *Anthurium andreanum* Cv.Temptation

S.No.	Culture Medium	Number of shoots per explant	Shoot length (cm)	Days for shoot emergence
1.	MS + BAP (0.5 mg l-1) + GA3 (1.0 mgl-1)	3.94	1.02	31.80
2.	MS + BAP (1.0 mg l-1) + GA3 (1.0 mgl-1)	5.94	1.56	29.55
3.	MS + BAP (1.5 mg l-1) + GA3 (1.0 mgl-1)	4.05	1.13	33.11
4.	MS + BAP (2.0 mg l-1) + GA3 (1.0 mgl-1)	3.38	1.08	34.44
5.	MS + BAP (2.5 mg l-1) + GA3 (1.0 mgl-1)	2.56	1.00	33.15
	SE	0.318	0.089	1.07
	CD (P=0.05)	0.709	0.200	2.28

Formation of microshoot was noticed after 4 weeks of inoculation. A two-fold increase in multiplication was observed in 8 to 10 weeks. A similar observation was made earlier by George and Ravishankar (1996) in *Vanilla planifolia*. Further, transfer to the MS media supplemented with BAP (1.0 mgl^{-1}) and GA_3 (1.0 mg l^{-1}) resulted in a two-fold proliferation of shoots at every subculture (Table 2).

The elongated shoots (4 to 5 cm long) were excised and cultured separately in NN medium containing various concentration of IBA to induce roots. The highest number of roots per shoot and the longest roots were obtained on the media containing NN+1.0 mgl^{-1} IBA (Table 3). Earlier, Somaya *et al.*(1998) observed high rooting percentage of *Anthurium andreanum* cultured in NN medium supplemented with IBA (1.0 mg l^{-1}).

From the present study, an effective plant multiplication protocol has been standardized which can be exploited on the commercial scale.

Table 3. Effect of IBA on rhizogenesis of *Anthurium andreanum* Cv. Temptation

S.No.	Culture medium	Percentage response to rhizogenesis	No. of roots per plant	Days taken for rooting	Length of roots (cm)
1.	NN + IBA (0.25 mg l-1)	50.64	95	5.0	3.9
2.	NN + IBA (0.50mg l-1)	69.23	93	7.0	5.1
3.	NN + IBA (0.75 mg l-1)	70.18	89	6.0	4.2
4.	NN + IBA (1.00 mg l-1)	89.09	81	9.1	5.3
5.	NN + IBA (1.25 mg l-1)	45.31	83	8.9	5.0
	SE	1.56	1.30	0.337	0.176
	CD(P=0.05)	3.32	2.78	0.751	0.393

Acknowledgement

Authors thank the Dean, HC & RI, TNAU, Coimbatore for the financial assistance and man power extended towards the successful conduct of the experiment.

References

Cen,Y.Q., Jiang, R.M., Deng, Z.L. and Ni, D.X. 1993. *In vitro* propagation of *Anthurium andreanum* morphogehesis and effects of physical and chemical factors. *Acta Hort. Sinica.,* **20**: 187-192.

Chen, F.C., Kuehnle, A.R. and Sugii, N. 1997. *Anthurium* roots for micropropagation and *Agrobacterium tumefaciens*-mediated gene transfer. *Plant Cell Tis. Organ Cult.*, **49**: 71-74.

Dufour, L. and Guerin, V. 2003. Growth, developmental features and flower production of *Anthurium andreanum* Lind. in tropical conditions. *Scientia Horti.* **98**: 25-35.

George, P.S. and Ravishankar, G.A. 1996. *In vitro* multiplication of *Vanilla planifolia* using axillary bud explant. *Plant Cell Rep.,* **16**: 490-494.

Kuehnle, A.R. and Sugii, N. 1991. Callus induction and plantlet regeneration in tissue cultures of Hawaiian anthuriums. *Hort. Sci.,* **26**: 919-921.

Kuehnle, A.R., Chen, F.C. and Sugii, N. 1992. Somatic embryogenesis and plant regeneration in *Anthurium andreanum* hybrids. *Plant Cell Rep.,* **11**: 438-442.

Kunisaki, J.T. 1980. *In vitro* propagation of *anthurium andreanum* lind. *Hort Sci*., **15**: 508-509.

Martin, K.P., Joseph, D., Madassery, J. and Philip, V.J. 2003. Direct shoot regeneration from lamina explants of two commercial cut flower cultivars of *Anthurium andreanum* hort. *In vitro cellular and develop. biol-plant.,* **39**: 500-504.

Murashige, T. and Skoog, F. 1962. A revised medium for the rapid growth and bioassays with tobacco tissue cultures. *Physiol. Plant.*, **15**: 473-497.

Nitsch, J.P. 1969. Experimental androgenesis in Nicotiana. *Phytomorph.,* **19**: 389-404.

Pierik, R.L.M. and Steegmans, H.H.M. 1976. Vegetative propagation of *Anthurium scherzerianum* Schott through callus cultures. *Scientia Horti.,* **4**: 291-292.

Pierik, R.L.M., Steegmans, H.H.M. and Van Der Meys, J.A.J. 1974. Plantlet formation in callus tissues of *Anthurium andreanum* Lind. *Scientia Horti*., **2**: 193-198.

Puchooa, D. and Sookun, D. 2003. Induced mutation and *in vitro* culture of *Anthurium andreanum*. AMAS: 17-27.

Somaya, K.U., Narayanaswamy, P. and Jayaprasad, K.V. 1998.Micropropagation studies in *Anthurium andreanum* Lind. *Karnataka J. Agric. Sci.* **11**: 466 - 470.

Teresa, E., Vargas, Alexander Mejías, Maira Oropeza and Eva de García. 2004. Plant regeneration of *Anthurium andreanum* cv. Rubrun. *J. Biotech*., **7**: 3-11

Received: March 4, 2011; Accepted: June 18, 2011