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MICROPROPAGATION OF PINK PULP DRAGON FRUIT (*SELENICEREUS COSTARICENSIS* F.A.C. WEBER)

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Abstract

The dragon (*Selenicereus costaricensis* F.A.C. Weber) fruit is an important fruit crop of India. In the present study, shoot tips of *S. costaricensis* were used as explants and were inoculated into MS medium supplemented with different growth regulator combinations to produce quality and genuine planting materials. Maximum shoot initiation and proliferation were observed on MS medium supplemented with 3 mg/l BAP + 0.5 mg/l NAA + 1 mg/l Kn. Similarly, maximum number of multiple shoots (8) and highest shoot length (4.05 cm) were noticed on MS medium supplemented with 7 mg/l of BAP + 0.5 mg/l NAA + 1 mg/l Kn. However, maximum root proliferation (82.50%) and highest root length (2.86 cm) were achieved on half MS medium fortified with 0.5 mg/l NAA + 1.0 mg/l IBA + 1 g/l AC. Maximum survival percentage was recorded in the hardening media which contains sand, soil and FYM in the ratio of 1 : 2 : 1. This protocol may be used for large scale production of genuine and quality planting materials.

Dragon (*Selenicereus costaricensis* F.A.C. Weber) fruit, one of the potential fruit crops of India, is getting commercial importance in the recent past (Karunakaran *et al*. 2019). It is rich in antioxidants, phosphorous, calcium, iron, vitamin B_1, vitamin B_2, vitamin B, dietary fibre and many other compounds and its production in India is steadily expanding (Wakchaure *et al*. 2021). A nitrogen containing compound known as "betalains" especially red betalain is present in red and pink color pulp and peel of fruits and it is commercially used as natural food colourant (Choo *et al*. 2019). Lately, area expansion under dragon fruit is gaining momentum in different parts of India which is originated in Mexico and Central and South America (Karunakaran *et al.* 2019). Even though, dragon fruit is commercially propagated through cuttings, the demand is mismatched with availability for quality planting materials due to larger vines required for mass multiplication through cuttings. Plant tissue culture is an alternative method for large scale plantlets production. The method is a more efficient alternative to traditional cuttings, which are slow and can transmit diseases. Few studies have been examined for micropropagation of dragon fruit for the production of rapid and high-quality planting material (Bozkurt *et al.* 2022, Lee and Chang 2022, Ismail *et al*. 2023). Hence, in the present study, it is aimed to establish an efficient micropropagation protocol for dragon fruit so that more plantlets with healthy shoot and root system can be produced to meet out the growing demand of quality and genuine planting materials.

The present study was carried out at Tissue culture laboratory, Department of Fruit Science, Horticultural College and Research Institute, Tamil Nadu Agricultural University, Coimbatore. Shoot tips (2 cm) of *S. costaricensis* were used as explants and the planting materials were collected from university orchard, Department of Fruit Science, Tamil Nadu Agricultural University, Coimbatore, Tamil Nadu, India. Shoot tips were washed thoroughly under running tap water for 10 min and then treated with Tween 20 for 5-10 min. followed by pretreatment with bavistin (0.2%) for 10-15 min and 0.5% streptomycin for 30 min and then washed with distilled

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water. It was disinfected with 70% ethanol for 30 sec followed by surface sterilized with 0.1% $HgCl_2$ for 3 min and the explants were thoroughly washed under sterile water for 3 to 4 times and then dried on a sterile filter paper.

Murashige and Skoog (MS) medium supplemented with different growth regulator combinations used for plantlets development. Explants were cultured in bottles, which were kept at a temperature of 25 ± 2°C, 60-70% relative humidity, a photoperiod of 16 hrs of light and 8 hrs of darkness and a light intensity of 3000 lux. This experiment was carried out in Completely Randomized Design (CRD) with three replications in each treatment and observations recorded as percentage. Data was recorded at regular intervals and was analysed using ANOVA as suggested by Panse and Sukhatme (1985). The results were interpreted using MS Excel spreadsheet.

In the present study, the lowest time taken for shoot initiation (34.50 days) and maximum shoot proliferation (72.14%) was achieved on MS medium supplemented with 3 mg/l of BAP + 0.5 mg/l of NAA + 1 mg/l of Kn. Maximum number of multiple shoots (8) and maximum shoot length (4.05 cm) were attained on MS medium supplemented with 7 mg/l of BAP and 1 mg/l of Kn compared to other treatments (Table 1). Similarly, number of roots (3.35) and root length (1.59 cm) were recorded maximum on half strength MS + 1 g/l of activated charcoal + 0.5 mg/l of NAA + 1.0 mg/l of IBA (Table 2).

Table 1. Influence of growth regulator combinations on the growth of dragon fruit.

SI. No.	Treatments	Days for shoot initiation	Shoot proliferation (%)	No. of multiple shoots	Shoot length (cm)
T_1	MS + 2 mg/l BAP + 0.5 mg/l NAA+ 1 mg/l Kn	45.25	55.65 (48.26)	2	3.12
T_2	MS + 2.5 mg/l BAP + 0.5 mg/l NAA+ 1 mg/l Kn	39.00	64.84 (53.65)	2	3.38
T_3	MS + 3.0 mg/l BAP + 0.5 mg/l NAA+ 1 mg/l Kn	34.50	72.14 (58.17)	3	3.53
T_4	MS + 4.0 mg/l BAP + 0.5 mg/l NAA+ 1 mg/l Kn	41.75	67.39 (55.20)	4	3.94
T_5	MS + 5.0 mg/l BAP + 0.5 mg/l NAA+ 1 mg/l Kn	43.69	63.42 (52.81)	5	3.87
T_6	MS + 6.0 mg/l BAP + 0.5 mg/l NAA+ 1 mg/l Kn	40.21	59.53 (50.51)	6	3.66
T_7	MS + 7.0 mg/l BAP + 0.5 mg/l NAA+ 1 mg/l Kn	37.50	70.91 (57.38)	8	4.05
	SEd	2.18	2.68	0.22	0.19
	CD (0.05%)	4.54	5.63	0.48	0.73

Table 2. Effects of plant growth regulator combinations on roots.

SI. No.	Treatments	No. of roots	Root length (cm)
R_1	½ MS + 0.5 mg/l NAA + 0.5 mg/l IBA + *AC 1 g/l	2.23	1.31
R_2	½ MS + 0.5 mg/l NAA + 1.0 mg/l IBA + AC 1 g/l	3.35	1.59
R_3	½ MS + 0.5 mg/l NAA + 2.0 mg/l IBA + AC 1 g/l	2.62	1.43
R_4	½ MS + 1.0 mg/l NAA + 0.5 mg/l IBA + AC 1 g/l	2.90	1.57
R_5	½ MS + 1.0 mg/l NAA + 1.0 mg/l IBA + AC 1 g/l	2.49	1.48
R_6	½ MS + 1.0 mg/l NAA + 2.0 mg/l IBA + AC 1 g/l	2.23	1.35
R_7	½ MS Basal (Control)	1.14	0.33
	SEd	0.14	0.07
	CD (0.05%)	0.30	0.16

*AC – Activated Charcoal.

In the present study, hardening media such as soil, sand, compost, coco peat and potting mixture of soil, sand and FYM was employed for hardening of micropropagated dragon fruit plantlets (Fig. 1). The maximum *ex vitro* survival percentage (75%) of dragon fruit plantlets using shoot tips was recorded in hardening media of pot mixture containing sand: soil: FYM in the ratio of 1: 2: 1. Primary hardening was carried out at mist chamber for 30 days and secondary hardening were happened in shade net conditions for 30 days. For one week, plantlets were allowed to acclimatize at 25°C of fluorescent light and a 16 hrs light/8 hrs dark cycle. Then, they were transferred to a greenhouse and the plastic bag was removed after the second week. Once a week, watering was done. After two months, plantlets were transferred into individual pots and given 15 days once watering intervals and it grows up to a height of 15-20 cm.

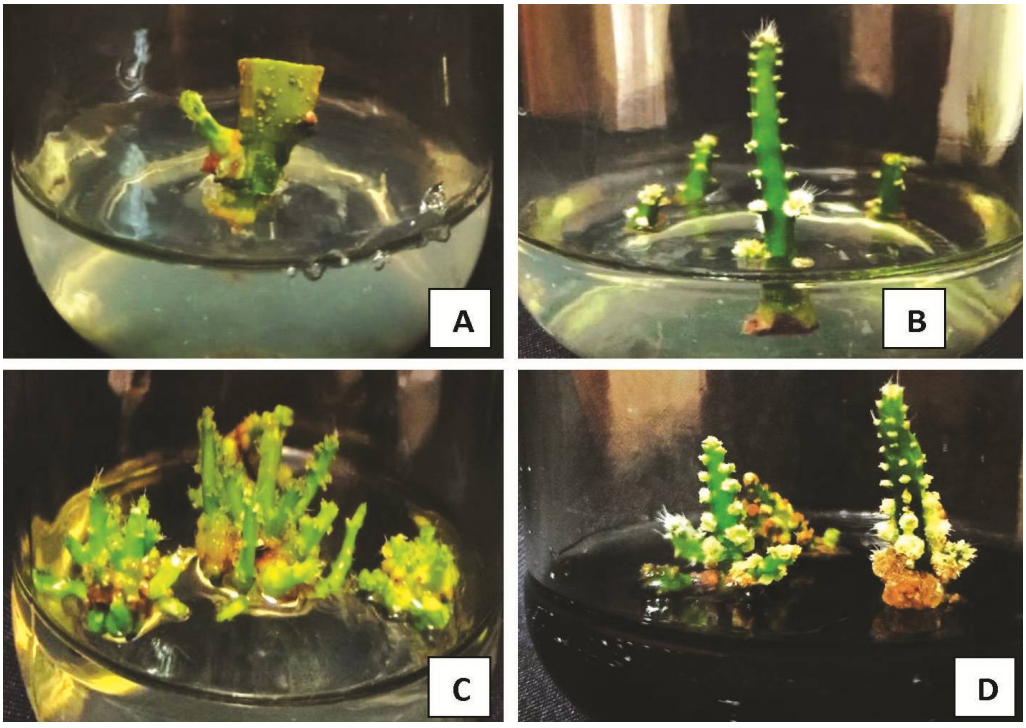

Fig. 1. *In vitro* propagation of dragon fruit using shoot tips. A: Shoot initiation stage. B: Shoot proliferation stage, C: Multiplication of shoots and D: Rooting of *in vitro* derived shoots.

In the present study, BAP and Kn resulted in progressive increase of multiplication rate of 1: 2. During the initial establishment stage, shoot tips containing areoles produced two number of shoots from a single explant and when go first subculture it resulted in four number of shoots whereas, at the end of third subculture it produced eight number of multiple shoots. From the above experiment, it is concluded that hundreds of dragon fruit plants could be produced by enhancing the number of subcultures in order to meet out the growing demands.

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