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Development of micropropagation protocol for jackfruit (*Artocarpus heterophyllus* Lam.) KJ 182

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Abstract

The most common method of propagating jackfruit is by seed, yet vegetative propagation techniques are labor-intensive and seasonal, and plantlets cultivated from seeds are heterozygous and do not conform to type. Hence, micropropagation can be used to overcome the limitations of conventional vegetative propagation methods. A better culture establishment was obtained with shoot tips cultured on MS medium with 2 mg L^{-1} BA. On MS media treated with BA (1-4 mg L^{-1}) and 0.35 mg L^{-1} GA_3, shoots proliferate. The highest number of shoots were obtained in MS medium supplemented with 3mg L^{-1} BA and 0.35 mg $L^{-1}GA_3$ and the highest shoot length and maximum number of leaves were recorded in MS medium supplemented with 2.5 mg L^{-1}BA and 0.35 mg $L^{-1}GA_3$. These proliferating shoots failed to yield any roots. Therefore, this could be a possible future career path.

Keywords: BA, Jackfruit, Micropropagation, Shoot multiplication

Introduction

Jackfruit (*Artocarpus heterophyllus* Lam.) belongs to the mulberry family Moraceae. It is a native of the Western Ghats of India and is commonly found in Asia, Africa, and some regions in South America. Fruits are a great source of fiber, calcium, phosphorus, potassium, magnesium, proteins, vitamins, and phytochemicals (Prem et al., 2015). Jackfruit is referred to as a poor man's fruit because it contributes to the food supply when there is an adequate shortage of food grains (Singh et al., 1963). Jackfruit is monoecious, producing male and female inflorescence on the same tree (Bose, 1985) and is a cross-pollinated crop. It is an evergreen medium sized latex producing tree, growing up to a height of 8-25 m, with a somatic chromosome number of 2n = 4x = 56 (Prakash et al., 2009). The tree is designated as the national fruit of Bangladesh and Indonesia. The jackfruit tree produces the world's largest known edible fruit, with an individual fruit weight of up to 50 kg, measuring up to 90 cm in length and 50cm in diameter, and yielding 20 to 250 fruits per tree per annum, and sometimes even up to 500 fruits on a fully mature tree (Haq, 2006; Shyamalamma et al., 2008).

The most common method of propagation of jackfruit is by seed, but because the crop is cross-pollinated and highly heterozygous, the plants raised from seeds are not true to type. Furthermore, due to the recalcitrant nature of the seeds, seeds stored even for a short time lose viability, resulting in poor germination. The vegetative propagation methods are generally tiresome, time consuming and seasonal with a low multiplication rate, making it difficult for effective and commercial level propagation. Therefore, the shortcomings of traditional vegetative propagation can be addressed via micropropagation.

The main aim of the present study was to establish a micropropagation protocol for jackfruit

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(*Artocarpus heterophyllus* Lam.). KJ 182 Chirapuram cluster jack, which is a promising jackfruit accession in Kasaragod, Kerala (Ajeesh, 2018). The tree produces an average of 8 fruits per cluster, and the fruits mature one after another, ensuring a long harvesting period for clustered jackfruit types. Thus, there is a need to multiply such jack plants in a shorter period of time.

Materials and methods

This investigation on "Development of micropropagation protocol for jackfruit (*Artocarpus heterophyllus* Lam.)" was carried out in the plant tissue culture laboratory of the Regional Agricultural Research Station (North Zone), Pilicode, Kasaragod (Dt.), Kerala, during the years 2018-2020.

Explant collection and surface sterilization of explants

Shoot tips collected from the current-season growth of mature KJ 182 jack were used for the experiment. Shoot tips of 5 cm length were thoroughly cleaned using a detergent solution for 10 minutes, then rinsed with distilled water for three times, followed by treating with Carbendazim+ Mancozeb (Saaf) (Saaf ,1 gram in 200 ml water) for one hour and thoroughly rinsing with sterilized distilled water for five to six times. After that, explants were surface sterilized using mercuric chloride (0.16g/ 200 ml of water) for 10 minutes, then rinsed with sterile distilled water five to six times to remove the traces of $HgCl_2$.

Establishment stage of jackfruit explants

The sterilized explants were cultured individually into culture bottles containing 20-30 ml of media (MS medium, ½ MS medium, modified MS medium, woody plant medium (WPM), and Gamborg B) as per the treatments. Each medium was provided with 2 mg L^{-1} BA. Modified MS medium was prepared by reducing the concentration of macroelements by half while retaining the concentration of microelements the same as that of MS medium. Cultures were incubated at 25 ± 3°C under white fluorescent light with an intensity of about 2000 lux for 16 hours per day. The number of days for shoot emergence, the number of days for shoot multiplication and branching, the number of shoots per explant, shoot length (cm), and the number of leaves per shoot were recorded.

Multiplication stage

Explants were cultured on MS medium with different concentrations (1, 1.5, 2.0, 2.5, 3.0, 3.5, 4.0, and 4.5 mg L^{-1}) of BA and 0.35 mg $L^{-1}GA_3$ for multiple shoot induction. Subculturing was done eight times at 21 days interval for shoot multiplication.

Rooting stage

Multiplied jackfruit shoots were cultured on half strength MS medium supplemented with various concentrations of NAA (1 and 3mg L^{-1}) and IBA (0.5 and 1.0mg L^{-1}) and their combinations.

All experiments were carried out in a completely randomized design. The first experiment had five treatments replicated four times. The second and third experiments had eight and nine treatments replicated three times. The observations recorded were subjected to statistical analysis using WASP 2.0 software prepared by the ICAR Research Complex for Goa and OPSTAT.

Results and Discussion

Establishment stage

Among the various media tried, MS medium fortified with 2 mg L^{-1}BA was found to be significantly superior with regard to number of days to shoot emergence (13.16), number of days to shoot multiplication and branching (15.83), shoot number per explant (1.83), shoot length (1.13), and number of leaves (1.66), followed by modified MS medium with 2 mg L^{-1}BA (Table 1 and Plate 1). The present study revealed that MS medium augmented with 2 mg L^{-1}BA showed the best result with respect to

Table 1: Effect of treatments on culture establishment of jackfruit explant

Sl. no	Treatment	No. of days to shoot emergence	No. of days for shoot multiplication and branching explant	No. of shoot/ length (cm)	Shoot	No. of leaves /shoot
1	T_1	13.16^{c}	15.83^{c}	1.83^{a}	1.13^{a}	1.66^{a}
2	T_2	14.41^{bc}	17.33^{b}	1.16^{b}	0.95^{b}	1.08^{b}
3	T_3	13.99^{c}	16.08^{c}	1.41^{ab}	0.90^{bc}	0.74^{bc}
4	T_4	16.08^{a}	18.66^{a}	1.00^{b}	0.76^{c}	0.33^{c}
5	T_5	15.41^{ab}	18.58^{a}	1.16^{b}	0.82^{bc}	1.00^{b}
	SE(±m)	0.41	0.40	0.15	0.05	0.14
	C. D. at 5 % level	1.25	1.20	0.47	0.17	0.44

T_1: MS medium + 2 mg L^{-1}; T_2: ½ MS medium + 2 mg L^{-1} ; T_3: Modified MS medium + 2 mg L^{-1} ; T_4: Woody plant medium (WPM) + 2 mg L^{-1}, T_5: Gamborg B… medium+ 2 mg L^{-}

growth parameters. WPM and B_5 medium provided with 2 mg L^{-1}BA showed poor results for all the growth parameters compared with other treatments. The composition of media has a greater influence on the establishment of culture. MS medium contains almost all nutrients required for plant growth in a higher amount compared with other media used in the study. Nitrogen is the prime component in MS medium, and it is found in the form of NH_4NO_3 and KNO_3, which influence plant growth and morphogenesis. This result was in agreement with the findings of Adiga (1996).

Multiplication stage

The multiplication stage of jackfruit shoots has been given in Plate 2 and recorded in Table 2. MS medium fortified with 2.5 mg L^{-1}BA and 0.35 mg L^{-1} GA_3 took fewer days for shoot emergence, followed by MS medium with 2 mg L^{-1}BA and 0.35 mg $L^{-1}GA_3$, and MS medium fortified with 4.5 mg L^{-1}BA and 0.35 mg $L^{-1}GA_3$ took more days to shoot emergence. BA at 3 mg L^{-1} and GA_3 at 0.35mg L^{-1} gave the highest number of shoots/ explant compared with other treatments (Fig. 1). BA at 2.5mg L^{-1} and 0.35 mg $L^{-1}GA_3$ gave the highest significant shoot length (2.26). The lowest shoot length was observed in MS medium + 4 mg L^{-1}BA + 0.35 mg L^{-1} GA_3. The number of leaves per explant showed significantly different values. The maximum number of leaves was recorded in MS medium augmented with 2.5 mg L^{-1}BA and 0.35 mg $L^{-1}GA_3$ (4.44). MS medium supplemented with 4.5 mg L^{-1}BA and 0.35 mg $L^{-1}GA_3$ (2.55) produced a lesser number of leaves. The shoot multiplication and elongation might be due to the role of cytokinin in overcoming the apical dominance and GA_3 for shoot elongation. The result clearly indicates that at a lower concentration of BA, shoot proliferation was lower, and it showed a positive response in all the growth parameters when the concentration was

Table 2: Effect of treatments on shoot emergence, no. of shoots/ explant, shoot length and no. of leaves/ explant

Sl no.	Treatment	No. of days to shoot emergence	No. of shoot/ explant	Shoot length (cm)	No. of leaves/ explant
1	T_1	11.22^{bcd}	2.21^{de}	1.15^{bc}	3.00^{bc}
2	T_2	12.10^{ab}	2.33^{cde}	1.30^{bc}	3.21^{bc}
3	T_3	10.88^{cd}	2.55^{bcd}	1.27^{bc}	3.33^{bc}
4	T_4	10.55^{d}	3.21^{ab}	2.26^{a}	4.44^{a}
5	T_5	10.99^{cd}	3.88^{a}	1.10^{bc}	3.88^{ab}
6	T_6	11.66^{abc}	3.00^{bc}	1.55^{b}	3.66^{ab}
7	T_7	12.32^{a}	1.88^{de}	1.03^{c}	2.99^{bc}
8	T_8	12.44^{a}	1.66^{e}	1.07^{bc}	2.55^{c}
SE(±m)		0.36	0.24	0.17	0.30
C. D. at 5 % level		1.09	0.73	0.52	0.92

T• : MS medium + 1 mg L^{-1}BA + 0.35 mg L^{-1} GA_3 ; T, : MS medium + 1.5 mg L^{-1}BA + 0.35 mg $L^{-1}GA_3$; T_3 : MS medium + 2 mg L^{-1} BA + 0.35 mg L^{-1} GA_3 ; T_4 : MS medium + 2.5 mg L^{-1}BA + 0.35 mg $L^{-1}GA_3$; T_5 : MS medium + 3 mg L^{-1}BA + 0.35 mg $L^{-1}GA_3$; T_6 : MS medium + 3.5 mg L^{-1}BA + 0.35 mg $L^{-1}GA_3$; T_7 : MS medium + 4 mg L^{-1}BA + 0.35 mg $L^{-1}GA_3$; T_8 : MS medium + 4.5 mg L^{-1}BA + 0.35 mg L^{-1}GA

increased, but further increases in BA concentration showed a reduction in growth character. This might be due to the inhibition of adventitious meristem elongation also reported by Sajid et al. (2006) and Borchetia et al. (2009). Gayathri and Sathyanarayana (2015) reported that MS medium supplemented with 2 mg L^{-1}BA produced more number of shoots and leaves, highest shoot length in jackfruit but in the present study BA concentration ranging from 2.5-3mg L^{-1} produced more number of multiple shoots, more number of leaves and highest shoot length.This variation might be due to the difference in endogenous level of hormones related to the genetic factor of the variety used in the present study.

Multiplied shoots were kept in the rooting media for about 3-4 months, and all treatments failed to produce roots (Plate 3). Inhibition of rooting might be due to the formation of a basal callus, which will affect the rooting competence of microplants by interfering with the physiological process by trapping growth constituents like growth regulators. Also, the exudation of phenolic compounds may be another reason for poor rooting in jack (Vengadeshan et al., 2002). Further detailed studies are warranted in this arena.

Conclusion

The present study describes that among the basal mediums, MS medium fortified with 2 mg L^{-1}BA is good for culture establishment. MS medium fortified with 2.5-3 mg L^{-1}BA along with 0.35 mg $L^{-1}GA_3$ can be used for shoot multiplication.

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References

Adiga, D.J. (1996) Clonal Propagation of Jackfruit (*Artocarpusheterophyllus* Lam) cv. Singapore Jack through tissue culture.*Curr. Sci*. 78: 1231- 1234.

Ajeesh, B.R. 2018. Field evaluation of promising jackfruit (*Artocarpus heterophyllus* Lam.) types. MSc Hortic.Thesis, KAU, Thrissur.

Borchetia, S., Das, S.C., Handique, P.J. and Das, S., 2009. High multiplication frequency and genetic stability for commercialization of the three varieties of micropropagated tea plants (*Camellia spp*.). *Sci. Hort.*, 120(4): 544-550.

Bose T.K. 1985. Jackfruit. In: Mitra, B.K. (ed.), *Fruits of India: Tropical and Subtropical*. NayaProkash, Calcutta, India, pp. 487-497.

Gayathri, R. and Sathyanarayana, B.N., 2015. *In vitro* culture of three elite clones of jack fruit.*The bioscan* 10(2): 713-717.

Haq, N. 2006. Jackfruit, *Artocarpus heterophyllus* L. International Centre for underutilized crops. Southampton, UK, 20p.

Prakash, O., Kumar, R., Mishra, A. and Gupta, R. 2009. *Artocarpus heterophyllus* (Jackfruit): An overview. *Pharmacognosy Rev.* 3(6): 353–358.

Prem, J.V., Sajesh, N.K., Jiby, J.M., Ajesh, C.K., Benchamin, A., Renjith, J.M., Alen, N.A., Deenamol, T., Riny, S.T., Nijamol, V. and Sophyiamol, J. 2015. Chemistry and medicinal properties of jackfruit (*Artocarpus heterophyllus*): A review on current status of knowledge. *Int. J. Innovative Res. Rev* 3(2): 83-95.

Sajid, G.M., Ilyas, M.K. and Anwar, R. 2006. Effect of diverse hormonal regimes on *in vitro* growth of grape germplasm. *Pak. J. Bot*. 38(2): 385-391.

Shyamalamma, S., Chandra, S.B., Hedge, M. and Naryanswamy, N. 2008. Evaluation of genetic diversity in jackfruit (*Artocarpus heterophyllus* Lam.) based on amplified fragment length polymorphism markers. *Genet. Mol. Res.* 7(3): 645-656.

Singh, S., Krishnamurthi, S. and Katyal, S.L. 1963. *Fruit Culture in India.* ICAR, New Delhi, India. 451p.

Vengadesan, G., Ganapathi, A., Amutha, S. and Selvaraj, N. 2002. *In vitro* propagation of *Acacia* species - a review. *Plant Sci.* 163(4): 663-671.