

Micropropagation of *Psidium guajava* from root tips of mature plant

Momamed Ismail^{*}, Abdul Razak TK, Tajuj Kumar and Mohamed Ali

Department of Biochemistry, Priyadarshini College of Pharmacy, Koratagere, Karnataka, India
And

NIMS University, Jaipur, Rajasthan, India.

Abstract

In the present work, tissue culture of *Psidium guajava* (guava) through root tip culture from well developed plants was carried out. Root tips after sterilization was cultured on modified Murashige and Skoog (MS) medium supplemented with different concentration and combination of BA (Benzyl Adenine), IAA (Indole acetic acid), and L-glutamine. More number of roots (70%) was developed into plantlets when MS was supplemented with BA 1mg/L combined with IAA 2.5mg/L glutamine 500mg/L. MS control and BA gave 1mg/L combined with IAA 2mg/L and glutamine 250mg/L gave minimum (20%) plantlets. .

*Corresponding
author

Key words: Tissue culture, *Psidium guajava*, micropropagation.

1.0 Introduction

Psidium guajava belongs to the family myrtaaceae. It is rich sources of vitamins, minerals organic acids and pectins [1]. It is considered as the “poor man’s fruit”. So many different phytochemicals provide guava with many unique properties and its actions including anti-microbial, astringent, bactericidal, hypoglycemic, laxative, anthelmintic, nutritive and spasmolytic [2]. It is also reported that regular consumption of guava fruit significantly increases the good cholesterol and blood pressure [3]. Medicinal uses of guava have been reported involving gastroenteritis, dysentery, wounds, ulcers, rheumatics, and toothache [4]. Our own primary research has demonstrated antimicrobial and anthelmintic activity of guava leaf extracts [5,6]. It is our belief that with appropriate tissue culture techniques it is possible to improve overall health and well-being of the general public. The research objective were to study the growth rate of root tips of *Psidium guajava* on various concentration and combinations of growth hormones on rooting of guava culturing on MS medium.

2.0 Materials and Methods.

Root tips of *Psidium guajava* were obtained from farmland. Guava plants root tips were used as explant materials, which were disinfected for 15 minutes in 15% bleach solution (0.78% NaOCL) containing 20 drops Tween-20 per liter, and rinsed immediately with sterile distilled water 4 times. The softwood roots was cut into 0.5cm segments as explants, Then the root tips were rinsed three times with sterile distilled water under laminar flow bench [7]. Murashige & skoog basal medium was supplemented with different concentrations of Benzyl Adeninine (BA), Indole acetic acid (IAA) and glutamine and 3.0% sucrose,

0.7% agar was used for culturing the root tips. The pH of the medium was adjusted to 5.8 with 1N KOH and 1N HCL. The culture media were sterilized by autoclaving at 121 °C for 20 minutes.

The Explants were placed vertically on the medium in the test tubes containing 10ml medium. The test tubes were covered with autoclaved polypropylene sheet and tightened with rubber band. The cultures were kept in growth chamber at $24 \pm 2^\circ\text{C}$ under a 16 hours of light per day provided by cool white fluorescent tubes [8].

After every 4weeks, sub culture was transferred to baby food jars, 12 weeks plantlets were transferred to modified MS medium with different concentration of auxins for root induction. The experiment was arranged in a completely Randomized Design with three replications per treatment, each with 10 explants.

Plantlets were then transferred to medium in Jiffy pots containing clay, silt and sand. Before transplanting the plantlets were washed with distilled water to remove medium from the roots. Initially the plantlets were covered with transparent plastic in order to maintain high humidity and then small holes were made in the plastic to acclimatize it gradually. The plantlets were watered with half knop’s solution and plastic cover was removed after a week . A regular irrigation to these plantlets were practiced at 15days interval and later on shifted to natural environment [7,8].The data is given in Table-1 and Table-2.

3.0 Result and Discussion

Action of Indole acetic acid (Auxin) on rooting

The data related to the effect of indole acetic acids on rooting of guava when cultured on MS medium showed that rooting percentage is high (52) when IAA

2.5mg/l combined with BA 1mg/L followed by IAA 2mg/L and BA 1mg/L with average number of roots 3.6 and 3.15 respectively [9]. The most inferior results were obtained when BA 1mg/L was used alone, and no rooting was observed in auxin free medium. In the present studies both BA and IAA 2.5mg/L gave highest percentage of rooting.

Action of growth hormones on shoots developed into plantlets

The data reveals that action of growth hormones on shoots developed into plantlets was effective. BA 1mg/L along with glutamine 500mg/L gave higher shoot development (70%) when cultured on MS medium. When the concentration of BA was reduced to 0.5mg/L, The number of shoots decreased with the same concentration of glutamine.

Media concentration	No of plants Failed to root	No of plants rooted	Average No of roots/plants
MS(control)	50	0	-
MS+BA1mg/L	26	14	2.4
MS+BA1mg/L	14	26	3.4
MS+IAA2.5mg/L	26	24	2.6
MS+BA1mg/L+IAA2.5mg/L	6	44	3.8

Table-1: Effect of various concentration of IAA on rooting of guava culturing on MS medium,

Media concentrations	No. of Shoot failed	No of shoots Developed into plantlets	AV. no. of Shoots/plantlet
MS(control)	37	13	1.5
MS+BA0.5mg/L	25	25	2.0
MS+BA0.5mg/L+ glutation250 mg/L	28	22	2.9
MS+BA 1mg/L+ glutation500mg/L	15	35	3.0

Table-2: Action of different concentration of growth hormones on development of guava shoots when supplemented to MS Medium

Reference

1. Guochen Yang and zhongge. Guava callus production under difficult culture medium and plant growth regulator conditions. Proceedings 33rd PGRSA Annual Meeting.
2. Samson IA. Tropical fruits, 2nd ed, Tropical Agriculture series, long man Scientific & Technical, Long man Inc., New York. 1986.
3. Conde Garcia EA, Nascimento VT, Santiago Santos AB. Inotropic effects of extracts of *Psidium guajava* L. (guava) leaves on the guinea pig atrium. Braz J Med Biol Res. 2003; 36(5):661-668.
4. Jemenz- Escrig A et al. Guava fruits as a new source of antioxidant dietary fiber. J Agric Food Chem. 2001; 49 (11):5489-5493.
5. Mohamed Ismail et al. Antibacterial Activity of Guava leaves extracts. J Pharm Pract. 2011;1(2); 47-48.

6. Mohamed Ismail et al. Anthelmintic Activity of Guava leaf extract. *Int J Res Pharm Biol Sci.* 2012; 3(1):1-2.
7. Khattak MS, Malik MN, Khan MA. *In vitro* micro-propagation of guava. 1993. Thesis submitted to Department of Botany, University of Peshawar, Pakistan.
8. Singh SK, Meghwal PR, Shamand HC, Singh SP. Direct shoot organogenesis and hopocotyl explants from In-vitro germinated seedlings of *psidium guajava* L. *Scientia Horticulture.* 2002; 95: 213-221.
9. Roshan Zamir N Ali et al. In-vitro Re-Generation of Guava (*Psidium guajava*) from shoot tips of mature trees. *Pak J Bot.* 2007; 39(7):2395-2398.