

## Establishment of a rapid *in vitro* system for regeneration of papaya plantlets in a liquid media

Gatambia, E.K.<sup>1</sup>, Kihurani, A.W.<sup>2</sup>, Wanzala F.K.R.<sup>2</sup> & Waiganjo, M.M.<sup>1</sup>

<sup>1</sup>Kenya Agricultural Research Institute -THIKA, Thika, Kenya

<sup>2</sup>Department of Horticulture, Jomo Kenyatta University of Agriculture and Technology, Nairobi, Kenya

**Corresponding author:** eliudgatambia@yahoo.com

### Abstract

This study was an attempt to determine rapid micro- propagation methods for regeneration of papaya plantlets *in vitro* using liquid media. Different concentrations and combinations of growth regulators in liquid MS namely BAP (0-2 mg l<sup>-1</sup>) and NAA (0-1 mg l<sup>-1</sup>), CPPU (0-2 mg l<sup>-1</sup>), and IBA (0-3 mg l<sup>-1</sup>) were used in the experiments. Results showed that 0.2 mg l<sup>-1</sup> BAP, 0.5mg l<sup>-1</sup> CPPU were best in shoot multiplication, while 3.0 mg l<sup>-1</sup> of IBA in root initiation. Shoot proliferation, root induction reported in this study is an indicator that liquid medium could be a rapid method for papaya regeneration. Further experiments geared towards optimisation of shoot elongation and rooting are ongoing.

Key words: *Carica papaya*, efficient regeneration, micro propagation, plant growth regulators

### Résumé

Cette étude était une tentative pour déterminer des méthodes rapides de micro-propagation pour la régénération de plantules de papaye *in vitro* en utilisant des milieux liquides. Différentes concentrations et combinaisons de régulateurs de croissance à l'état liquide de MS à savoir, BAP (0-2 mg l<sup>-1</sup>) et NAA (0-1 mg l<sup>-1</sup>), CPPU (0-2 mg l<sup>-1</sup>), et IBA (0-3 mg l<sup>-1</sup>) ont été utilisées dans les expériences. Les résultats ont montré que 0,2 mg l<sup>-1</sup> de BAP, 0.5 mg l<sup>-1</sup> de CPPU étaient les meilleures dans la multiplication des pousses, tandis que 3,0 mg l<sup>-1</sup> de l'IBA dans l'initiation des racines. La prolifération des pousses et l'induction des racines, rapportées dans cette étude, sont un indicateur que le milieu liquide peut être une méthode rapide pour la régénération de la papaye. D'autres expériences orientées vers l'optimisation de l'allongement des pousses et l'enracinement sont en cours.

Mots clés: *Carica papaya*, régénération efficace, micro propagation, régulateurs de croissance des plantes

## Background

Papaya originated from Eastern Central America, but today, it is naturalised in many areas including East Africa (Morton, 1987). It is polygamous with three sex types namely, male, female, and hermaphrodite. It has potential to produce fruits throughout the year. However, papaya producers are constrained by many challenges like, the polygamous nature of the crop which makes it difficult to distinguish among the sex types at seedling stages (Louw, 1999). Besides, the recent outbreak of papaya ring spot virus (PRSV), which is very difficult to control, in all papaya producing areas of the world has not spared East Africa. *In vitro* meristem culture has proved useful in eradication of viral and other plant diseases (Poehlman, 1987). This study attempts to address the aforementioned sex paradox and generate large quantities of clean plantlets through *in vitro* culture of papaya meristems in liquid media.

## Literature Summary

Rapid regeneration of papaya through liquid medium increases contact with tissue which stimulates and facilitates the uptake of nutrient, phytohormones, leading to better shoot and root growth. Continuous shaking reduces expression of apical dominance leading to induction and proliferation of numerous axillary buds. The shake culture conditions, boosts multiplication rate of shoots due to improved aeration (Shakti Mehrotra1, 2007).

## Study Description

The present study was conducted at Jomo Kenyatta University of Agriculture and Technology (JKUAT) and Kenya Agricultural Research Institute (KARI) tissue culture labs in Kenya. Meristems of selected lines were harvested from greenhouse, washed, sterilised and cultured in Murashige and Skoog (MS) liquid basal medium (Murashige and Skoog, 1962) supplemented with BAP (6-benzylaminopurine) ( $0.2 \text{ mg l}^{-1}$ ) and NAA (Naphthalene acetic acid) ( $0.1 \text{ mg l}^{-1}$ ), CPPU ( $0.2 \text{ mg l}^{-1}$ ), and IBA (Indole-3-butyric acid) ( $0.3 \text{ mg l}^{-1}$ ) 3% sucrose for shoot induction, elongation and IBA for rooting. They were put in a growth room at  $27 \pm 1^\circ\text{C}$ , under white florescent lamp ( $40 \mu \text{ mol, } \mu 125^{-1}$ ). They were mounted on an orbital shaker with a speed of 120 rpm for 8 weeks. Data collection was done weekly. The data collected were analysed using (SAS) ver. 9.

## Research Application

Results showed that  $0.2 \text{ mg l}^{-1}$  BAP,  $0.5 \text{ mg l}^{-1}$  CPPU were best in shoot induction, while  $3.0 \text{ mg l}^{-1}$  of IBA in root initiation. Results corroborate with those obtained by Be and Debergh (2006). (2006) that more shoots were obtained when BAP concentration

was increased. Shoot proliferation, root induction reported in this study is an indicator that liquid medium could be a rapid method of papaya regeneration. Further experiments towards optimisation of shoot elongation and rooting are ongoing.

**Table 1. Effects of different concentration of hormones on leaf number shoot number and shoot length.**

Hormone conc (mg <sup>l</sup> <sup>-1</sup> )	Leaves	Length	Shoots
1. BAP 0.5	1.23 cd	0.27	0.90 c
2. BAP 0.1	2.47ab	0.73	2.25 ab
3. BAP 0.125	1.43 cd	0.33	1.78 b
4. BAP 0.2	2.94 a	0.85	2.8 a
5. BAP 0.1 + NAA 0.05	1.94bc	0.60	2.12 ab
6. BAP 0.125 + NAA 0.07	0.56 d	0.16	0.49 c
7. Plain MS Liquid	1.25 cd	0.38	0.92 c
Cv	32.67328	15.12373	31.53168
p-value	<.0001	<.0001	<.0001

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