Internat. J. Plant Sci. Vol.2 No.2 July 2007 : 79-84

Micropropagation of *Phyllanthus amarus* (Schum and Thom) through nodal shoot segment culture

MAHIPAL S. SHEKHAWAT* AND A. K. DIXIT[1]
Department of Botany, Arignar Anna Govt. Arts College, KARAIKAL (PONDICHERRY) INDIA
[1]Department of Botany, K.M. Center for P.G. Studies, (PONDICHERRY) INDIA

(Accepted :February, 2007)

SUMMARY

Methods were developed in present investigation for cloning and large scale plant production of *Phyllanthus amarus* (Shum &Thom) from the germplasm selected from the arid areas of Rajasthan, India. Nodal shoots segments were used as explants. These explants were dressed and surface sterilized. Multiple shoots were induced by proliferation of axillary buds/meristem on Murashige & Skoog's (MS) semisolid medium + 2.0 mgl^{-1} 6-Benzylaminopurine (BAP). The shoots of *P. amarus* were further multiplied on modified MS (MMS) + 0.1 mgl^{-1} Indole 3-acetic acid (IAA) + 0.5 mgl^{-1} each of BAP and Kinetin. The *in vitro* generated shoots were rooted on half-strength MS medium containing 2.0 mgl^{-1} Indole 3-butyric acid (IBA) + 2.0 mgl^{-1} IAA with 100 mgl^{-1} activated charcoal. By this method 78% shoots were rooted. The shoots could also be rooted by pulse treatment with 300 mgl^{-1} IBA solution for 4 minutes. Only 46% shoots were rooted within 2-3 weeks in this method. The *in vitro* rooted plants could be hardened and acclimatized in green house and transplanted to the field successfully.

Key words : *Phyllanthus amarus*, Cloning, Multiple shoots, *In vitro.*

The genus *Phyllanthus* (Family Euphorbiaceae) is widely distributed throughout the tropical and subtropical countries. *Phyllanthus amarus* (Bhumi amalika/Bhui Aonla) is found in semi-temperate and tropical parts of India. The infusion of leaves, stems, and roots of most species of plants of this genus has long been used in folk medicine in the world for thousands of years for treatment of broad spectrum of diseases. Clinical studies indicate that *Phyllanthus* plant have potential clinical utility on the treatment of hepatitis-B and Nephrolitiase and in painful disorders. *Phyllanthus amarus* is known to have constituents that are effective against jaundice (Hepatitis-A, Hepatitis-B and Hepatitis-E). The plant derived products reduce levels of bilirubin in liver cells infected by Hepatitis-B (Prasad, 1999). Phyllanthin is the major hepatoprotective compound of *P. amarus* and is concentrated maximum in leaves (1.56%) followed by stem (0.0104%) and root (0.007%). (CIMAP Report, 1998).

There have been limited studies on tissue cultures of *Phyllanthus* species. Tissue cultures of *Aonla* (*Phyllanthus emblica*) were analyzed for the presence of phyllantidine and phyllantine. Sehgal and Khurana (1985) reported on the tissue culture as mean of propagation of *P. emblica*. Shekhawat *et al*. (1999) reported on micropropagation of *Phyllanthus emblica*. Cultures of several species of *Phyllanthus* were established and their chemical constituents were isolated and characterized (Ishimaru *et al*., 1992 and cited by Calixto *et al*., 1998). Survey of existing literature reviewing that the micropropagation of selected *Phyllanthus amarus* has not been achieved. Sehgal and Khurana (1985) reported on morphogenesis and plant regeneration from cultured endosperm of *Emblica officinalis* (*Phyllanthus emblica*). Rajasubramaniam and Pardha Saradhi (1997) described a rapid multiplication method for *Phyllanthus fraternus*: a plant with anti-hepatitis virus DNA polymerase activity.

It has been pointed out that efficacy of *Phyllanthus amarus* to cure Hepatitis-B is affected by the soil and the geographical factors. It is cross-pollinated and seed raised plant show phenotypic variation. It seems that there is genetic variability among the *P. amarus* plants. This variability is probably responsible for variable constituents of antiviral components of the plant. There is need for developing technologies for propagation of germplasms selected for production of biomass (for harvesting desirable constituents) under particular environmental set of conditions.

MATERIALS AND METHODS

Explants and their sterilization:

The basic explanting materials (Candidate Plus Plants) of *Phyllanthus amarus* were collected from Ajmer and Sikar districts of Rajasthan, India and from the University New Campus (J. N. Vyas University,

* Author for correspondence.