Review

In vitro propagation of jasmine- a critical review

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ABSTRACT

Jasmine, *Jasminum sambac* (L) Aiton belonging to family Oleaceae is an ornamental plant that produces white flowers with a very pleasant fragrance and is extensively used in perfumery and for religious purposes. Jasmine does not bear seeds hence the plant is propagated solely by cuttings and layering and these methods of propagation are dependent on the season. Layering involves more time and restricts the number of plants propagated from a bush. However long term cutting/layering causes varietal degeneration, resistance weakness and declining flower production. In recent years plant tissue culture technique has become a powerful tool for the propagation of many plant species. Vegetative parts like nodes, shoot apices, stems and leaves have been used as explants for in vitro propagation. The explants are usually collected from field grown mature plants and thereafter inoculated on variously supplemented Murashige and Skoog's (MS) medium for multiple shoot proliferation and callus induction. Callus induction from leaf and meristem explants has been achieved using different concentrations of auxins and cytokinins. Maximum percentage of cultures showing callus proliferation from leaf explants was observed in the medium containing MS basal salts supplemented with 3.0 mg/l BAP and 2.0 mg/l IAA in leaf explants. White and friable callus was obtained in the medium containing MS basal salts supplemented with 2.0 mg/l BAP, 2.0 mg/l Kn and 2.0 mg/l IAA from meristem explants.

Keywords: Micropropagation; jasmine; growth regulators; explant; callus

INTRODUCTION

Among commercial loose flowers jasmine (*Jasminum sambac*) of the family Oleaceae is a genus comprising approximately 600 species of small trees and shrubs and is a native of India. It is an ornamental plant extensively used in perfume industry and for religious purposes which produces white flowers with a very pleasant fragrance (Davallo et al 2014). Jasmine's essential oil produces a distinctive fragrance and is used as a natural perfume and also for aromatherapy (Satuhu 2004). Its essential oil is used in preparation of different products such as industrial perfumes, soaps, cosmetics, foods and pharmaceuticals (Davallo et al 2014). It is endowed with large spectrum of commercial potentialities in perfumery and essential oil sector (Ambasta 1986, Chadha1976). The flowers are also used as ornamental for the production of flowers and tea. The species is highly variable possibly a result of spontaneous mutation, natural hybridization and autopolyploidy. Only a few varieties are propagated by seed in the wild. Cultivate *J sambac* generally does not bear seeds as a result it is difficult to develop new varieties through conventional breeding. The only way to develop new cultivars is either through somatic mutations or gene manipulation. Plant tissue culture is the first step in somatic mutation or gene manipulation.

In recent years plant tissue culture technique has become a powerful tool for the propagation of many plant species. Plant tissue culture offers an alternative method for the conservation of rare, valuable and endangered plant genotypes. This technique can successfully preserve the plant species which do not produce seeds or some of the seeds that cannot be stored for an extended period. However in vitro methods can be applied for preservation of vegetative tissues to keep the genetic background (Hussain et al 2012). Jasmine does not bear seeds and the plant is reproduced solely by cuttings and layering and these methods of propagation are dependent on the season. Layering involves more time and restricts the number

of plants propagated from a bush. It can also be propagated by cuttings. However long term cutting/ layering causes varietal degeneration, resistance weakness and declining flower production (Cai et al 2007).

In vitro culture presents itself as an attractive tool for mass multiplication of important fragrant plant species and for the production of secondary metabolites. There are few reports on regeneration of jasmine through tissue culture techniques (Khoder et al 1979).

Establishment of aseptic culture

Development of aseptic culture is most important for establishment of in vitro culture. Knowledge of physiological status and the susceptibility of the plant species to different pathological contaminants are desirable to establish disease free cultures in vitro. Usually young shoots are used as explants in *J officinale*. The explants were prepared by removing large leaves and cutting the shoots into 3 cm long pieces each containing either one or two axillary buds. Nodal explants (1.0-1.5 cm long) were prepared by trimming off the excess tissue. These were washed for 30 minutes in running tap water, treated with 0.03 per cent cetrimide solution for 10 minutes followed by thorough washing (5-6 times) with distilled water. Surface sterilization was carried out with 0.1 per cent $HgCl_2$ (w/v) for 10 minutes and the tissues were washed four times with sterile distilled water (Bhattacharya and Bhattacharyya 1997). Aseptic cultures were established in *J nudiflorum* by Zhou-min et al (2008). The explants were surface-sterilized by soaking them in saturated laundry powder solution for 10 minutes followed by washing them 2-3 times in running tap water. In the second step the explants were treated with 75 per cent ethyl alcohol for 20 seconds followed by washing 2-3 times with sterilized distilled water. Finally the explants were surface-sterilized by treating with 0.1 per cent $HgCl_2$ for 8 minutes followed by washing with sterilized distilled water thrice to remove traces of $HgCl_2$ adhering to the explant.

The leaves were collected from *J sambac* and washed with running tap water for 60 minutes followed by soaking in 3 per cent of Teepol soap solution for 15 minutes and rinsed in distilled water to reduce contamination. The leaves were disinfected by treating with 70 per cent ethanol for 45 seconds and washed with sterile distilled water thrice followed by treating with 0.1 per cent (w/v) aqueous $HgCl_2$ for 3 minutes and washing again with sterile distilled water (Davallo et al 2014). Young shoots of *J sambac* either apical or axillary bud were disinfected in two steps. In the first step the shoots were washed for 15 minutes in running tap water, cut into 1.5-2 cm, treated with 0.1 per cent Bavistin solution for 15 minutes followed by thorough washing (5-6 times) with distilled water. In the second step surface sterilization was carried out with 0.1 per cent $HgCl_2$ (w/v) for 10 minutes followed by washing 3 times with sterile distilled water (Biswal et al 2016). Salim (2016) used nodes and internodes as explants collected from an adult shrub of *J azoricum*. After removing leaves explants were cleaned with liquid soap to remove dirt followed by washing under running tap water for 1 h. Surface sterilization was achieved in the laminar air flow cabinet by treating the explants with 2 per cent (v/v) Clorox (6% NaOCl) solution containing 2 drops of Tween 20 for 18 minutes followed by rinsing with sterile distilled water (Salim 2016).

In vitro culture

The effect of growth regulators on multiplication of various plant species was reviewed by many workers (Bhattacharya and Bhattacharyya 1997, Zhou-min et al 2008, U-Kong et al 2012, Davallo et al 2014, Biswal et al 2016, Salim 2016). The apical meristem was capable of having unlimited proliferation potential to develop into new shoots. Shoot buds arose from individual cells in calli developed as a result of wound reaction from cut and injured tissues.

Meristem culture

Some of the workers used apical meristem for mass multiplication of *Jasminum* species. A single meristem produced a single shoot or multiple shoots depending on the variety and composition of the medium. Several plant tissue culture media were used including formulations suggested by Murashige and Skoog (1962). Micropropagation of *Jasminum* species using apical meristem or shoot primordium was reported by various researchers (Cai et al 2007, He et al 2011, Asanica et al 2008). The application of BA in the culture medium induced bud germination in jasmine. Sun et al (2009) observed highest bud induction rate in the WPM medium supplemented with 2.0 mg/l BA and 0.1 mg/l IBA. Shoot proliferation was maximum in the WPM medium supplemented with 1.0 mg/l BA and 0.2 mg/l IBA at an incubation temperature of 35°C. According to Cai et al (2007) medium suitable for optimum bud generation of jasmine is MS basal medium combined with benzyl adenine (2.0 mg/l) and NAA (0.1 mg/l). They also observed that MS basal medium

combined with benzyl adenine (1.5 mg/l) and NAA (0.3 mg/l) is optimum for multiple shoot induction in axillary bud. Bhattacharya and Bhattacharyya (1997) successfully micropropagated *J officinale* by culturing nodal segments. Maximum shoot proliferation was obtained by culturing nodal segments of *J officinale* in MS basal media with 3 per cent sucrose supplemented with 17.76 µM benzyl adenine and 0.53 µM NAA. U-Kong et al (2012) reported that shoot tips were formed from shoot buds on modified MS medium supplemented with BA 4 mg/l (50%) and kinetin 1 mg/l of *J sambac* strains 1 (60%) and the results showed that 75 per cent shoot buds were induced from shoot tip explants when cultured on modified MS medium supplemented with BA 4 mg/l whereas 54.54 per cent shoot buds were induced when cultured on modified MS medium supplemented with kinetin 1 mg/l *J sambac* strain 2. Farzinebrahimi et al (2014) used young stems (each contained one nodal part) of *J sambac* for shoot development on MS medium supplemented with a combination of BAP (3.0 mg/l) and NAA (1 mg/l) that showed 20 per cent shoot regeneration. Biswal et al (2016) observed early bud break (25-26 days) in *J sambac* when MS medium was supplemented with BAP (2.0 mg/l), Kn (1.0 mg/l) and Ads (50 mg/l). Maximum percentage of bud break and multiple shoot formation (83.7 and 90.5) in *J sambac* was observed on MS medium having BAP (2.0 mg/l), Kn (1.0 mg/l), Ads (50 mg/l) and 3 per cent sucrose within 4 weeks of subculture. Salim (2016) stated that BA (2.0 mg/l) gave the best result in shoot number (2.5) and number of nodes per shoot (4.5 nodes/shoot) from nodal explants of *J azoricum.*

Callus culture and plant regeneration

Culture medium consisting of Murashige and Skoog (1962) medium supplemented with 2,4-D (2.0 mg/l), BAP (2.5 mg/l), NAA (0.01 mg/l) and TDZ (0.5 mg/l) significantly initiated the growth of callus in *J malbaricum*. Rajasekaran et al (2000) tried different explants of jasmine like leaf, stem and shoot tip to induce callus on MS medium supplemented with BAP and IAA. Maximum callus growth was observed from leaf explants in MS medium supplemented with 1 mg/l IAA and 10 mg/l BAP within 23 days of culture. Shoot development in jasmine was observed from leaf bit callus cultured on MS medium supplemented with 12 mg/l BAP. Similar results were reported by Gomathi et al (2007) in *J grandiflorum.* They found that leaf explants of *J grandiflorum* were ideal for callus induction. MS medium supplemented with 2,4-D (1.25 ppm) was found suitable for callus induction in leaf explants whereas Miller medium supplemented with BAP (1.25 ppm) and NAA (1.25 ppm) was suitable for callus induction in petal explants. Khoder et al (1979) obtained green callus from apical bud, petal, anther and internodes of *J officinale.* Complete plants were regenerated from stem pieces cultured on Millers's medium containing glucose, vitamins and growth regulators. Callus formation in leaf explants of *J nudiflorum* Lindl was favoured by 2,4-D (Zhong-min et al 2008). Zhehuang et al (2011) also obtained callus from petals of *J sambac* with some phytohormones at different concentrations. Biswal et al (2016) observed the maximum percentage of cultures showing callus proliferation from leaf explants of *J sambac* in the medium containing MS basal salts supplemented with 3.0 mg/l BAP and 2.0 mg/l IAA in leaf explants. The percentage of cultures showing callus was similar in the medium containing MS basal salts supplemented with 5.0 mg/l BAP and 2.0 mg/l NAA. However white and friable callus was obtained in the medium containing MS basal salts supplemented with 2.0 mg/l BAP, 2.0 mg/l Kn and 2.0 mg/l IAA from meristem explants. Davallo et al (2014) observed that highest callus diameter was observed in medium containing 0.3 mg/l 2,4-D. Maximum callus weight was obtained in 0.3 mg/l 2,4-D from leaf explants of *J sambac*. Farzinebrahimi et al (2014) observed that young stems of *J sambac* inoculated in MS medium supplemented with 2,4-D (0.5 mg/l) developed callus. Suryaningsih et al (2015) observed that essential oil (benzyl acetate and jasmine) content was highest in *J sambac* explant calli grown on MS + 20 per cent sucrose. The content of benzyl acetate reached 1.27 per cent and jasmone content reached 1.15 per cent in 12 weeks old calli. Salim (2016) reported that the highest percentage of callus induction was in MS supplemented with 4.0 mg/l BA + 0.1 mg/l 2,4-D and 6.0 mg/l BA + 0.1 mg/l 2,4-D and the highest number of buds (10.1) was recorded in the combination 4.0 mg/l BA + 2.0 mg/l Kin from inter-nodal explants of *J azoricum.* Bhattacharya and Bhattacharyya (2010) induced in vitro shoot regeneration in *J officinale* in MS medium supplemented with BAP (4.0 mg/l), NAA (0.1 mg/l) and sucrose 40 g/l. The elongation of regenerated shoots occurred when the cultures were transferred to MS medium supplemented with Kn (2.0 mg/l). U-Kong et al (2012) reported that modified MS medium supplemented with BA (4 mg/l) combination with NAA (0.1 mg/l) induced 2.8 shoots/explant in average and could form callus in 18 days while supplemented with kinetin 1 mg/l combination with NAA (0.1 mg/l) induced 3.2 shoots/explant in average and could form callus in

26 days of *J sambac* strain 1 and BA (4 mg/l) combination with NAA (0.1 mg/l) could induce 2.8 shoots of *J sambac* strain 2. Young leaves formed callus on modified MS medium supplemented with kinetin (1 mg/l) combination with 2,4-D (0.1 mg/l) supplemented with BA (1 mg/l) in 12 days of *J sambac* strain 1.

Root induction

The induction of root in the micropropagated shoots of *Jasminum* species was dependent on the composition of mineral nutrients and growth regulators. Cai et al (2007) induced rooting in micro-shoots of *J sambac* in half strength MS medium devoid of sugar and supplemented with NAA (1.2 mg/l). Sun et al (2009) adopted two step rooting method in jasmine. They cultured micro-shoots of jasmine in half strength WPM medium supplemented with NAA (0.1 mg/l) for 7 days. These micro-shoots were transferred to half strength WPM medium without any growth regulator supplement to obtain profuse rooting. He et al (2011) isolated the in vitro grown micro-shoots of *J sambac* and dipped them in NAA solution (450 mg/l) for 10 minutes. These NAA treated micro-shoots were sub-cultured in half-strength WPM medium for rooting. NAA was considered the most effective auxin in inducing roots in *J sambac.*

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