PROPAGATION OF MULBERRY, *Morus alba* THROUGH A SINGLE NODE CULTURE

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Abstract

A research study was undertaken to determine the requirement for initiation and multiplication rate of axillary buds of mature tree of Mulberry. Axillary buds derived from actively growing young shoots of 6 years old *Morus alba* were used to initiate the culture. Successful bud shoot induction and shoot elongation was achieved on MS media with 5μM BAP concentration. High multiplication rate i.e.12 micro shoots per culture was obtained with 15μM BAP. After 4-5 weeks the shoots developed roots in the same media without adding any rooting hormones. These plantlets showed a good growth response in earthen pots and 85 % survival of plants in soil was recorded.

Introduction

Mulberry a moderate sized deciduous tree species is planted almost in all the irrigated plantation of Punjab. This is a good tree species for reforestation projects because of its multiple uses. There is a great demand for this plant, which yields best quality wood for sports. The leaves are used for rearing silkworm, medicine and fodder for cattle, sheep & goats (Sheikh, 1993). Mulberry propagation is one of the important techniques for establishing plantation. There are two methods of propagation, viz. sexual (seedling) and asexual/vegetative (grafting, cuttings, layering, tissue culture, etc). Mulberry is genetically so heterogeneous that it is difficult to produce a pure line or an individual of the same characters as their parents through sexual propagation. Therefore, vegetation propagation is preferred (Singh and Sanatchandra, 2004). The present study was initiated to find out the in vitro requirements for initiation and maximizing its multiplication rate. This could be profitably utilized for clonal propagation of selected families as well as mass yield of mulberry leaves for silkworm rearing.

Materials and Methods

Experiment was conducted at Plant Propagation Laboratory, Punjab Forestry Research Institute (PFRI), Faisalabad. Axillary buds of *Morus alba* were collected from trees growing at PFRI Arboretum. These explants were surface sterilized with 1% commercial bleach solution for 25 minutes and then washed 5 times with sterile water to

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remove the possible traces of sodium hypochlorite in the bleach. These surface sterilized explants were inoculated on MS (Murashige and Skoog, 1962) basal media supplemented with different concentrations of BAP (Benzylaminopurine).

In general BAP has been frequently reported to induce better shoot multiplication than other cytokinins particularly in tree species (Ahmad, 1989). Rao and Lee (1982) have reported its effectiveness in juvenile as well as mature tissues of *Callophyllum inophyllum*, *Eugenia* spp. and *Fragraea fragrans*. Gupta, *et al*. (1981) and Zaman, *et al*. (1998) have reported its effectiveness in *Eucalyptus* spp and *M. alba,* respectively.

Culture Media

MS basal media containing 3 % sucrose and 0.9 % bacteriological agar was used with 5μM BAP for initiation of sprouting of buds. Multiplication of shoots was tried on MS media with different concentrations of BAP (Table 1). Due to the formation of rosette clumps of shoots, elongation of shoots was required. Elongation of shoots was tried on MS + 5μM BAP media. pH of all the media was adjusted to 5.8 before autoclaving.

No rooting hormones were required for the development of roots. Explants developed their roots in the same media within 4-5 weeks.

Culture Condition

Culture was maintained at 25-27^{0}C with 16 hours light and 8 hours dark period. Observations were recorded on percent culture responding to multiple shoot formation, average number of shoots per explant, length of shoots, percent shoots rooted and number of days taken for rooting, etc. Experiment was repeated 15 times and average data were calculated. The experiment was repeated 15 times and average data were calculated for each parameter.

Results and Discussion

After initiation of sprouting of buds on MS with 5 μM BAP, the multiplication of shoots started on MS media supplemented with different concentration of BAP. Shoot proliferation occurred within 25-30 days of incubation. Maximum multiplication rate was observed in shoots on medium containing 15 μM of BAP (Table 1).

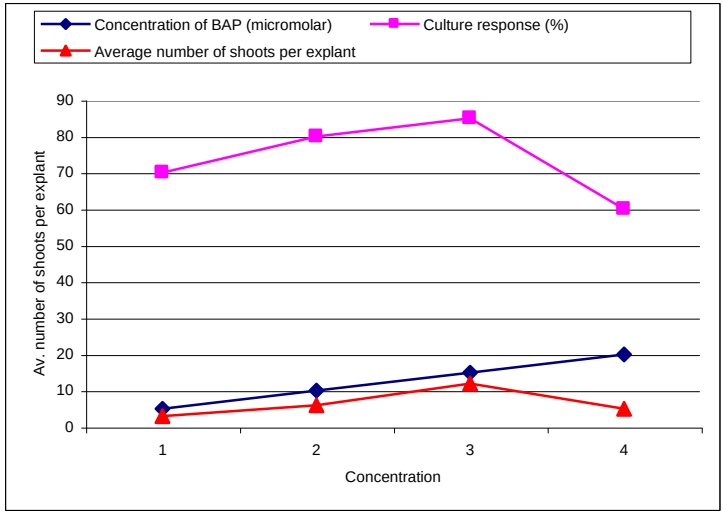
Table 1. Effect of BAP on multiple shoot formation

Concentration of BAP in MS media (μM)	Culture response (%)	Average number of shoots per explant (after 4 weeks)
5	70	3
10	80	6
15	85	12
20	60	5

Maximum 85% culture responded to the multiple shoot formation among the media combinations tested. Maximum average number of shoots developed was 12 after 4 weeks of inoculation on the media with 15 μM BAP.

The shoots were very small in size; hence these shoots were again kept on MS media with 5 μM BAP for elongation. Elongation of shoots was achieved within 30-35 days of incubation on elongation media.

Figure 1. Response of culture and shoots to BAP



Rooting and Acclimatization of rooted Shoots

When shoots attained the height of 4-5 cm, these were left in the same media for next 4-5 weeks and these developed their roots without adding any rooting hormones.

When the rooted shoots attained the height of 7-8 cm, these were transferred to earthen pots containing soil taken from tree growing area. These earthen pots were covered with polythene sheet to maintain humidity. Polythene sheet was gradually removed to acclimatize the plantlets. On the emergence of first pair of leaves, the polythene sheet was completely removed. The survival %age in pots was recorded as 85%.

Conclusions

It is concluded from the study that *M. alba* can be successfully propagated through axillary bud by in vitro techniques. So far as the concentration of BAP in MS media is concerned, 15 μM proved to be optimum for maximum multiplication rate and 5 μM was found best for elongation of shoot and no rooting hormones are required for root development.

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