

PROJECT PROFILE

Project Title: Development of macro and micropropagation techniques for *Melia dubia* Cav. for planting stock production

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Start and Completion dates : 2009-2011

Objectives:

1. Development of a macropropagation protocol for large scale production of planting stock.
2. Formulation of micropropagation techniques for continuous seedling production.

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Summary

The supply of fruits and seeds is abundant every year in *Melia dubia*, however, large scale planting is limited due to problems in germination. Therefore, this project examined the possibility of raising planting stock of *Melia* using vegetative methods and tissue culture. A survey was conducted and fruits of *Melia dubia* from twenty identified sources were collected to serve as explants for further studies. X ray and cut test were carried out to obtain information on seed filling studies. Different pretreatments were tested for enhancing germination. It was observed that the source of seeds and medium of germination was the major influencing factor. About 1000 seedlings from different sources were assembled. They were used for further experiments.

Micropropagation: Optimisation of sterilisation procedures for explants for contamination free cultures was carried out. It was observed that season played a significant role in the initiation of cultures. Experiments initiated during the period from January – June when there was a new flush of leaves, were contaminant free. Subcultured tissues revealed contamination after repeated subculturing, when initiated during the off seasons. This could possibly be due to endophytes. Nodal explants were excised from seedlings, thereafter planted on Murashige and Skoog's medium supplemented with various growth hormones for multiple shoot formation. Twenty different combinations were tested in two basal media. Multiple shoot formation occurred on MS supplemented with BAP (0.1 – 2 ppm) either alone or in combination with Kinetin (0.5 ppm). Best results, however, were obtained on MS + BAP (1 ppm) where 4-5 shoots regenerated from a single shoot apex. It was also observed that initial treatment with BAP (2 – 4 ppm) for 2 - 3

weeks followed by transfer to lower concentration of BAP (0.01 - 0.1 ppm) or simple MS medium proved most effective in terms of new shoot production and shoot growth rate. WPM medium was also tested but shoot proliferation was low. Vitrification of shoots was also observed. The shoots were excised and transferred to different root inducing media both *in vitro* and *ex vitro*. For *ex vitro*, micro cuttings of various sizes (2-3, 3-4, 4-5 cm) in varying concentrations of rooting hormones – 1000, 2000 and 3000 ppm IBA (liquid formulations) for 5 minutes and rooted in vermiculite. 75 % rooting was obtained; however, transplantation resulted in poor survival of plantlets.

Macropropagation: It involved determination of coppicing ability at different stump heights (30, 60, 90 and 120 cm), followed by rooting of these coppices. Also branch cuttings were included in the rooting experiment. Factors such as different hormonal treatments (control; IBA 1000, 2000, 3000 ppm – powder and liquid formulations), different media namely sand, vermiculite and coir pith were included in this experiment. Results indicated that 120 cm stumps gave the best survival percentage (60%) with the best coppice ability in terms of sprout number production (10). Though stumps of height 90 cm also produced sufficient coppice, the shoots did not survive. The difference observed in the coppice shoots arising from stumps of varied heights was diameter of the sprouts. Sprouts with diameter 0.5 cm or less (pencil thickness) showed poor rooting and survival ability. The rooting ability recorded was relatively low i.e. 16%, 18% and 11 % from 30, 60 and 90 cm stumps respectively. Sprouts from branch cuttings of pencil thickness diameter also rooted well, though the branch cuttings did not respond to hormonal treatments. They were also observed to be highly susceptible to fungal attack.