# TISSUE CULTURE OF FOREST TREES IN INDIA

A. F. MASCARENHAS and E. M. MURALIDHARAN
Division of Biochemical Sciences, National Chemical Laboratory, Poona 411 008, India.

## ABSTRACT

Tissue culture studies carried out in India on forest tree species are reviewed and presented in a tabulated form. Many of these studies are directed at large-scale micropropagation of economically important trees yielding fuelwood, pulpwood or timber. Plantlet regeneration has been achieved in several species, but commercially viable technologies have been developed only in a few instances.

## INTRODUCTION

The tree wealth of India is unrivalled in its diversity of species and of utility. Forests of various types cover the subcontinent ranging from coniferous in the Himalayas to lush tropical forests in the south. Trees like teak, sandalwood and rosewood have enjoyed world-wide reputation since ancient times. The pressures of a growing population in recent decades has however resulted in the exploitation of the forests to disastrous proportions. The increasing demands for biomass for fuelwood, timber and the pulp and paper industry can no longer be met from the existing natural forests alone. Large-scale plantation forestry and afforestation of degraded forest lands is the solution to ensure future sufficiency in biomass.

Several issues need to be examined if trees are to be treated as crops for biomass production. The choice of tree species would depend on the end uses and the agroclimate of the area to be planted. Fast-growing, short-rotation species with good pulping characteristics or calorific value are selected for paper-pulp or firewood. Several million hectares of land all over India lie waste because of degraded soil, extreme salinity or aridity. By a suitable choice of stress-tolerant species, waste land could be reclaimed for afforestation.

Forest tree improvement by selection and breeding began in India only recently[1]. Traditional breeding methods in trees are limited by their large size and long life-cycles. Clonal forestry is now gaining increasing recognition as a quicker alternative in tree improvement[2]. *In vitro* techniques are being increasingly applied to supplement conventional methods of vegetative propagation. The benefits of this technique include high multiplication rates, generation of disease-free stock and stress-tolerant variants, and long-term storage of valuable germplasm.

Tissue culture of tree species in India began only in the 1960s. At the University of Delhi, callus cultures of *Pinus* were established and studied by Konar[3]. The embryos and endosperm of *Santalum* were studied in culture by Rangaswamy and Rao[4]. Later research on *Santalum* led to a highly successful system for regeneration through somatic embryogenesis and protoplasts. During the late 1960s tissue culture studies on tree species were initiated at the National Chemical Laboratory, Pune[5].

Research in tissue culture of tree species is now carried out in several research institutes and universities. The entry of private commercial firms into this area is only recent and these have yet to make an impact.

In this review work on tissue culture of forest tree species carried out in India is given in the form of a table (table 1). Unpublished work is also included wherever the information was available to the authors. Fruit trees have been excluded from this review.

## CONCLUSION

As table 1 indicates, most of the forest trees which have been cultured are economically important as sources of timber, pulpwood or firewood. Many trees, especially the tree legumes, are multi-purpose trees suitable for rural agroforestry. Research work in a majority of cases is directed towards micropropagation, from juvenile tissue, seedlings and mature trees.

Although plantlet regeneration has been reported in a large number of tree species only a few have

**Table 1** *Work on tissue culture of forest trees in India*

| Species | Explant | Result, studies | Reference |
|---|---|---|---|
| **Angiosperms** | | | |
| *Acacia auriculiformis* | Seedling node | Budbreak, plantlet | Gupta, S. C. (unpublished) |
| *A. nilotica* | Mature stem | Adventitious (advt.) buds, plantlets | 8 |
| | Cotyledons | Embryogenesis | Mascarenhas, A. F. (unpublished) |
| *A. senegal* | Seedling explants | Budbreak, plantlets | 9 |
| *Ailanthus malabarica* | Nodes | Budbreak | D'Souza (unpublished) |
| *Albizzia amara* | Hypocotyl | Callus, advt. buds, plantlets, embryogenesis | 10 |
| *A. lebbeck* | Hypocotyl | Embryogenesis, plantlets | 11 |
| | Seedling explants | Advt. buds, plantlets in soil | 12 |
| | | Callus, advt. buds, plantlets in soil | 13 |
| | Anthers | Plantlet (haploid) | 14 |
| | | Callus | 15 |
| *A. procera* | Seedling explants | Callus, advt. buds, plantlets | 16 |
| | | Callus, embryogenesis, plantlets | 17 |
| *A. richardiana* | Seedling explants | Callus, advt. buds, plantlets | 10 |
| *Azadirachta indica* | Embryo | Callus, advt. buds, plantlets | 18 |
| | Mature nodes | Callus, advt. buds, plantlets | 19 |
| | Mature leaflets | Callus, advt. buds, plantlets | 20 |
| | Stem | Callus, biochemical studies | 21 |
| | Cotyledons | Advt. buds, plantlets | 6 |
| *Bambusa arundinacea* | Seed | Callus, embryogenesis, plantlets | 22 |
| *B. vulgaris* | Mature nodes | Budbreak | 23 |
| *Bauhinia purpurea* | Seedling nodes | Budbreak, plantlets | 16 |
| *Butea monosperma* | Nodes | Budbreak | D'Souza (unpublished) |
| | Pollen | Callus | 24 |
| *Cassia fistula* | Anthers | Callus, embryogenesis | 25 |
| *C. siamea* | Anthers | Callus, embryogenesis, plantlets | 26 |
| *Ceratonia siliqua* | Mature nodes | Callus, advt. buds, plantlets | 27 |
| *Commiphora wightii* | Leaf, stem | Callus, suspension | 28 |
| *Crataeva nurvala* | Leaf | Advt. buds, plantlets | 29 |
| *Dalbergia lanceolaria* | Seedling explants | Callus, advt. buds, plantlets | 30 |
| *D. latifolia* | Seedling explants | Callus, advt. buds, plantlets | 31 |
| | | budbreak, plantlets | 32 |
| | Roots, nodes (5-yr tree) | Advt. buds, plantlets in soil | 33 |
| | Mature stem | Callus, advt. buds, plantlets | 34 |
| *D. sissoo* | Seedling, roots | Advt. buds, plantlets | 35 |
| | Hypocotyl | Callus, advt. buds | 36 |
| | Mature nodes | Budbreak, plantlets | 37 |
| *Delonix elata* | Leaf | Callus, advt. buds | 38 |
| *Dendrocalamus strictus* | Seedling nodes | Budbreak, plantlets in soil and field | 23 |
| | Seeds | Embryogenesis, plantlets in soil | 39 |
| | Mature nodes | Budbreak, plantlets | 23 |
| *Duboisia myoporoides* | Mature nodes | Budbreak, plantlets | 40 |
| | Leaves | Advt. buds, plantlets | 41 |
| | | suspension, plantlets | 42 |
| *Erythrina indica* | Seedling explants | Budbreak, callus, plantlets | David, S. B. (unpublished) |

**Table 1.** (*Contd.*)

| Species | Explant | Result, studies | Reference |
|---|---|---|---|
| *Eucalyptus camaldulensis* | Mature nodes | Budbreak, plantlets in soil and field | 43 |
| | Mature leaves from *in vitro* shoots | Callus, advt. buds, plantlets in soil | 44 |
| *E. citriodora* | Lignotuber | Callus, plantlets | 45 |
| | Seedling nodes | Budbreak, plantlets | 46, 47 |
| | Cotyledons | Callus, advt. buds, plantlets | 48 |
| | Shoot tip | Budbreak | 49 |
| | Mature nodes | Budbreak, plantlets in soil and field | 50 |
| | Seed | Embryogenesis, plantlet in soil, artificial seeds | 44, 6 |
| | Mature leaf | Callus, estimation of oil | 51 |
| *E. globulus* | Mature nodes | Budbreak, plantlets | 33 |
| *E. grandis* | Mature nodes | Budbreak, plantlets in soil | 52, 53 |
| *E. tereticornis* | Mature nodes | Budbreak, plantlets in soil and field | 33 |
| | Leaves | Callus, biochemical studies | 54 |
| *E. torelliana* | Mature nodes | Budbreak, plantlets in soil and field | 43 |
| *Ficus religiosa* | Mature stem explants | Callus, advt. buds, plantlets in soil | 55 |
| *Holarrena antidysenterica* | Mature nodes | Budbreak, plantlets | 56 |
| | Hypocotyl | Callus, secondary-metabolite production | 57, 58 |
| *Jacaranda acutifolia* | Anthers | Callus, embryogenesis | 26 |
| *Lagestroemia flos-reginae* | Mature nodes | Budbreak, plantlets in soil | 59 |
| *Leucaena leucocephala* | Mature nodes | Budbreak, plantlets in soil | 60, 61, 62, 63, 64 |
| | | *In vitro* nodulation | 65 |
| *Leucosceptrum canum* | Mature leaves | Callus, advt. buds, embryogenesis, plantlets in soil | 66 |
| | Mature nodes | Budbreak, plantlets | 67 |
| *Melia azadirachta* | Embryo | *In vitro* flowering | Mascarenhas, A. F. (unpublished) |
| *Mitragyna parviflora* | Shoots, roots | Long-term preservation | 68 |
| | Mature nodes | Budbreak, plantlets | 69 |
| *Morus alba* | Mature stem | Callus, biochemical studies | 5 |
| | Seedling nodes | Budbreak, plantlets | 16 |
| *M. indica* | Mature nodes | Budbreak, plantlets in soil | 70 |
| | Mature leaf | Callus, advt. buds, plantlets in soil | 70 |
| | Mature nodes | Plantlets, *in vitro* fruiting | 71 |
| | Mature buds | Encapsulated buds, plantlets | 72 |
| *Peltophorum pterocarpum* | Anthers | Callus, plantlets (haploid) | 73 |
| *Poinciana regia* | Anthers | Callus, embryogenesis | 26 |
| *Populus alba* | Mature nodes | Budbreak | 74 |
| *P. ciliata* | Mature stem explants | Callus, suspension, embryogenesis, plantlet in soil | 75 |
| | Mature leaf | Advt. buds, plantlets in soil | 74, 76 |
| | Mature buds | Callus, advt. buds, plantlets in soil | 76 |
| *P. nigra* | Mature stem explants | Callus, advt. buds | 77 |
| *P. deltoides* × *P. nigra* | Leaves, petioles | Callus, advt. buds, plantlets | 78 |
| *Prosopis cineraria* | Hypocotyl | Advt. buds, plantlets | 79 |
| | Mature nodes | Budbreak, plantlets in soil | 80 |
| | Gall | Callus | 81 |
| *Pterocarpus santalinus* | Seedling explants | Budbreak, callus | Mascarenhas, A. F. (unpublished) |
| *Putranjiva roxburghii* | Endosperm | Callus, plantlets (triploid) | 82 |

**Table 1.** (*Contd.*)

| Species | Explant | Result, studies | Reference |
|---|---|---|---|
| *Salix babylonica* | Mature buds | *In vitro* flowering | 83 |
| | Mature buds | Budbreak, plantlets | 84, 85 |
| *S. tetrasperma* | Mature buds | *In vitro* flowering | 84, 86 |
| *Salvadora persica* | Mature nodes | Budbreak, plantlets in soil and field | 87 |
| | Stem gall | Callus | 88 |
| *Santalum album* | Endosperm | Callus | 4 |
| | | Advt. buds, plantlets (triploid) in soil and field | 89 |
| | Hypocotyl | Advt. buds, plantlets | 90 |
| | | Callus, embryogenesis, plantlets in soil and field | 91 |
| | Suspension | Embryogenesis, plantlets | 92 |
| | | Protoplasts, callus, embryogenesis, plantlets | 93 |
| | Mature stem, hypocotyl | Protoplasts, callus, embryogenesis, plantlets | 94 |
| | Mature shoot tip | Callus, embryogenesis, plantlets in field | 95 |
| *Sapindus trifoliatus* | Mature leaf | Callus, embryogenesis, plantlets | 96 |
| *Sapium sebiferum* | Mature nodes | Budbreak, plantlets in soil and field | 97 |
| *Sesbania grandiflora* | Hypocotyl, cotyledon | Callus, advt. buds, plantlets | 98 |
| *Shorea robusta* | Shoots | Long-term preservation | 68 |
| *Solanum grandiflorum* | Anthers | Callus, embryogenesis | 99 |
| *Syzygium travancorium* | Shoots | Long-term preservation | Mascarenhas, A. F. (unpublished) |
| *Tamarindus indica* | Seedling explants | Budbreak, plantlets in soil and field | 100 |
| | Mature nodes | Budbreak, plantlets in soil | 101 |
| | Anthers | Callus, embryogenesis | 15 |
| *Tectona grandis* | Mature stem explants | Callus, biochemical studies | 5 |
| | Seedling, mature nodes | Budbreak, plantlets in soil and field | 102 |
| | Mature shoots | Callus, embryogenesis | 103 |
| | Endosperm | Callus | 104 |
| | Shoots, roots | Long-term preservation | 68 |
| *Tecomella undulata* | Mature buds | Budbreak, plantlets | 105 |
| *Terminalia bellerica* | Nodes | Budbreak, callus, plantlets | 106 |
| *Vateria macrocarpa* | Shoots | Long-term preservation | Mascarenhas, A. F. (unpublished) |
| *Zizyphus xylopyra* | Nodes | Budbreak, plantlets | 106 |
| **Gymnosperms** | | | |
| *Biota orientalis* | Embryo | Embryogenesis | 107 |
| *Cedrus deodara* | Embryo, female gametophyte | Callus, advt. buds | 108 |
| *Cryptomeria japonica* | Cotyledons | Callus, cytological studies | 109 |
| *Picea smithiana* | Cotyledons | Callus, advt. buds | 74,110 |
| | | Cytological studies | 74,110 |
| *Pinus gerardiana* | Hypocotyl | Callus, advt. buds | 3, 111, 112 |
| | | Callus, cytological studies | 113 |
| *P. roxburghii* | Embryo | Callus, advt. buds | 28 |
| | Seedling explants | Callus, cytological studies | 114 |
| *P. wallichiana* | Embryo | Callus, advt. buds, plantlets | 115 |

been successfully transferred to soil and established in the field. This step is essential for the technique to be applicable for large-scale micropropagation and for confirmation of clonal fidelity and economic feasibility.

There has been an increasing interest in recent years in commercializing *in vitro* propagation of forest trees, especially by the pulp and paper industries. Commercialization will bring about refinements in the existing procedures to make these more cost-effective. The development of automated culture procedures and plant delivery systems using somatic embryos and artificial seeds is also being investigated. The production of artificial seeds by encapsulation of somatic embryos in *Eucalyptus*[6] marks a beginning in this direction.

The recent demonstration of the possibility of genetic transformation and *in vitro* regeneration in conifers[7] points to the potential application of these methods to the improvement of trees in general in the near future. While *in vitro* propagation methods have been successful in an increasing number of tree species, the realization of the benefits of these procedures would depend on the implementation of comprehensive tree improvement programmes. The opportunities for forest tree biotechnology through improved productivity is tremendous in a country like India with expanding biomass requirements.

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# ANNOUNCEMENTS

## BIOSENSORS '89

(28 and 29 September 1989, St John's College, Cambridge, UK)

The realization that the unique recognition and catalytic capabilities of biological systems can be coupled with electrochemical or optical transducers has spawned a new branch of analytic science—biosensors. This emerging technology is likely to have a profound effect on the way analytical procedures are performed. Recent developments in the technology and its applications will be discussed. For details contact: Dr Renata Duke, IBC Technical Services Ltd, Bath House (3rd Floor) 56 Holborn Viaduct, London EC1A 2EX, UK.

## SIXTH EUROPEAN SEMINAR AND EXHIBITION ON COMPUTER-AIDED MOLECULAR DESIGN

(5 and 6 October 1989, London Press Centre, UK)

The meeting and accompanying exhibition will provide a forum for discussion and display of the latest novelties in both hardware and software, demonstrating that rational design of molecules has become a reality. For details contact: Dr Renata Duke, IBC Technical Services Ltd, Bath House (3rd Floor), 56 Holborn Viaduct, London EC1A 2EX, UK.