area immediately below the low tide. According to their frequent occurrence in stratified limestones at the lower flank of Jurassic bioherms in southern Germany they possibly lived in water as deep as 70 m. How far they extend into deeper water is not known. In the light of the above-mentioned points it could be summarized that, the association of the oncolite-bearing olistoliths and other *in situ* and reworked fossils bearing olistoliths of the Dras and the Lamayuru formations indicate that, before the commencement of convergence, i.e. during the tectonically stable regime, the carbonate sequence was deposited at a shallow marine condition. In the Upper Cretaceous time the convergence of the Indian Plate beneath the Eurasian led to the compression of the shallow marine sediments of shelf, reef and basin margin (slope) environments. Ongoing compression of the shelf sediments led to the development of minor shelf and ramp for a shorter duration, resulted in the formation of small, isolated shallow marine basins all along the Tethyan platform margin, which enhances to the amount of reworked sediments. The process is concomitently associated with the Tethyan sedimentation which is finally ceased during the Eocene time (45 Ma) when the final collision between the Indian and Eurasian plate took place. In the tectonically active phase of subduction, i.e. during subduction accretion[15], the collapse of the Zanskar shelf margin (Tethyan zone) initiated the process of sliding down of these shallow marine sediments towards deeper levels. These sediments were ultimately kneaded[15] with the sediments of the Dras and the Lamayuru formations which were being deposited at that time at the deeper levels. The main emphasis here is to highlight the occurrence of different olistoliths of limestone and the discovery of oncolite structures in the NW sector of the Indus–Tsangpo Suture Zone of Ladakh Himalaya. The study is significant in the study of Cenozoic continent to continent tectonics and building of the Himalayan mountain.

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Plantlet production from shoot tip cultures of red sandalwood (*Pterocarpus santalinus* L.)

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Red sandalwood (*Pterocarpus santalinus* L.), belonging to the family Fabaceae, is one of the most valuable trees, and has limited distribution in India. In view of its high price, restricted distribution and usefulness as a timber tree, there is urgent need to obtain improved lines, in both quality and quantity. We have established a method for production of complete plantlets by tissue culture. We report here the successful development of red sandalwood plantlets by induction of multiple shoots from shoot tips, and successful transfer of micropropagated plants to soil.

RED sandalwood is restricted to certain forest tracts in Andhra Pradesh and Tamil Nadu in South India. In view of its value, in countries like Japan, its trade has become much more significant than what it was a few decades ago. The red colouring principle santalin is valued highly as a natural dye. The wavy and rippled grain wood is preferred to that of straight grained wood. In view of its high price, restricted distribution and usefulness as a timber tree, there is an urgent need to obtain improved lines, in both quality and quantity.

Earlier studies using conventional clonal propagation methods like grafting and rooted cuttings have not been very successful. Importance of plant tissue culture for successful mass propagation of forest tress like eucalyptus[1], sandalwood[2] and teak wood[3] has already been demonstrated. Forest trees in general and Fabaceae in particular have proved to be difficult to mass propagate by tissue culture. So far there are very few reports of timber trees of Fabaceae successfully established by tissue culture. Rosewood[4] is the only timber tree of this family successfully established by tissue culture. However, tissue culture propagation of a few fast-growing fuelwood trees like leucaena[5,6], sesbania[7], albizia[8,9] and acacia[10], belonging to this family, have been reported, both by the method of organogenesis and by multiple shoot production of

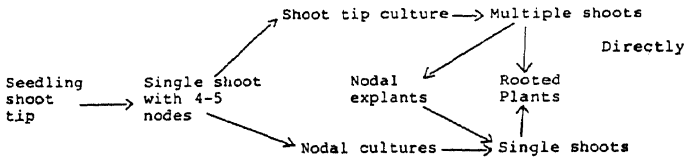
axillary meristem of seedling explants. Red sandalwood, like rosewood, is equally important as a timber tree and is described as an underexploited luxury wood by the advisory committee on technology innovation (NRC 1979)[11].

Gamborgs' medium (B5)[12] containing 2% sucrose and 0.8% agar was used as basal medium throughout the present study, unless otherwise stated. The medium was adjusted to pH 5.6–5.8 before autoclaving at 121°C for 20 min. Corning testtubes containing 15 ml medium were used with cotton plugs covered in cloth. All cultures were incubated at 25°C in a photoperiod of 16 h per day of fluorescent light (about 1200 lux). The experiments were repeated at least thrice for all cultures.

Shoot tip cultures were established from germinating seedlings. Seeds were soaked and dried alternatively for 15 days prior to placing them in seed beds for successful germination. Shoot tips obtained from 15-day-old seedlings were initially washed in running tap-water followed by soap solution and water thoroughly to remove soil and other contaminants. This was followed by 15-min treatment with 0.1% mercuric chloride solution and then rinsed with sterile-distilled water three to four times before inoculation. Shoot tips were cultured on B_5 basal medium supplemented with 0.2 mg/l benzylaminopurine (BA) and 0.1 mg/l kinetin, where they elongated into shoots measuring 4–5 cm with four or five nodes. Explants, both shoot tip and nodal cuttings, were taken from these sterile shoots for all the following experiments. Shoot tips measuring about 5–8 mm and nodal explants with single axillary bud were cultured separately on B5 medium supplemented with the cytokinins BA and kinetin at concentration ranging from 0.5 to 5 mg/l individually or in combination with other cytokinins or an auxin, mostly naphthaleneacetic acid (NAA) (0.5 mg/l).

For the development of single shoots from seedling shoot tip with four to six nodes, basal medium alone was found to be inadequate. Culturing of shoot tip on B5-medium supplemented with 0.2 mg/l BA and 0.1 mg/l kinetin was found to be necessary for single shoot growth. Healthy shoots with well-developed leaves were obtained by this method in about five to six weeks. Shoot tips cultured on B5 medium supplemented with single cytokinin did not give more than 2–3 shoots. Hence combinations of two cytokinins were tried. Among the various combinations on B5 media supplemented with 1 mg/l BA along with 1 mg/l kinetin multiple shoots, up to eight, were obtained in four to six weeks. These shoots were measuring nearly 3–5 cm (Figure 1). Multiple shoots obtained by this method were divided into 2–3 clumps for further proliferation to increase the number of shoots. These clumps, in turn, proliferated into multiple shoots. For continuous production these shoots are subcultured every 4–6 weeks. The vigour of the multiple shoots has declined after 5–6 subcultures. Shoots can also be rooted on rooting media and subsequently plantlets can be developed. For root development, MS medium[13] was supplemented with indoleacetic acid (IAA), NAA or indolebutyric acid (IBA) in concentrations ranging from 1 to 5 mg/l. Among these shoots rooted on MS medium supplemented with IAA were better than the other two. Even with IAA, at high concentration of 5 mg/l, a single thick root induction was observed, but at low concentrations (1–2 mg/l) adventitious roots were obtained. Nearly 80% of the shoots have rooted on IAA medium compared to IBA where it was 30–40%. In addition to the shoot tip cultures from seedling shoots, nodal explants with single axillary buds were also cultured to induce single or multiple shoots. From these explants only single shoots were obtained. The flow chart shows the sequence of cultures starting from the seedlings.

Figure 1. Multiple shoots from seedling shoot tip cultured on B5 medium supplemented with 1 mg/l BA + 1 mg/l kinetin and 200 mg/l adenine sulphate.



Rooted shoots were established by gradual acclimatization in sterile soil mixtures of 1:1 soil and sand (Figures 2 and 3). Establishment of plants was nearly 50%. The percentage could be further improved if sophisticated growth chambers are available. Acclimatization was done by transferring the rooted plants into liquid medium of the same composition for hardening; after two weeks these were transferred into test tubes containing only water and covered with Parafilm for a week prior to potting. Even the pots were covered with plastic cover to keep the humidity for two weeks, before keeping them in shaded areas for protection. By this method 60% survived. Loss of plants was mainly due to necrosis. About 20 plants were established by this method.

In a previous study[14] in red sandalwood, in which a single cytokinin (BA) was used, resultant shoots showed scaly leaves. In the present investigation, both single shoots on low cytokinins (0.2 mg/l BA + 0.1 mg/l kinetin) and multiple shoots obtained on 1 mg/l BA + 1 mg/l kinetin showed well-expanded normal leaves.

Figure 2. Single shoot rooted on MS + 5 mg/l IAA.

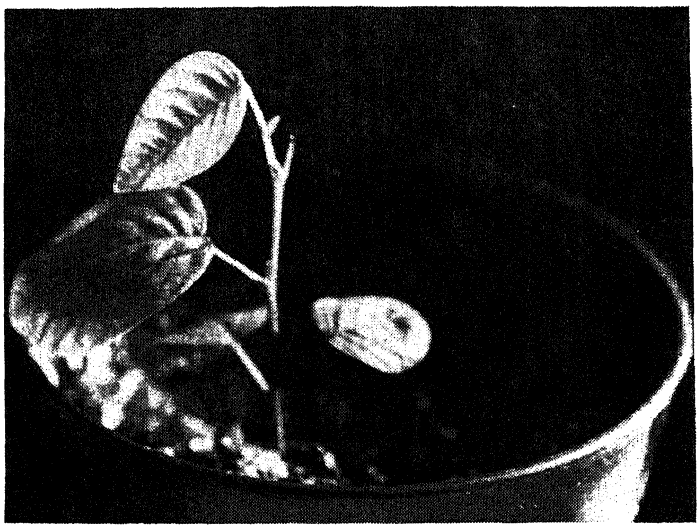


Figure 3. Established plant after one month.

Minor variations of the above concentrations also showed normal leaf development. Combination of two cytokinins was found to be better than the use of a single cytokinin. Comparative studies with other tree legumes showed that there is no precocious leaf drop as reported in leucaena[5] and rosewood[4]. Addition of adenine sulphate to the multiple shoot medium has shown improved vigour of the cultures. Experiments conducted with shoot medium has show improved vigour of the cultures. Experiments conducted with shoot tips or nodal explants from field-grown mature trees from the forest areas have not so far been successful. Even the establishment of the cultures was not achieved successfully. This could be due to problems arising while transporting the material from the forest areas, which usually resulted in contamination. The method of multiplication as reported here shows great potential. Multiplication from seedling explants may not be desirable unless it is a precious hybrid seed where planting material is limited. However, the technique developed in the present study can be adapted for the multiplication of mature trees once the initial cultures are established. Also, multiplication by multiple shoot methods from shoot tip or axillary meristems will be more desirable than by the method of morphogenesis from callus cultures, where variations can come due to changes in the ploidy level. These studies demonstrate the potential for mass multiplication of this species.

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Vincristine can cause giant cell formation in rat testis

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While it is known that several cancer chemotherapeutic regimens containing the *Vinca* alkaloid vincristine bring about abnormalities in male reproduction, like azoospermia, gynaecomastia, etc., the precise mode of toxicity of vincristine is not known. We administered vincristine sulphate to sexually mature Wistar strain male albino rats, and show that vincristine causes absence of sperm in seminiferous tubules, depletion of germinal epithelial elements, and formation of hypertypic giant cells. We discuss the origin of the giant cells in the light of the antimitotic activity of vincristine, and suggest that vincristine affects spermatogonial mitosis, consequent upon which there is endomitosis, resulting in giant cells.

COMBINATION chemotherapy is one of the most advocated therapeutics for cancer. MOPP (mustine, oncovin, procarbazine and prednisone)[1], COPP (cyclophosphamide, oncovin, procarbazine and prednisone)[2], MOMP (mustine, oncovin, methotrexate and prednisone)[3], CVP (cyclophosphamide, vincristine and prednisone)[4], MVPP (mustine, vinblastine, procarbazine and prednisone)[5], CMM (cyclophosphamide, methotrexate and mercaptopurine)[6] and MCCB (mustine, cyclophosphamide, chlorambucil and busulphan)[7], are examples of combination cancer chemotherapeutic regimens. Most of these drugs are by nature cytotoxic; they are either alkylating agents, antimetabolites, antibiotics or mitotic spindle poisons[8]. There have also been reports implicating these regimens in several side-effects like nausea and vomiting, leukopenia, alopecia, stomatitis, peripheral neuropathy, cardiopathy, hepatocellular damage and pulmonary fibrosis[9]. Male gonadal dysfunction including azoospermia, oligospermia, gynaecomastia and germinal aplasia, probably culminating in male sterility[3,10–14], has also been reported.

Vincristine is advocated as one of the drugs in combination chemotherapy[1–4]. It is established as a mitotic-spindle poison and is believed to prevent cancer growth by arresting mitotic-spindle formation through prevention of tubulin polymerization and disruption of microtubules. Use of such spindle poisons in cancer chemotherapy is also likely to affect other dividing cells, including those connected with spermatogenesis[15]. While alkylating agents, antimetabolites and antibiotics—chlorambucil[16], cyclophosphamide[17], prednisone, methotrexate, 5-fluorouracil, mitomycin C, actinomycin D, procarbazine[18] and cisplatin[19]—, when administered in cancer chemotherapy, have been shown to lead to testicular atrophy, azoospermia, germinal aplasia and sterility, the specific gonadal toxicity of spindle poisons like vincristine has not yet been studied. On the other hand, administration of total alkaloids of *Vinca rosea* (*Catharanthus roseus*) (West Indian periwinkle, Apocynaceae) to adult male rats and mice has been shown to bring about arrest of spermatogenesis, regression of Leydig cells, and derangements in sperm[20–24]. Vincristine being one of the binary indole-indolin alkaloids isolated from this plant, it is highly probable that this drug, when used in cancer treatment, might cause testicular derangements. In this paper we report that vincristine causes giant-cell formation in seminiferous tubules of rat.

We examined stained sections of testes from rats that had been given intraperitoneal injections of vincristine sulphate (see Figure 1). The typical histoarchitecture of seminiferous tubules of normal rats show cells arranged in spermatogenic sequence (Figure 1,*a*). In rats treated with 10 or 20 μg (per animal) of vincristine, the seminiferous tubules were thoroughly disorganized, highly regressed, and contained far fewer layers of cells (Figure 2,*b*). Meiotic elements were never seen in the cells. Most of the tubules contained hypertypic giant cells of different sizes along the border of the widened lumen as well as lying free in the lumen (Figure 1,*c*). There were a few tubules in which the size of the germinal epithelial cells increased towards the luminal border (Figure 1,*d*), in contrast to the decrease in size of