BIOLOGY OF THE SEEDLING OF SANDAL WOOD

(Santalum album Linn.)

By

YAYATI B. TAIDE

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Dedicated

То

My Parents,

Dr. B.G. Taide,

Dr. (Mrs.) B.B. Taide

whose love and blessing have been a source of inspiration, courage and stamina to undertake studies in Master of Science in Forestry in persuit of knowledge. I owe them for what I am to day and even hope to be in future.

DECLARATION

I hereby declare that this thesis entitled "Biology of the seedling of sandal wood (<u>Santalam album Linn.</u>)" is a bonafide record of research work done by me during the course of research and that the thesis has not previously formed the basis for the award to me of any degree, diploma, associateship, fellowship, or other similar title, of any other University or Society.

Yayati Taide)

Place: Vellanikkara Date : // -//- 9/

CERTIFICATE

Certified that this thesis, entitled "Biology of the seedling of Sandal Wood (Santalum album Linn.)" is a record of research work done independently by Shri. Yayati. B. Taide under my guidance and supervision and that it has not previously formed the basis for the award of any degree, fellowship or associateship to him.

Place: Vellanikkara Date : l = l - l + 99l

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Dr. Luckins.C.Babu, Associate Professor and Chairman, Advisory Committee, College of Forestry, Kerala Agricultural University. Approved By :

Chairman

South Chy

Dr. Luckins.C.Babu, Associate Professor, College of Forestry.

Members

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Dr. C.C.Abraham, Special Officer, College of Forestry.

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External Examiner

Dr. B. Mohan Kumar, Associate Professor, College of Forestry.

Dr. T.V. Viswanatham, Associate Professor, College of Horticulture.

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Yayati Taide

Introduction

1. INTRODUCTION

sandalwood tree, botanically known as Santalum album, The a member of the family Santalaceae. The tree is is Linn. economically a very important forest species in India and is the source of the world famous East Indian sandalwood oil. It is small to medium sized, evergreen tree growing to a height up to 10 to 15 m and 2-4 m in girth (Parthasarathi and Rai, 1989) and is native to India (Dymock et al., 1892; Arctander, 1960; Troupe, limited Ramaswami, 1956 b). The species has а 1909, 1921; distribution in the world, confined to India and Indonesia. Τn India, it is commonly found in the comparatively dry regions of peninsular India from Vindhya mountains southwards, especially in Karnataka and Tamil Nadu states. The tree flourishes well the from sea level up to 1800 m altitude in different soil types and climates except in highly alkaline, water logged or very cold The tree has been introduced in Rajasthan, parts of places. Uttar Pradesh, Madhya Pradesh and Orissa.

The heart wood of <u>Santalum album</u>, Linn. is commercially known as East Indian sandalwood and the essential oil form it as East Indian sandalwood oil. Both wood and oil are used in incenses, perfumes, medicines and are of great commercial value. In old Ayurvedic literature sandalwood has been described as bitter, cooling, astringent, and useful in biliousness, vomiting, fever, thirst and heat of the body (Watt, 1893). Ninety per cent of the world annual demand of sandalwood oil is met by India. India enjoys a virtual monopoly on this oil trade, earning over Rupees Ten crores in foreign exchange (Husain and Punnuswamy, 1982). Sandalwood oil is a real vegetable gold, and it deserves the attention of foresters, botanists, entomologists and industrialists for its protection, preservation and promotion, for it is an asset and an earner of foreign exchange.

Late in the last century, sandal was heavily exploited for commercial purposes. This necessitated its regeneration to Attempts were made to raise sandal future supply. ensure plantations as far back in 1870 or even earlier. But majority of these cases, failed and the main reason for the failure was the ignorance of the parasitic nature of sandal. Root parasitic nature of sandal was reported as early as in 19th century in India. The association of root of sandal with other plant species came to light during the course of investigation on "spike", (a very common disease affecting leaves of sandal) by foresters. The absence of root hairs in sandal and the constant assocation of roots of sandal with other species are known as haustoria have led to the conclusion that nutrients are passed from host roots to those of sandal. Doubts were expressed by foresters as to whether the entire food requirement of a big sandal tree could be

from the host-parasite association. Many researchers were met sandal also sceptical about the parasitic nature of (Iyanger, Barber (1906) conducted a detailed examination on the 1965). rootlet and haustoria. He stressed the fact that sandal derives its nutrient only from the hosts through the haustoria and in no other manner. Presence of large number of unattached haustoria in many sandal trees led many to doubt its parasitic nature. Further scope of investigation on the parasitic nature of sandal has been indicated by Barber.

Later it was established that sandal is both a autophyte and a root parasite. Barber (1906) clearly showed that sandal derives nitrogen and phosphorous from the host and lime and potash from the soil through root tips.

In the available literature, there is a long and exhaustive list of sandal hosts but the information regarding the best host parasite relation however is lacking. Sandal depends mainly on host for its vigorous growth, heart wood formation and its resistance to diseases and insects. Identification of the best host parasitic relationship might help in increasing the quality and quantity of sandal wood and sandal oil in the long run. With objective an experiment was carried out to identify and this recommend the choice of the host species to be preferred while raising sandal plantations, particularly in Kerala.

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The present study was undertaken with the following objectives:

To identify the best sandal - host combinations,
To study seedling growth characteristics,

3. To study the anatomical structure of haustoria.

Review of Literature

2. REVIEW OF LITERATURE

Ninety per cent of the world's demand of sandal wood oil is met by India (Husain and Punnuswamy, 1982). This demand has been ever increasing. To meet this escalating demand the cultivation of sandal is to be extended into unconventional sandal growing areas (Kushalappa, 1983). The major problem in the expansion of sandal cultivation is, the fact that, sandal seedlings are incapable of growing beyond a year unless they develop haustoria with host plants, through which they meet their nutritional An attempt is made to requirements (Rao and Sahib, 1911). literature Morphology, briefly the available on review Physiology, Chemistry and utilization of sandal.

1. Morphology and Physiology

1.1. Parasitism of sandal

Sandal is a hemi-root parasite drawing nutrition from the host plants through haustorial connections. The parasitic nature of sandal was first noticed by Scott (1871). Later independent studies by Barber (1903), Rao (1903) and Rao (1942) revealed the presence of haustoria in sandal roots. Barber (1905) noted that haustoria formation occurred only on certain roots of sandal and not on all of them. A detailed description of the structure of the haustoria was also given by Barber (1906, 1907). He noted that formation of haustoria is more or less confined to the younger roots. Haustoria on the main roots, probably take little part in the absorption of nutrients. He also observed that olđ haustoria died off leaving scars behind the newer ones which are formed from the tiny rcots. Acording to Rao (1911), young seedlings established roct attachments with other plants at a very early age, when less than even two months old. Rao (1933a) reported that osmotic pressure in the tissues of sandal was higher as compared to that in the tissues of the host plants which ensured a unidirectional flow of the nutrients from the host to the parasite. Though sandal plants can survive without host, experiments have proved beyond doubt that the host plants are absolutely necessary for the healthy growth sandal of plants (Ananthapadmanabha et al., 1984).

Iyanger (1960) inferred from his experiments that sandal drew nitrogen and phosphorus from the host plants, while potassium and calcium were taken directly from the soil. Rao (1933a, 1933b) from his studies on sandal plants grown in pots, with and without host plants concluded that sandal depends on the host plants for its requirement of nitrogen, phosphorus and potassium and derives its other mineral constituents including calcium and iron from the soil. Sreenivasaya (1931) emphasised that sandal depends mainly on its host for its vigorous growth,

heart wood formation and resistance to diseases insects. or (1973) reported that spike disease could be controlled Sarma through vector host plant control. Ramaiah et al., (1962) in their histochemical study of sandal root haustoria, found the presence of nutrients such as potassium, calcium, magnesium, iron, zinc and copper in the vascular strands of the haustorium. suggested that these minerals are possibly taken by sandal They from the host plants. The possibility of absorption of these nutrients directly from the soil also is very much in evidence as sandal roots do possess cation exchange capacity at a level comparable to the levels occurring in its hosts (Parthasarathi The cation exchange capacity of the white 1974). al., et succulent roots of young seedlings of sandal is, however, reported to be at a slightly low level (Kunda et al., 1971).

showed that Using radiotracer technique, Kunda (1974) calcium and phosphate could be absorbed directly by the roots of the sandal seedlings. He also showed that sandal seedlings could organic substances, amino acids, sugars and mineral draw phosphate from the host plants. Similar results of (1924),Rao Srimathi and Sreenivasaya (1963a) have also shown that there was translocation of nutrients from the host plant to the leaves of which was not associated with а host, was Sandal sandal.

reported to be poor in basic amino acid content (Srimathi et al., Total nitrogen in the leaves of sandal was found to be 1961). influenced by the nature of the host plant with which it was associated (Rao 1933b). The chemical nature of the sap of xylem sandal was also found to be influenced by the host plants of The core of the the sandal haustorium was reported (Rao, 1938). 1962a). have contained auxin (Srimathi and Sreenivasaya, to Occurrence of endopolyploidy in sandal haustorium was noted by Srimathi and Sreenivasaya (1962b). Iyanger (1965) described at length the physiology of root parasitism in sandal.

on a differential response in the cation exchange Based capacity (CEC) of the roots of host plants three categories of host plants were identified (Parthasarathi et al., 1974). In the first category of the host plants, the CEC of the host roots tended to increased on parasitisation by sandal, such hosts were good hosts. In the second category, the CEC did not taken as any marked variation, subsequent to parasitisation by show In the third sandal, such hosts were taken as of medium type. category of the host plants, the CEC showed a decrease subsequent to parasitisation of sandal and such hosts were taken as poor for sandal. Similarly based on the number of haustorial hosts connections as it might provide an index of nutritional level Rao (1938) and Rangaswamy and Griffth (1939) have classified the host good, medium and poor in pot culture experiments. plants as

According to them, when the number of haustorial connections made with the host was above 100, it was taken as good host, when this number ranged between 25 and 100 it was considered as a medium and when the number was less than 25, it was considered as host This basis assumed that higher number of haustoria poor host. would provide a greater efficiency of nutrition (Parthasarathi and Rai, 1989). Rao (1935) observed the growth of sandal plant in pot cultures with 108 different host species and categorised the host into three classes as determined by their capability to help the parasite to grow (i) vigorously (ii) normally and (iii) The chlorophyll activity and photosynthetic efficiency poorly. were found to increase progressively from poor to good hosts. The co-efficient of host efficiency also could be taken as an index the hosts as good, medium and poor in classifying (Ananthapadmanabha et al., 1988).

1965 of hosts (Iyanger, and wide range Sandal has а (1911) has pointed out that sandal Bhatnagar, 1965). Rao the roots of almost all species, though in varying attacked degrees. An instance of self-parasitism has been observed by Rao However, Whitehead (1916) warned that selfin sandal. (1911)sandal was a positive danger to its healthy parasitism in Rao (1938) reported that there were certain hosts which growth. were easily killed by sandal, similarly there were hosts which

might kill sandal. He further reported that sandal could feed on a hosts over 200 feet away from it. According to Mitchell (1938) association with evergreen or semi evergreen species helped sandal to grow better.

The parasitic nature of sandal was noticed by Scott (1871) and it was Brandis (1903) who suggested that sandal may derive part of its nutrition from soil as well. It is, however reported Iyanger (1965) that plants belonging to Annonaceae were not by parasitised by sandal. Though the young sandal seedlings possess root hairs, once the haustoria are formed on the roots, the root hairs degenerate (Rao, 1903). Deep rooted species were reported serve as better hosts than shallow rooted one (Rao, 1904). to their experiments on well established field sandal grown From plants, Lushington (1918) and Iyer (1918) showed that sandal is an obligate parasite. However, association of sandal with not host promoted luxuriant growth in sandal (Ananthapadmanabha et al., 1984). It is, however, of interest to note that out of sandal seedlings raised for experimental purpose at the the sandal research centre, Bangalore about three per cent of the seedlings were not seen to possess haustoria up to one year after germination (Nagaveni and Srimathi, 1985).

1.2. Studies on germination of sandal

produce "seeds" twice a year during March-Sandal trees collected during September-October. The seeds and April September-October and sown in April-May were found to give maximum germination (Nagaveni and Srimathi 1981b). Srimathi and (1983) reported that some trees flowered, once a year Kulkarni while some twice a year and still others throughout the year. flowering and germination pattern were differential The attributed for the survival of the species in adverse conditions Seeds collected from trees of different et al., 1985). (Bagchi age groups were reported to show similar percentage of viability, germination and plant percentage (Nagaveni and Srimathi, 1981b). is generally known that seeds which float, when immersed in It are non-viable and have less germinability. Ιt is seen water floatation method cannot be reliably applied to separate that viable and non-viable seeds in sandal (Nagaveni and Srimathi, 1985). It has been reported that chemicals like Hydrogen peroxide (Thiourea et al., 1960; Roberts and Narashova, indole 1964) acetic acid and indole butyric acid (Chatterjee, 1960) and zinc sulphate (Masev and Kutacek, 1966) were found to accelerate certain hard coat seeds. The normal period of germination in germination of sandal seeds is 30 to 90 days. But it was found

"seeds" - Morphologically the fruit of sandal is a one seeded berry. The dried fruit as such is sown as seed.

removal of mesocarp and treatment with Ceresan (an that organomercurial fungicide) hasten germination (10 to 20 days; Srimathi and Rao, 1969). Also, studies on sandal seeds involving (i) treatment with hot and cold water, (ii) machanical removal of the seed coat, and (iii) chemical scarification of the seed coat acid or sulphuric acid, showed that the alcohol with considerably increase percentage could the scarification germination and bring down the dormancy period (Nagaveni and Application of gibberellic acid was found to Srimathi,1981a). bring down the dormancy period and induce quick and uniform germination in sandal seeds (Nagaveni and Srimathi, 1980). Alpha and $oldsymbol{eta}$ amylases were two enzymes involved in the break down of starch in germinating seeds (Bewley and Black, 1978). The 🛠 amylase activity is due to the de novo synthesis of this enzyme under the control of endogenous gibberellic acid, $oldsymbol{eta}$ amylase activity, however, is due to the activation of the latent enzyme in the imbibing seed (Goodwin and Mercer, 1975). It is also known that endogenously applied gibberlic acid helps breaking off dormancy in seeds (Ovcharov, 1977). Another study on the effect of exogenously applied gibberellic acid on the relative levels of lpha and eta amylase activity in germinating sandal seeds revealed that gibberellic acid treatment brought a decrease in $oldsymbol{eta}$ amylase activity and an increase in \measuredangle amylase activity both in decoated those with intact seed coat (Ananthapadmanabha in seeds and et al., 1986).

1.3. Storage of sandal seeds

Nagaveni and Srimathi (1981b) who extensively investigated different methods of storage of sandal seeds under different conditions, namely, (i) gunny bag under room temperature, (ii) sealed polythene bags under room temperature, (iii) sealed bags at 4-5°C and (iv) seeds mixed with moist polythene vermiculate packed in polythene bags and kept at 4-5°C, found that, while simple gunny bag storage was a cheap and effective method of storage for a period of 8-9 months, storage in sealed polythene bags at 4-5°C was better for storing seeds beyond 12 an experiment at the Sandal Research Institute, months. In Bangalore (1980) when seeds were soaked in water and dried and then stored in polythene bags or gunny bags gave better result. Fresh seeds usually have a dormancy period of two months and 80 per cent of the seeds remain viable up to nine months (Nagaveni and Srimathi, 1981b).

1.4. Vegetative propagation of sandal

Uniyal <u>et al</u>., (1985) developed a method of vegetative propagation of sandal through root cuttings. The cuttings were treated with Seradix (IAA) powder and planted horizontally in a nursery bed. The cuttings with shoots and roots were transplanted in pots along with host. Success up to 60 per cent was achieved

this method. Vegetative propagation of sandal through air in layering or through root suckers also has been standardised (Rao In the method involving air layering, and Srimathi, 1977). branches with approximately two centimetre of diameter were The rings were dusted with Seradix B with active chosen. ingredient IAA (0.5 g) and covered with spagnum moss. Callus formation occured within 35 to 45 days in 95 per cent of the treated wounds. The rooted branch, thus separated was planted out along with a host.

In the method involving root-sucker, a trench was made around a mature sandal tree about 1.5 m from the base. The cuttings were treated with root promoting hormones (IAA or NAA, 50 ppm solution for a period of two hours) and planted in pots along with hosts. In about five to six months both shoot and root systems developed very well.

1.5. Tissue, organ and cell culture.

Induction of embryonal proliferation in sandal was attempted and it was found that similarity existed in the morphogenetic potentialities of the embryo of sandal and other angiospermic parasites (Rao, 1965). Yeast extract was found to initiate and stimulate the prolification of sandal endosperm. However, for satisfactory proliferation, the endosperm of sandal required an

auxin (2,4-D) and kinin (kinetin) in conjunction with yeast extract (Rangaswamy and Rao, 1963). Culturing method of lateral buds, root tips and haustoria of sandal in different method was attempted by Srimathi and Sreenivasaya (1963b). Bapat and Rao (1979) observed somatic embryogenesis in seedling callus of sandalwood. Lakshmi Sita (1986) made exhaustive studies on tissue culture of sandal from nodal and internodal segments, young shoots, suspension cultures, endosperm, anthers, shootcallus, leaf callus, embryo and hypocotyledonary segments. She has reported somatic embryogenesis from shoot callus cultures derived from 20-25 year old sandalwood trees.

1.6. Role of trace elements on growth of sandal seedlings

A systematic study of the deficiency symptoms caused in sandal seedlings under conditions of deficiency of copper, zinc, manganese, molybdenum and boron has been done by Kamala <u>et al</u>., (1986). They have observed characteristic differences in the peroxidase isoenzyme pattern in the leaves of sandal seedlings under deficiency of different trace elements.

Raising sandal seedlings successfully in the nursery has been facing serious problem in many sandal growing areas. As high as 100 per cent mortality of the seedlings was reported in some areas. This malady has been attributed to be the result of

and nematode infection (Sandal Research funqal combined Institute, Bangalore, 1983). Inorganic fertilizer application in doses to sandal seeedlings have resulted in fertilizer small toxicity and subsequent mortality which indicated that fertilizer application has to be avoided at the nursery stage in sandal (Rangaswamy et al., 1990). The diseases commonly found in sandal tree in seedling as well as mature state are sandal spike, leaf and damping off (Choudhari 1963). Bulter (1903) suggested curb that sandal thrives best when the ground is covered by a dense undergrowth. The accumulation of starch in the leaves of sandal first noticed by Bulter (1912). Rao (1911) reported that was sandal spike is caused by an insufficient supply of water to the plants owing to its relations with unfavourable hosts. Seedlings grown without host association did not thrive well (Rao, 1910). He also showed that the best method of raising sandal seedlings in a nursery was to sow the seeds in tile-cylinder with the host.

1.7. <u>Sandal types</u> and biochemical marker for oil bearing capacity in sandal.

A study of the isozyme pattern in respect of peroxidase, malate, dehydrogenase and esterase in the fully expanded green healthy leaves of different types of sandal plants showing variations in their leaf shape (Ovate, lanceolate, linear and elliptic) has shown that (a) characteristic differences existed

between the sandal leaf types in their pattern of isozymes of peroxidase and malate dehydrogenase at vegetative as well as flowering stages (b) in the sandal plants with big ovate leaves three sub-types existed and (c) the sandal plants with normal ovate wavy and normal ovate non-wavy leaves were genetically very close (Parthasarathi <u>et al.</u>, 1985).

Parthasarathi <u>et al</u>., (1986) found that the peroxidase isozyme pattern in the living bark tissue of any given sandal type remained characteristic of that type and the pattern was unaffected by seasonal variations or stage of development of the plant. It was however found that plants with ovate normal wavy, ovate normal non-wavy and ovate small leaves in the one hand and plants having ovate big leaves, sub-types II and III on the other, showed a common isozyme pattern in the living bark tissue. It was possible that the types showing a common pattern were genetically close.

Rao (1928) observed that the leaves of sandal plants growing in forest areas in close proximity to Strychnos plants tasted characteristically bitter. A few hosts have been found to produce discoloration in the leaves of the sandal feeding upon them (Rao, 1938). At the same time when sandal was found to feed on certain hosts such as <u>Crotalaria anergioides</u>, the leaves and the shape of the crown underwent remarkable transformations. A

careful reading of the chromatograms revealed that the leaves from sandal plants without any host plant, had practically no basic amino acid although they had a heavy concentration of exypoline. Leaves from sandals growing in associations with Leguminous plants, which are good hosts showed high concentration of the basic amino acids (Srimathi <u>et al.</u>, 1961).

Quantitative study of the activity of the specific peroxidase isozyme ("spi") in the common band showed that it remained at a stationary level, throughout in any given plant. Further spi. activity showed a strong negative correlation to the oil content in mature sandal plants. The spi. activity in a young sandal plant, irrespective of its age can be used in forecasting sandal oil per cent at maturity (Parthasarathi et al., 1986).

1.8. Physiological studies on sandal in relation to spike disease

No other aspect of sandal has attracted so much work as on of the physiology sandal affected by spike disease (Parthasarathi, 1979). Parthasarathi and Venkatesan (1982)investigated the pathological physiology in spiked sandal. In spiked sandal there is severe reduction in the leaf size and internodal distance, chlorosis of the leaves, necrosis of phloem elements, death of root ends and haustorical connections, followed by the death of the plant within one to three years after infection of the disease (Parthasarathi et al., 1986).

1.8.1. Mineral metabolism: The haustoria die in the spiked sandal, which cuts off the channel of nutrition from host plants. The cation exchange capacity of the sandal roots is, however, not affected in the diseased state (Parthasarathi <u>et al.</u>, 1971). The observation of a considerable decrease in the mineral content of spiked sandal (Rao and Sreenivasaya, 1928, Iyanger, 1928) indicated that a good part of the mineral nutrients is derived by sandal from the host plants through the haustoria which die in the diseased plant.

Chlorosis of the spike leaves: Parthasarathi and Rao 1.8.2. (1962a) and Parthasarathi et al., (1976) have studied in detail the chlorosis of spiked sandal leaves distribution of iron in the healthy and spiked sandal, and Ca/Fe ratio in the roots. The iron in the leaves was less while it was more in the level of stem and root regions of the spiked plant (Shestakov, 1930). The occurrence of high chlorophyllase activity at the time of chlorophyll accumulation in both healthy and spiked leaves was reported by Looney and Patterson (1967).

1.8.3. Carbohydrate metabolism: One of the prominent characteristics of spiked sandal is an abnormal accumulation of starch and sugars (Iyanger, 1928). The diastatic activity of the spiked sandal leaves, however, was found to be at a higher level (Sreenivasaya and Sastri, 1928; Sastri and Sreenivasaya, 1929;

Iyanger, 1937) despite the presence of high level of tannin and a reduced level of calcium therein, which were conducive for a lowering in the diastatic activity (Iyanger, 1937). A study of phosphorylase and phophoglucomutase activities and starch levels in the healthy and spiked sandal leaves showed that a lowering in the phophoglucomutase activity in the spiked leaves, could be a cause for the starch accumulation therein (Parthasarathi <u>et al</u>., 1977).

1.8.4. Nitrogen metabolism: In a study of the nitrogen fractions in the spiked leaves, Narasimhamurthy and Sreenivasaya (1929)found an increase in the total nitrogen and in the soluble N, basic N and amino N fractions, while there was а decrease in protein N. Parthasarathi et al., (1962b) found the nitrate reductase activity and nitrate N to increase in the spiked Parthasarathi et al., (1973a) mentioned that deficiency leaves. of trace elements, copper, zinc, manganese, molybdenum and cobalt in the soils of healthy and spiked sandal areas does not serve as a pre-disposing factor for the onset of the spike disease.

1.8.5. Organic acids: Iyanger (1933) found a specific accumulation of succinic acid and a decrease in the malic and oxalic acid levels in the spiked sandal levels. Ramaiah <u>et al.</u>, (1964) noticed a considerable accumulation of pyruvic acid while the level of oxaloacetic acid was almost negligible in the spiked leaves.
Osmotic pressure, pH buffering action: The pH of 1.8.6. healthy sandal leaf sap was found to lie between 5.2 and 5.7 while that for spiked sandal leaf was between 4.7 to 5.0. The osmotic concentration was higher in the diseased sample as compared to that in normal plant (Iyanger, 1928). The buffering capacities of the tissue fluids of the spiked sandal were higher than those of the healthy (Sreenivasan and Sreenivasaya 1934). The buffering capacity showed a decreasing gradient irrespective of the sandal host combination, from leaf bark, root and to wood.

1.8.7. Soil studies: Sreenivasaya (1930); Iyanger (1937,1960) suggested that the visible effects resulting from spike infection could be traced to the deficiency of lime as the immediate cause. Parthasarathi <u>et al</u>., (1973a) studied the role of four minor elements in sandal and showed that no deficiency either of the major elements or of the minor elements acted as a pre-disposing factor for the onset of a sandal spike disease.

Khan and Yadav (1962) reported calcareous soils with low amount of available nutrients resulted in incidence of spike disease in sandal. Physical properties of soil and its nutrient status showed positive correlation on growth of sandal in terms of increment in height and girth (Alexander and Thomas, 1985; Jain al., 1988). Sreenivasaya et and Rangaswami (1931) made observations on the role of soil on the growth of sandal.

Krishnamurthy <u>et al</u>. (1983) observed that the nature of top soil (0-30 cm) and available nitrogen have some influence on the growth of sandal.

2. Chemistry and Utilisation of sandal oil

2.1. <u>Characteristics and physiochemical properties</u>

Sandal oil possesses a sweet, fragrant, persistent and wood odour and is one of the most important essential oils produced in India (Parthasarathi and Rai, 1989). Sandal oil possesses the following physiochemical characteristics. Appearance: Nearly colourless to golden yellow, viscous oil liquid, specific gravity: 0.962 to 0.985, Refractive index, n: 1.499 to 1.506, Esters (as santalyl acetate): 1.6 to 5.4 per cent, solubility in 70 per cent aqueous ethanol: 1.5 vols.

2.2. Chemical composition

The chemical constituents of sandal oil have been studied by several workers, Guha and Bhattacharya; 1944, Ghatgey and Bhattacharya, 1956, Karawya and Wahba, 1962; Walker, 1968; Mouir and Takacs 1969; Gibson and Barneis, 1972; Adams <u>et al.</u>, 1975; Kretshmas <u>et al.</u>, 1976; Demole <u>et al.</u>, 1976. The general composition of sandal oil is as (i) \propto and β santalos-90 per cent,

(ii) $\not{\sim}$ and β santalones, epi β santalenes and santalyl acetate-6 per cent, (iii) Ketones: small percentage, (iv) Phenols - small percentage (v) Acids and Heterocyclic compound - small percent. The major odoriferous components of the sandal oil are the sesquiterpenoid constituents, $\not{\sim}$ santalol and β santalol. When sandal oil extracted from young and old trees were compared it was found that, the sandal oil from young trees showed lesser amount of santalols (Shankaranarayana and Parthasarathi, 1984). The biogenesis of santalenes is discussed by Bhati (1970).

Singh (1915) found that sandal trees grown in good fertile soil yield heartwood poorer in oil content than those grown in poor rocky soils.

2.3. Distillation/extraction

Sastry (1944) and Ramaswami (1972) have given an account of distillation of sandalwood by water distillation method and by steam distillation method. More than 90 per cent of the present production of sandal oil comes from modern steam distillation. Production of sandal oil by solvent extraction process could also be possible as reported by Rao (1939). Rectification of the benezene extract of sandal powder by 70 per cent ethanol followed by extraction with petrol, gives a higher yield of oil (Shankaranarayana and Venkatesan, 1981). Shankaranarayana and

Parthasarathi (1984) found that addition of polyethylene glycol 200 to the ethylacetate or acetone extracts of sandal powder before steam or vacuum distillation, yielded a higher percentage of sandalwood oil.

2.4. Adulterants of sandal oil

Cedarwood oil, copaiba balsam oil, Araucaria oil and organic compounds such as benzyl benzoate, benzyl alcohol, glyceryl triacetate, diethyl phythalate, isoprophyl myristate, terpineol and liquid paraffin are used as adulterants for sandal oil (Finnemore, 1926; Sastry, 1944; Guenther, 1952). Materials and Methods

3. MATERIALS AND METHODS

A pot culture experiment was conducted with the objective of studying the biology of the seedling of sandal (Santalum album Linn.) with special interest on the parasitic nature of sandal. The major objectives was to identify a few of the best host parasite combination. The experiment was conducted during July to March 1991 at the College of Forestry, Vellanikkara, 1990 Thrissur, Kerala, India, which is situated at 10 32' N latitude 76 to 10' E longitude at an altitude of 22.25 metres above and mean sea level. The maximum and minimum temperatures, rainfall relative humidity during the period of the experiment are and recorded and presented in Fig. 1.

3.1.Preparation of containers for potting

The polythene bags (250 gauge) of size 35 x 18 cm were used as the containers for potting. They were filled with potting mixture of sand, soil and powdered cowdung thoroughly mixed in the ratio of 1:1:1. The potting mixture was slightly acidic with a pH of 6.02. The bags were provided with sufficient number of holes at the bottom to facilitate drainage. The bags were then filled up to about 5 cm below the surface and the bags were serially numbered in each host parasite combination and were kept ready for planting.





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3.2. Selection of host species

Fifteen host species relevant to the State of Kerala were selected from the exhaustive list of host species listed by Barbar (1906). One month old host seedlings were collected from the Forest Nursery, Social Forestry Wing, Thrissur Forest Division. The selected host species, forty each, were then transplanted into the polythene bags and watered regularly till they got established. The selected host species were;

3.2.1. Ailanthus malabarica, DC.

It is a large ornamental deciduous tree from the family Simarubaceae. The tree occurs in evergreen forests of the Western Ghats, from the Konkan southwards on the eastern slopes of the Pegu Yoma in Burma. It is often planted for ornamental purpose in Southern India. The wood, which is soft and white, is used in match box industry, the tree yields a fragrant resin used as incense and in native medicine (Troupe,1921).

3.2.2. Albizia lebbek, Benth.

It is a moderate large sized deciduous tree from family Leguminosae Sub family Mimoseae. The tree is beleived to be wild in the Sub-Himalayan tract, Bengal, Chota Nagpur, the Indian

Peninsula, Burma, the Andaman and Cocos islands. The wood is dark brown with lighter or darker streaks and ornamental. It is used for making furniture and other agricultural implements, carvings etc. (Troupe, 1921).

3.2.3. Acacia auriculiformis, A. Cunn. ex Benth.

This is a small spreading evergreen tree belonging to family Leguminosae, sub family, Mimoseae. Its native range is Australia and Papua New Guinea. This species has been introduced to several countries because of its ability to grow on poor soils and produce high quality fuel wood (Troupe, 1921).

3.2.4. Azadirachta indica, A. Juss.

This is a moderate sized to large, usually evergreen tree from family Meliaceae. This tree is common in the open shrubforests of the dry zone of Burma, and is found apparently wild on the Shiwalik hills. Tree is wild and found everywhere in India, most commonly in the forests of Karnataka and parts of the Deccan. The wood is red, hard, and durable, and is used for house-buildings, furniture and many other purposes. The bark gum flowers and seed kernel oil are used in medicine.

3.2.5. Cassia siamea, Lam.

This is a moderate-sized evergreen tree from the family Leguminosae sub family Caesalpinieae. The tree is indigenous to Burma and the Southern part of Madras, largely planted for ornamental purpose. The tree grows fairly fast and it is easy to cultivate (Troupe, 1921).

3.2.6. Casuarina equisetifolia, Forst.

Casuarina is a large evergreen tree from the family Casuarinaceae, which is indigenous to the sandy shores and dunes along the coast of Chittagong, Tenasserim and the Andamans, particularly in little Andamans, also in the Malay Archipelago, the Malay peninsula chiefly on the east coast, the Pacific islands, North Australia. Its wood is very hard, used sometimes for poles and rafters but chiefly for fuel for which it is excellent (Troupe, 1921).

3.2.7. Dalbergia latifolia, Roxb.

This also is a large deciduous (nearly evergreen) tree from the family Leguminosae sub family Papilionaceae and is indigenous to sub-Himalayan tract from Oudh to Sikkim, Chota Nagpur, Central, Western, and Southern India. The wood is very hard,

strong, durable and ornamental. This is used for a large number of purposes, such as furniture, panelling, ornamental work, agricultural implements, etc (Troupe, 1921).

3.2.8. Delonix regia, Bojer.

This is a large evergreen tree from the family Leguminosae sub family Caesalpinioideae found in West Indies, Indonesia, North Central America, Australia. It is not native to India, but is extensively cultivated as an ornamental tree in tropical and subtropical areas of India (Tiwari and Singh, 1984).

3.2.9. Emblica officinalis, Gaertn.

is a moderate sized deciduous tree from the This family Euphorbiaceae Commonly found in most parts of deciduous forests of the country except in arid regions (Brandis, 1906). It is also found in the Himalayan ranges, moist peninsular low level, moist peninsular high level, moist sawannah and dry sawannah forests (Champion and Seth, 1968). It is considered to be a fairly good fodder tree and is lopped for fodder in Maharashtra, Madhya Pradesh, Orissa and Uttar Pradesh (Gupta, 1942; Lauric 1945; Shabnam, 1959).

3.2.10. Lagerstroemia speciosa, Pers.

It is a large deciduous tree from the family Lythraceae. This tree is found in Assam, Bengal, Chittagong, Western and Southern North Kanara and southern Konkan southward through India from Malabar to Travancore, Chota Nagpur and is common throughout Its wood is light red, hard and durable and is used Burma. for construction of boats and canoes, carts and other purposes. This is one of the most important timber trees of Burma and Assam (Troupe, 1921).

3.2.11. Leucaena leucocephala, Lam. de wit.

This is a medium sized evergreen tree from the family, Leguminosae, sub family Mimoseae. It is a multipurpose tree (MPT) native to Mexico, Gautemala and Honduras. It is introduced and grown in Philippines, Indonesia, Papua New Guinea, Malaysia, Australia, East and West Africa. In India it has been successfully introduced to Andhra Pradesh, Karnataka, Tamil Nadu, Himachal Pradesh and Uttar Pradesh (Lohani, 1979).

3.2.12. Pongamia glabra, Vent.

It is a moderate sized nearly evergreen tree from the family Leguminosae, sub family Papilionaceae. This is found throughout

India and Burma, chiefly along streams and rivers. Also found in Andamans and in Sundarbans as one of the characteristic species of the mixed forests of the littoral fringe. The wood, which is yellowish white, is not durable and is used mainly for fuel (Troupe,1921).

3.2.13. Psidium guajava, Linn.

This is an arborescent evergreen shrub or tree belonging to family Myrtaceae which attains a height up to ten meters and grows well in most tropical and subtropical soils and is indigenous to tropical America. Guavas have been thoroughly naturalised throughout the tropics and subtropics and are of commercial importance in Florida and Hawali (United States of America), India, Egypt, South Africa, Brazil, Colombia and the West Indies (Steven and Philip , 1980).

3.2.14. Swietenia mahagoni, Linn.

Mahogani is a large evergreen tree from the family Meliaceae. The cultivation of the tree was first attempted in India in 1975, when plants from the West Indies were introduced into the Royal Botanic gardens, Calcutta and since then it has been grown with varying success in many parts of India (Troupe, 1921).

3.2.15. Terminalia catappa, Linn.

It is a large handsome deciduous tree with whorled branches and large glabrous leaves, belonging to family Combretaceae. The tree is native to the Andamans and adjacent islands and of the Malay Peninsula, in coastal forests. It is extensively planted in tropical India and Burma, particularly around monasteries, both for ornament and for the sake of its fruit, the kernels of which are eaten (Troup 1921).

3.3. Raising sandal seedlings

3.3.1. Seed Collection

Sandal seeds were collected from the Institute of Forest Genetics and Tree Breeding, Coimbatore. Details regarding flowering and seed characterestics are given below:

Flowering season	-	May-June
Fruiting season	-	Oct-Dec
Seed per kilograms	-	6000 Nos
Purity percentage	-	Before cleaning 18 per cent After cleaning 100 per cent
Moisture per cent	-	7.6 per cent
Time taken for germination	-	30 days
Germination percentage	-	35 per cent
Plant percentage	-	15 per cent
Seedlings obtained per kilogram of seeds	_	900

3.3.2. Preparation of seed bed

An area of 12 x 1 meter was demarcated at the experimental field of the College of Forestry, Vellanikkara. This demarcated area was then dug to a depth of 0.3 to 0.45 meter and the stones, pebbles, roots etc. lying underneath were removed. After this a standard seed bed of size 12 m x 1 m x 10 cm with a flat surface was prepared (Khanna, 1981).

3.3.3. Pretreatment of seeds

Sandal seeds were given soaking treatment in cold water for 24 hours. This method is the normal procedure followed by the Forest Department in Kerala for raising sandal seedling in nursery. For this treatment, sandal seeds were completely soaked in cold water taken in plastic bucket for 24 hours. The soaking process was done at room temperature. The 24 hours soaked seeds were then immediately sown in the seed bed prepared.

3.3.4. Sowing

Seeds were sown on 16.7.1990 using broad-cast sowing method. After broadcasting the seed-beds were covered with fine sand and soil so as to cover the seeds. The seed-beds were then covered with thin layer of straw to hasten germination.

3.3.5. Cultural operations

The bed was watered regularly using a rose can except on rainy days. Hand weeding was done once in a month. To avoid termite attack, H C H 10 per cent (Hexa Chloro Cyclo Hexane) was dusted on the bed.

3.4. Design and layout of experiment

The experiment was conducted in a Completely Randomised Design (CRD) with fifteen treatments and one control. Each host species was treated as one treatment and was replicated 40 times. After transplanting, during the field establishment of host and sandal seedlings in pots, there was some mortality. Similarly, mortality was also seen during the experimental period which has finally resulted in the unequal replications. The treatements were as follows:

T ₁	Sandal alone (Control)
^т 2	Sandal + <u>Ailanthus</u> <u>malabarica</u>
^т 3	Sandal + <u>Casuarina</u> equisetifolia
${}^{\mathrm{T}}_{4}$	Sandal + <u>Albizia</u> <u>lebbek</u>
^т 5	Sandal + Emblica officinalis
т _б	Sandal + <u>Leucaena</u> <u>leucocephala</u>

^т 7	Sandal	Ŧ	<u>Pongamia</u> glabra
^т 8 -	Sandal	+	<u>Terminalia</u> <u>catappa</u>
т ₉	Sandal	+	Legerstroemia speciosa
^T 10	Sandal	+	Dalbergia latifolia
T_11	Sandal	+	<u>Swietenia</u> <u>mahagoni</u>
^T 12	Sandal	+	Acacia auriculiformis
^T 13	Sandal	+	Azadirachta indica
^T 14	Sandal	+	<u>Cassia siamea</u>
^T 15	sandal	+	<u>Delonix</u> <u>regia</u>

T₁₆ Sandal + <u>Psidium</u> guajava

3.5. Planting of seedlings to polybags

Four weeks old seedlings with average height of 7.0 cm with number leaves and girth measuring around 0.3 mm, 6-8 of were used for transplanting. The seedlings were carefully removed from Initially two seedlings per polythene bag, containing the bed. host, were planted, which was subsequently thinned down the to per bag as soon as the seedlings were fully established. one This ensured uniform number of seedlings in all polythene bags.

Before planting the seedlings to the polybags containing hosts, the following data on seedling height, number of leaves, collar diameter and tap root length of the sandal seedlings were recorded.

3.6. Observations recorded

Biometrical observations were recorded from the established sandal seedlings at fortnightly intervals. Observations were also recorded after destructive sampling at 30 days interval.

3.6.1. Fortnightly observations

At an interval of 15 days, observations of sandal seedlings growing with the host species were recorded as follows:

3.6.1.1. Seedling height

Seedling height was measured from tip of the growing point to the collar region using a measuring tape and the mean height in centimeters was worked out.

3.6.1.2. Collar diameter

The collar diameter of the seedling was measured using vernier callipers, in millimeter and mean was worked out.

3.6.1.3. Number of Leaves

The number of leaves of seedling was counted and mean was recorded.

3.6.2. Monthly observations

The first sampling was done on the 30th day and subsequent sampling were at 30 days interval. At each sampling three seedlings of sandal from each host combinations were taken, by carefully spliting open the polythene bags using a clean blade keeping the root system intact. The mud, debris, weeds, etc. sticking on the roots were carefully removed by repeatedly washing the samples in running water. The seedlings were then spread out for a brief period for drying and the following observations were recorded.

3.6.2.1. Shoot height

The length of the shoot was measured from the tip of the growing point to the collar region using a measuring tape. From this the mean length was calculated.

3.6.2.2. Tap root length

The length of the tap root was measured from the collar region to the tip of the main tap root using a measuring tape and their mean length was calculated.

3.6.2.3. Dry matter production per sandal seedling

The stem was first detached from the tap root at the collar region and its fresh weight recorded and then kept in paper bags separately. Similarly the fresh weight of the root portion was also recorded and kept separately in paper bags. These bags were then kept in an hot air oven maintained at $80\pm 2^{\circ}$ C for 48 hours and dried to a constant weight. The final weights were then determined using an electronic balance (Sartorius).

3.7. Statistical analysis

The experimental data were statistically analysed by applying techniques of analysis of variance for CRD and their significance tested by 'F' test (Snedecor and Cochran, 1967).

3.8. Anatomical study

Hand sections of the haustorial connections of sandal seedlings with different host plants were taken. Uniformly thin sections were stained using Safranin and were mounted in glycerin medium. The section were carefully observed under the microscope, and necessary microphotographs were taken to examine nature of connections andother anatomical details.

Results

4. RESULTS

experiment was carried out in the College of Forestry, An Vellanikkara to identify promising sandal- host combinations to recommended and adopted for large scale sandal plantations. be growth parameters like height, girth and number of leaves When it was found that they could be were critically examined, into three broad categories which for the sake of classified convenience are recognised as (i) * sandal-casuarina (ii) sandalsandal-emblica combinations. Sandal-casuarina (iii) dalbergia category includes species namely terminalia, pongamia, albizia, azadirachta. Sandal-dalbergia category includes legerstromia, sandal-emblica category leucaena, acacia and cassia, whereas includes ailanthus, delonix, swietenia, psidium and sandal alone. Sandal-casuarina showed the best performance followed by sandal-Sandal emblica combination showed some amount of dalbergia. or allelopathic effect. The results from the antagonistic investigation, are summarised in this chapter.

4.1. Height

The data on mean height of sandal seedling at different stages of development with the host species after transplantation

 The generic names of the hosts are used as common names and hence they are used in lower case letter.

	_	height (cm)						
Sl. Species	Species	Days after transplanting						
No.		30	60	90	120	150		
 1.	<u>Santalum</u> album (alone)	9.20	12.50	14.27	15.93	19.60		
2.	<u>Ailanthus</u> malabarica	8.81	8.99	9.29	9.29	10.46		
3.	Casuarina equisetifolia	8.46	9.55	13,48	20.40	27.78		
4.	<u>Albizia lebbek</u>	9.28	11.07	13.65	15.84	20.51		
5.	Emblica officinalis	8.08	7.57	8.59	9.47	9.93		
6.	Leucaena leucocephala	7.90	7.99	8.72	9.77	10.13		
7.	<u>Pongamia</u> glabra	8.10	9.48	12.31	14.98	19.57		
8.	Terminalia catappa	7.29	8.49	11.70	17.08	25.48		
9.	Lagerstroemia speciosa	6.87	7.40	8.88	11.21	14.10		
10.	<u>Dalbergia</u> <u>latifolia</u>	7.24	8.48	10.06	12.10	16.04		
11.	<u>Swietenia</u> <u>mahagoni</u>	7.32	7.57	8.36	9.15	10.34		
12.	<u>Acacia</u> auriculiformis	7.64	8.44	9.14	9.93	13.53		
13.	<u>Azadirachta</u> <u>indica</u>	7.45	9.10	11.03	12.39	17.07		
14.	<u>Cassia</u> <u>siamea</u>	6.55	7.85	9.14	10.44	16.62		
15.	<u>Delonix</u> <u>regia</u>	7.22	8.30	9.25	12.44	15.71		
16.	<u>Psidium guajava</u>	6.90	7.95	10.68	12.50	15.81		

Table 1. Mean height of sandal seedling at various stages of development with different host species.



Fig. 2. Height of sandal seedling at different stages of development with different host species. (---: Sandal - casumina; o-o-o: Sandal - dalbergia; -A-4-: Sandal - emblica)

statistically analysed and are presented in Table were 1. The three categories are ploted in Figure 2. Observations on 🦾 all ahowed a significant difference in height (Appendix 1). stages On 30th day after transplanting (DAT) among the three categories, sandal-casuarina recorded the maximum height of 8.46 cm, sandalemblica recorded 8.08 cm and the minimum height of 7.24 сm was recorded by sandal-dalbergia. However, on the 60th and 90th DAT it was sandal-emblica which showed the minimum height growth of 7.57 cm and 8.59 cm respectively. Sandal-dalbergia recorded and intermediate height (8.48 and 10.06 cm) and sandal-casuarina showed the maximum height of 9.55 cm and 13.48 cm respectively. Observation on 120th and 150th DAT showed that sandal-casuarina was statistically superior (Appendix VIII) and this combination showed the maximum height of 20.40 cm and 27.78 cm respectively and sandal-dalbergia recorded (12.10 and 16.04 cm) respectively. both this observations sandal-emblica combination showed On the minimum height of 9.47 and 9.93 cm respectively. Throughout the investigation period a steady increase in the height of sandal seedling in sandal-casuarina and sandal-dalbergia combinations was observed.

4.2.Collar girth

Statistically analysed data on mean collar girth of sandal seedlings at different stages of development in combination with 15 host species are presented in Table 2. Figure 3 represents

Table 2. Mean collar girth of sandal seedling at various stages of development with different host species.

Collar girth (cm) Days after transplanting Species S1. No. 60 90 120 30 150 ____ ____ 1. Santalum album (alone) 0.184 0.221 0.275 0.301 0.3540.353 0.374 0.396 0.470 2. Ailanthus malabarica 0.276 Casuarina equisetifolia 0.320 0.336 0.395 0.520 0.833 3. 0.300 0.356 0.382 0.489 0.566 Albizia lebbek 4. Emblica officinalis 0.263 0.280 0.310 0.319 0.400 5. 0.344 0.382 0.220 0.268 0.359 Leucaena leucocephala 6. 7. Pongamia glabra 0.240 0.319 0.330 0.366 0.410 0.560. 0.283 0.327 0.456 8. Terminalia catappa 0.175 9. Lagerstroemia speciosa 0.160 0.281 0.313 0.480 0.542 10. Dalbergia latifolia 0.179 0.200 0.320 0.392 0.544 0.317 0.344 11. Swietenia mahaqoni 0.160 0.200 0.401 0.264 0.333 0.391 12. Acacia auriculiformis 0.1580.194 13. Azadirachta indica 0.171 0.200 0.225 0.295 0.345 0.297 0.333 14. Cassia siamea 0.173 0.231 0.265 0.221 0.272 0.321 0.395 15. Delonix regia 0.173 0.183 0.231 0.284 0.341 0.410 16. Psidium guajava



Fig. 3. Collar girth of sandal seedling at different stages of development with different host species. (——: Sandal — casurina; o-o-o: Sandal — dalbergia; -△-△-: Sandal — emblica)

the collar girth of three categories of sandal combinations and their analysis of variance is given in Appendix II. With respect to collar girth the combinations were statistically superior throughout the period of investigation. However, observations on indicated that Sandal-casuarina recorded the maximum 30th DAT (0.320 cm) followed by sandal-emblica (0.263 cm) and the minimum was sandal-dalbergia (0.179 cm). At 60th DAT sandal-casuarina recorded 0.336 cm followed by sandal-dalbergia (0.283 cm) and sandal-emblica (0.280 cm). Observations on 90th, 120th and 150th DAT showed that sandal-casuarina showed the maximum collar girth of 0.395 cm, 0.520 cm, 0.833 cm respectively. This was followed 0.544 0.392 сm sandal-dalbergia 0.320 cm, CM, by respectively. Sandal-emblica combination recorded the minimum of cm, 0.319 cm and 0.400 cm respectively during the three 0.310 The results showed a significant difference on observations. 150th DAT, with sandal-casuarina differing significantly from the rest (Appendix IX). In all the three categories there was a steady increase in mean collar girth.

4.3. Number of leaves

Number of leaves is an indirect index of healthy seedling. The data on mean number of leaves of sandal seedlings at different stages of development with the 15 host combinations were statistically analysed and are presented in Table 3. Figure 4 represents the mean numbers of leaves of three sandal-host

	Species	Leaf number						
		Days after transplanting						
No.		30	60	90	1,20	150		
· 1.	Santalum album (alone)	12.93	17.80	15.67	17.20	22.53		
2.	<u>Ailanthus</u> <u>malabarica</u>	10.69	12.92	11.32	11.31	8.67		
3.	<u>Casuarina</u> <u>equisetifolia</u>	<u>a</u> 10.48	14.08	17.83	26.05	28.56		
4.	<u>Albizia</u> <u>lebbek</u> -	11.03	15.08	16.32	17.76	24.16		
5.	Emblica officinalis	9.03	9.62	7.91	10.69	8.52		
6.	<u>Leucaena</u> <u>leucocephala</u>	9.52	11.74	11.08	12.38	11.57		
7.	<u>Pongamia glabra</u>	9.37	13.53	18.48	20.32	22.56		
8.	<u>Terminalia</u> catappa	9.94	13.31	15.48	18.13	27.10		
9.	Lagerstroemia speciosa	9.32	11.16	13.65	15.48	18.48		
10.	<u>Dalbergia</u> <u>latifolia</u>	9.34	12.79	15.44	16.31	18.58		
11.	<u>Swietenia</u> <u>mahagoni</u>	10.85	12.56	13.41	14.61	12.21		
12.	<u>Acacia</u> auriculiformis	9.87	12,33	13.47	14.17	14.03		
13.	Azadirachta indica	10.75	13.50	14.84	16.24	18.00		
14.	<u>Cassia siamea</u>	. 9.27	11.00	12.12	13.61	16.14		
15.	<u>Delonix</u> <u>regia</u>	9.40	12.40	13.56	9.62	10.14		

9.20 12.10 12.20

13.67

14.38

16. Psidium guajava

Table 3. Mean leaf number of sandal seedling at various stages of development with different host species.



DAT Fig. 4. Number of leaves of sandal seedling at different stages of development with different host species. (---: Sandalcasurina; o-o-o: Sandal - dalbergia; -A-A-: Sandal - emblica)

categories. Their analysis of variance is given in Appendix III. statisticaly The data on analysis were found to be significant leaves throughout the mean number of with respect to investigation (Appendix III). Observation on 90th, 120th, 150th showed that sandal-casuarina produced the maximum number of DAT the numbers being 17.83, 26.05, 28.56 respectively. On leaves 150th DAT, the difference was significant (Appendix X). During sandal-dalbergia combination the above mentioned period the number of leaves 18.58 mean 15.44, 16.31 and produced On all the three occasions sandal-emblica respectively. minimum number of leaves the values being 7.91, produced the and 8.52 respectively. In sandal-casuarina and sandal-10.69 dalbergia there was an increasing trend in the number of leaves throughout the experimental period whereas sandal-emblica showed a decreasing trend.

4.4. Shoot and root length

The mean shoot and root length of sandal seedling with 15 host combinations at different stages of development were statistically analysed and the data are presented in Table 4 and 5 and the Analysis of variance in Appendix IV and V. At all the stages of observation, treatments showed significant difference except on 60th DAT. Figure 5 depicts the shoot and root length of three categories. At 30th DAT sandal-emblica recorded the maximum (9.0 cm) shoot length followed by sandal-dalbergia (7.4

cm) and sandal-casuarina (6.4 cm). On 60th DAT same pattern was sandal-emblica recording the maximum (10.3 cm) observed with followed by sandal-casuarina (9.9 cm) and sandal-dalbergia (9.8 cm). However on 90th DAT onwards it was sandal-casuarina which showed the maximum shoot length of 21.9 cm (90th DAT), 26.5 cm (120th DAT), 31.2 cm (150th DAT) and had significant differences from the rest, on 150th DAT (Appendix XI). On all these instances namely 90th, 120th and 150th DAT sandal-dalbergia recorded the 16.3 cm, 17.0 cm and 18.0 cm respectively, of shoot length 11.0 sandal-emblica 11.8 cm, cm 10.5 cm, followed by In the case of sandal-emblica, during 120th DAT a respectively. decline in shoot length was observed, as against sandal-casuarina and sandal-dalbergia combination which showed a steady increase.

regards root length, the treatments showed significant As difference at all stages of developement (Appendix V). Sandalcasuarina recorded the maximum root length throughout the for the 90th DAT and thereafter it showed investigation but а steady increase in root length whereas sandal-emblica showed а decline on the 60th DAT onwards. On 30th and 60th DAT sandalas 7.40 and 14.67 сm respectively casuarina recorded сm and sandal-emblica sandal-dalbergia (5.53 cm 6.97 cm) aqainst recorded 5.60 cm and 6.73 cm respectively. On 90th DAT sandalrecorded the maximum of 15.37 cm followed by sandaldalbergia Sandal-emblica recorded the least mean cm). casuarina (10.33

Mean shoot length of sandal seedling at various stages of development with different host species. Table 4.

			Shoot	L length	(cm)			
	- Species	Days after transplanting						
No.	-		60		120			
1.	Santalum album (alone)	8.7	11.0	12.1	13.0	15.1		
2.	Ailanthus malabarica	5.0	7.7	9.6	2.1	7.2		
3.	<u>Casuarina</u> equisetifolia	6.4	9.9	21.9	26.5	31.2		
4.	<u>Albizia lebbek</u>	6.9	10.4	10.3	21.1	19.5		
5.	Emblica officinalis	9.0	10.3	10.5	11.8	11.0		
6.	Leucaena leucocephala	7.9	9.2	8.8	7.5	10.3		
7.	Pongamia glabra	8.7	9.5	14.0	22.0	24.5		
8.	Terminalia catappa	8.2	9.0	14.8	23.7	25.7		
9.	Lagerstroemia <u>speciosa</u>	5.2	7.5	11.9	13.8	15.3		
10.	<u>Dalbergia</u> <u>latifolia</u>	7.4	9.8	16.3	17.0	18.0		
11.	Swietenia mahagoni	7.5	9.3	12.0	13.3	13.7		
12.	Acacia auriculiformis	6.2	. 8.8	10.2	12.3	13.3		
13.	<u>Azadirachta</u> indica	5.8	9.3	11.0	15.0	18.7		
14.	Cassia siamea	6.1	10.2	11.3	18.0	16.0		
15.	<u>Delonix regia</u>	6.0	8.5	8.3	9.2	10.7		
16.	<u>Psidium guajava</u>	7.1	8.7	8.2	12.0	12.0		

Table 5. Mean root length of sandal seedling at various stages of development with different host species.

				· ··· ··· ··· ··· ··· ··· ··· ··· ···		
	Root length (cm)					
Sl. Species No.		Days aft	ter trans	planting	[
	30	60	90	120	150	
1. <u>Santalum</u> <u>album</u> (alone)	5.67	7.17	8.50	7.83	8.10	
2. <u>Ailanthus</u> malabarica	4.70	9.73	11.17	9.67	10.47	
3. <u>Casuarina equisetifolia</u>	<u>a</u> 7.40	14.67	10.33	21.33	22.77	
4. <u>Albizia lebbek</u>	6.43	6.83	15.87	15.37	12.43	
5. <u>Emblica</u> officinalis	5.60	6.73	5.40	5.80	5.77	
6. <u>Leucaena</u> <u>leucocephala</u>	7.80	12.57	.16.20	15.67	13.23	
7. <u>Pongamia</u> glabra	9.60	10.90	10.43	14.33	13.10	
8. <u>Terminalia</u> <u>catappa</u>	8.20	8.17	17.90	19.33	17.10	
9. <u>Lagerstroemia</u> speciosa	4.40	4.73	12.57	12.03	11.47	
10. <u>Dalbergia</u> <u>latifolia</u>	5.53	6.97	15.37	13.07	15.27	
ll. <u>Swietenia</u> mahagoni	10.10	11.90	13.43	16.00	11.37	
12. <u>Acacia</u> <u>auriculiformis</u>	8.10	10.17	9.07	24.67	12.80	
13. <u>Azadirachta indica</u>	12.10	13.67	15.17	7.27	9.93	
14. <u>Cassia</u> <u>siamea</u>	9.07	6.67	11.50	5.90	9.53	
15. <u>Delonix</u> regia	7.37	6.93	6.63	8.57	9.33	
l6. <u>Psidium</u> guajava	8.27	9.23	770	10.70	10.10	

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stages of development with different host species. (22: Sandal - casurina; 2: Sandal - dalbergia; 2: Sandal - dal sandal - emblica).

root length at this stage (5.40 cm). Observations on 120th and 150th DAT showed that sandal-casuarina, recorded maximum of 21.33 cm and 22.77 cm. Sandal-dalbergia recorded a root length of 13.07 cm and 15.27 cm and sandal-emblica showed the least 5.80 cm and 5.77 cm. On 150th DAT sandal-casuarina was statistically superior to other treatments (Appendix XII).

4.5. Dry matter production

Dry matter accumulation as a function of growth was calculated at 30 day intervals. Table 6 and 7 depicts the accumulation of mean shoot dry weight and mean root dry weight at different stages of development of sandal seedling with 15 host combinations. The shoot dry weight and root dry weight of three categories are shown in Figure 6 and 7 and their analysis of variance in Appendix VI and VII.

With reference to shoot dry weight, treatments showed significant differences at all the stages of investigation except Shoot dry weight of sandal-casuarina and on the 30th DAT. sandal-dalbergia showed a steady increase during all the observations except on the 120th DAT, at the same time a decreasing trend was observed in the shoot dry weight of sandalemblica combination. On 30th DAT sandal-emblica recorded the maximum shoot dry weight of 0.392 g per plant followed bv sandal-dalbergia (0.076 g per plant). Sandal-casuarina recorded
	deveropment with							
			Dry weig	ht (g pe	er plant)			
sı.	- Species	Days after transplanting						
No.	-	30	60	90	.120	150		
1.	Santalum album (alone)	0.019	0.026	0.028	0.027	0.027		
2.	<u>Ailanthus</u> <u>malabarica</u>	0.030	0.069	0.087	0.149	0.096		
3.	Casuarina equisetifolia	0.054	0.175	0.666	0.511	0.591		
4.	<u>Albizia lebbek</u>	0.080	0.121	0.117	0.367	0.170		
5.	Emblica officinalis	0.392	0.116	0.077	0.062	0.063		
6.	Leucaena leucocephala	0.052	0.066	0.069	0.049	0.069		
7.	Pongamia glabra	0.050	0.091	0.150	0.493	0.548		
8.	<u>Terminalia</u> <u>catappa</u>	0.095	0.093	0.226	0.884	0.382		
9.	Lagerstroemia speciosa	0.047	0.060	0.114	0.214	0.276		
10.	<u>Dalbergia</u> latifolia	0.076	0.098	0.575	0.269	0.288		
11.	Swietenia mahagoni	0.119	0.077	0.091	0.117	0.135		
12.	<u>Acacia</u> <u>auriculiformis</u>	0.072	0.089	0.087	0.161	0.194		
13.	<u>Azadirachta</u> indica	0.063	0.104	0.121	0.157	0.183		
14.	Cassia siamea	0.051	0.134	0.185	0.087	0.077		
15.	<u>Delonix</u> <u>regia</u>	0.068	0.097	0.096	0.107	0.133		
16.	<u>Psidium</u> guajava	0.046	0.053	0.106	0.077	0.071		

Table 6. Mean dry weight of sandal shoot at various stages of development with different host species.



Fig. 6. Dry weight of sandal shoot at different stages of development with different species. (___: Sandal-Casurina; o-o-o: Sandal - Dalbergia; -△-△-: Sandal - emblica)

per plant) during this observation. minimum (0.054 g the However, observations on 60th DAT indicated that sandal-casuarina (0.175 g per plant) followed by sandalrecorded the maximum emblica (0.116 g per plant) and the least was sandal-dalbergia (0.098 g per plant). Sandal-casuarina recorded the maximum on 90th, 120th, 150th DAT (0.666 g per plant, 0.511 g per plant and 0.591 q per plant) respectively, and was statistically superior to all other treatments on the 150th DAT (Appendix It was sandal-emblica combination which produced the XIII). least shoot dry weight accumulation on all the observations (0.077 g per plant, 0.062g per plant, 0.063 g per plant).

Root dry weight also showed considerable variation among the treatments. The maximum root dry weight production by all the three combinations was observed on 90th DAT. Sandal-casuarina combination produced the maximum root dry weight throughout the investigation period and it differed significantly from all the other treatments on 150th DAT (Appendix IV). Observations on sandal-emblica combination showed an increasing trend upto 90th DAT and there after there was a steady decline in the root dry weight production.

It was sandal-emblica combination which produced the maximum root dry weight (0.038 g per plant) on 30th DAT followed by sandal-dalbergia (0.034 g per plant) and sandal-casuarina produced the minimum (0.012 g per plant). On 60th DAT the root

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Mean dry weight of sandal root at various stages of development with different host species. Table 7.

		Dry weight (g per plant)						
Sl.	Species	Days after transplanting						
No.	-	30	60	90	120	150		
					· · · · · · · · · · · · · · · · · · ·			
1.	<u>Santalum</u> <u>album</u> (alone)	0.014	0.017	0.019	0.016	0.016		
2.	Ailanthus malabarica	0.008	0.032	0.050	0.070	0.042		
3.	Casuarina equisetifolia	0.012	0.081	0.228	0.158	0.159		
4.	Albizia lebbek	0.030.	0.033	0.067	0.174	0.130		
5.	Emblica officinalis	0.038	0.053	0.053	0.018	0.014		
6.	Leucaena leucocephala	0.028	0.043	0.040	0.0065	0.062		
7.	Pongamia glabra	0.041	0.034	0.088	0.133	0.124		
8.	<u>Terminalia</u> catappa	0.049	0.036	0.118	0.186	0.164		
9.	Lagerstroemia speciosa	0.016	0.023	0.034	0.082	0.083		
10.	<u>Dalbergia latifolia</u>	0.034	0.043	0.195	0.075	0.088		
11.	Swietenia mahagoni	0.058	0.035	0.202	0.023	0.015		
12.	<u>Acacia</u> auriculiformis	0.025	0.141	0.025	0.081	0.043		
13.	<u>Azadirachta indica</u>	0.031	0.038	0.039	0.076	0.109		
14.	<u>Cassia</u> <u>siamea</u>	0.022	0.033	0.083	0.027	0.042		
15.	Delonix regia	0.021	0.021	0.020	0.027	0.028		
16.	Psidium guajava	0.035	0.044	0.032	0.060	0.043		



Fig. 7. Dry weight of sandal root at different stages of development with different host species. (----: Sandaldasurina ; o-o-o: Sandal - dalbergia; -△-△-: Sandalemblica)

dry weight production was maximum in sandal-casuarina (0.081 q per plant). This was followed by sandal-emblica (0.053 g per)and minimum was that of sandal-dalbergia combination plant) (0.043 g per plant). Observation of the 90th, 120th and the 150th DAT showed that sandal-casuarina recorded the maximum of 0.228, 0.158 and 0.159 g per plant respectively followed by sandal-dalbergia (0.195, 0.075 and 0.088 g per plant). Sandalemblica produced the minimum root dry weight of 0.053, 0.018 and 0.014 g per plant respectively.

4.6. Anatomical study of haustorium

The process of haustoria formation has been observed from the different sections. It is found that young haustorium appears as a small hemispherical outgrowth (Plate I). The free end after coming in contact with the host root gradually flattens. The young haustorium consists of a narrow neck, a massive parenchymatous body, and a broad apex (Plate II). The centre of the body is occupied by a nucleus having rich cytoplasmic cells.

When the haustorium comes in contact with the host root, the outermost cells of the apex become radially elongated and richly cytoplasmic, and appear glandular. The peripheral part of the body of the haustorium grows rapidly and extends around the glandular area of the sucker, finally coming in contact with the

host. This spreading tissue, on either side of the host, forms the clasping folds (Plate III).

In the surface layers of the haustorium there is glandular activity. When a hard root has to be attacked the glandular cells fail to effect penetration. In such cases an external gland is formed schizogenously (Plate IV).

The parent root and the host show direct vascular connection with that of the haustorium. In a mature haustorium, the vascular cylinder resembles an inverted flask (Plate V). The xylem consist of vessels which are in union with those of the host. The tissue of the mature haustorium, especially those of the sucker, on penetration of the host spread outwards, raising the cortex of the host along the cambial layer. firmly When established, secondary growth occurs in the haustorium which renders it permanent and functional for a long time (Plate VI). The vascular connections between the host and sandal becomes so intimate, that the host root and parasitic root become almost a physiological unit, catering to the nutritional requirements of sandal. In certain sandal-host combinations like sandal-emblica, sandal-acacia, sandal-ailanthus well developed haustorial connections were not seen.

Plate I. Haustorium of sandal on casuarina

🖂 - Haustorium

Plate II. L.S. of young haustorium of sandal when associated with dalbergia showing neck, parenchymatous body and apex. (x 380)







Plate III. L.S. of young haustorium of sandal showing the clasping folds. (x 490)

H - host rootlet, C - clasping folds.

Plate IV. L.S. of sandal haustorium when associated with emblica showing the failure of penetration. (x368)

H - host rootlet, C - glandular cells.



Plate V. L.S. of a mature haustorium showing the vascular cylinder. (x 410)

V - Vascular cylinder, H - host rootlet,

X - Xylem.

Plate VI. L.S. of mature haustorium showing the established connections with the host root (x 368)

H - host rootlet, V - Vascular cells, X - Xylem.





Discussion

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5. DISCUSSION

An investigation was carried out at the College of Forestry, Vellanikkara during 1990-1991 to study the biology of sandal particular reference to the sandal host with seedling experiment was conducted in The а interrelationship. Completely Randomised Design (CRD) with 15 host combinations. The result obtained with regards to the seven growth parameters namely height, collar girth, number of leaves, root length, shoot length, root biomass and shoot biomass are discussed in this chapter.

5.1 Seedling height

Seedling height is an important growth parameter for the successful establishment of plants. The results obtained in this respect showed that uniformly, throughout the growth period under observation, combinations such as sandal-casuarina, sandalterminalia, sandal-albizia, sandal-dalbergia, sandal-pongamia showed higher growth increments as compared to sandal-emblica, Combinations such sandal-swietenia and sandal-ailanthus. as sandal-lagerstoemia, sandal-delonix, sandal-psidium were in Sandal-casuarina showed the maximum height, this might between. to the cladophylous canopy of casuarina which gives due be sufficient protection for sandal seedling, at the same time, allows optimum sunlight for sandal to grow. Bhatnagar (1965)already reported that sandal needs lateral shade in early has

In sandal-leucaena combination, the host plant grew very days. leaving sandal in complete shade denying it sufficient fast light for normal growth. In sandal-emblica combination, even though the canopy of emblica is same as that of casuarina the poor performance of sandal might be due to some allelopathic effect. This is in agreement with the findings of Rao (1938) who showed that some host has adverse effect on sandal. Another possible reason for the better performance of combination such as sandal-terminalia, sandal-pongamia might be sandal-casuarina, due to the better haustorial connection established between this host and sandal which enabled better absorption of nutrients by from the host plants. Synergistic effect might be the sandal other possible reason for the better performance of sandalcasuarina, sandal-terminalia, sandal-pongamia, sandal-albizia which has to be further investigated.

5.2 Collar girth

Good collar girth is recognised as one of the most important characterestic of a healthy seedling. An uniform increase with regards to collar girth was observed for all the treatments throughout the period under observations. The best collar girth among the treatments was found in sandal-casuarina, this was closely followed by sandal-terminalia, sandal-dalbergia, sandalalbizia. Combination like sandal-ailanthus, sandal-emblica, sandal-leucaena, sandal-swietenia registered lower collar girth

comparative scale. The exceptionally better performance in a the former combinations could be attributed to with found similar beneficial effects of the particular svnergistic or Shade offered by the host species seems to have a combination. significant role in determining the collar girth of sandal seedling. This is evident from the fact that species like casuarina, albizia, terminalia, dalbergia which have a canopy allowing ample sunlight to penetrate, allowed better collar girth of sandal seedling. At the same time combinations like sandalsandal-swietenia, sandal-acacia and sandal-delonix emblica, exhibited a reduced collar girth which can be due to some The extent of haustorial connections allelopathic effects. the host and sandal tended to result in better between performance with respect to collar girth too.

5.3 Number of leaves

Leaves being the photosynthetic apparatus of plants, their number has a direct influence on the performance of seedlings. This important parameter, too was found to be significantly differing among the treatments through out the period under investigation. When the final obeservations were taken sandalcasuarina association performed better than the rest. Sandal produced a higher number of leaves when associated with some of the host species like casuarina, terminalia, dalbergia, pongamia and lagerstoemia. Similar observations about good growth of

sandal in association with certain host species has been reported Ananthapadmanabha et al., (1984). This again points to by the fact that host species has a significant effect on the performance of sandal. Sandal grown with species like emblica, leucaena, swietenia, etc. showed reduced growth with less number leaves. The leaves were narrow, small and slightly pale of in colour. Sandal being a hemiroot parasite characters like extent shade offered by the host species, number of haustorial of connection established and synergistic or allelopathic effects arising out of the particular combination assumes significant effect on sandal growth. These effects are confirmed from the observations on mean number of leaves.

5.4 Studies on biomass

Shoot biomass is an indicator of the net photosynthesis. Similarly, the root biomass gives us some indirect idea of the extent of nutrient absorbing surface of the seedling. Both of these are directly related to survival and growth of the seedling in the long run. Biomass production and photosynthetic activity are inter-related. The sandal plants with healthy green leaves showed significantly higher biomass.

In case of shoot and root biomass, statistically significant differences were observed between various sandal host combinations. With respect to biometric observations sandal-

casuarina, sandal-albizia, sandal-terminalia, sandal-pongamia and With performed better. respect to this sandal-dalbergia sandal-delonix, sandal-ailanthus, sandal-emblica, parameter relatively sandal-psidium, sandal-swietenia, showed poor performance. Host spècies namely, casuarina, terminalia, albizia, dalbergia and pongamia provided partial shade for sandal to grow. Due to synergistic effect better haustorial connections were produced in the above mentioned species, which resulted in the better performance of sandal. This in turn was reflected in shoot length and higher biomass production. better The poor performance of other combinations like sandal-emblica, sandalswietenia, sandal-delonix and sandal-acacia could be due to the heavy shade which resulted in the restricted growth of sandal reducing the shoot length and leaf number which further thus contributed to the low shoot biomass production.

healthy root system is a pre-requisite for seedling to Α From the observation we have seen that sandal root establish. system develops well, when it grows in association with certain host species like casuarina, dalbergia and terminalia. Further, when in association with desirable hosts the number of haustorial connections also were found to increase. It is presumed that а developed sandal root system induces initiation of more well of haustoria. On the other hand extensive development number of host root system does not have any influence on production of

It is the inherent capacity of sandal to stimulate haustoria. initiation of haustoria at various points in young roots, if it association with desirable hosts. Similar results were is in by (Ananthapadmanabha et al., 1988). Characterestic reported absence of root hairs in sandal leads to a complete dependence of the host species, to get its nutrition through the sandal on significant In the investigation, connections. haustorial differences were observed in the sandal root system in the Combinations like the various host species. combination with -sandal-casuarina, sandal-dalbergia, sandal-terminalia, sandalalbizia showed a well developed root system. One of the possible for the poor performance of combinations like sandalreasons sandal-ailanthus, sandal-swietenia, sandal-psidium, emblica, sandal-delonix might be due to the underdeveloped root system of sandal with less number of haustorial connections, due to some allelopathic effects between sandal and the host species.

5.5 Anatomical study of haustorium

The study revealed that there is a great amount of variation in the degree of parasitsm by sandal. Sandal haustoria has selective power and attack some host extensively and some others sparingly. Small lateral roots of sandal when coming in contact with host roots, try to attach themselves with the haustorial connections. The success in establishing a contact depends on the disintegration of the thick cortex layer in the root of the

In some preferred combinations ultimate host plants. the association of host and sandal roots becomes so intimate that it. becomes almost an anatomical and physiological entity, enabling absorb nutrients through the roots of host plants sandal to But in certain other combinations, the haustorial I). (Plate connections were established but subsequently they were separated as in sandal-emblica, sandal-ailanthus etc. This might be one of the possible reasons for the synergistic effect of certain sandal-host combinations like sandal-casuarina, sandal-terminalia and sandal-dalbergia. In Contrary there was not much haustorial formation in case of sandal-emblica, sandal-swietenia, sandaldelonix, sandal-acacia etc. This might be probably due to the reaction of some allelopathic substances secreted by these hosts which might have resulted in the separation of haustorial connections and the poor performance of the combinations.

5.6 -Selection index

The major objective of the experiment was to indentify sandal-host combinations which could be recommended for large scale plantations. The overall performances of the different combinations have been evaluated and selections have been made from these combinations using a selection index based on the seven growth parameters discussed above.

Height of sandal seedling is an important growth parameter that has been taken into account. The experimental combinations which showed mean height more than 22 cm were given a rating 'A' which carried a weightage of 5 points. The combinations between 22 cm - 16 cm were designated as 'B' category with a weightage of 3 and less than 16 cm were classed as 'C' category with 2 points. Similarly root biomass was considered for the selections and those having a mean value of more than 0.1 g were grouped in 'A' category, 0.1-0.5 g in 'B' and less than 0.5 g in 'C' category weightage alloted for class 'A', 'B', 'C' remained the same the for all the parameter studied. In the same way based on the root length, the different treatments were classed as 'A', 'B',& 'C' categories with greater than 20 cm, between 20-10 cm and less than 10 cm respectively. Girth of the seedling was another parameter that was found to contribute substantially for the future establishment of the seedling in the field and was also considered for selection. In the selection index proposed, seedlings according to girth were grouped into 'A', 'B'& 'C' categories with greater than 0.8 mm, 0.8-0.5mm and less than 0.5 mm respectively. It has been understood that shoot biomass has a direct correlation with the future biomass hence, this parameter was also taken into account, in the preparation of the selection More than 0.3 g seedling were classed as 'A', between index. 0.3-0.1 g as 'B' and less than 0.1 g as 'C'. Number of leaves of seedling is known to contribute the increase in photosynthetic

area which in turn will lead to better establishment of seedling in the field. The groups in this respect were, seedling having leaf number greater than 25 'A', between 25-15 'B' and less than the weightage given was as above. Length of shoot 'C' was 15 also weighted in the same manner seedlings having lenght greater 25 cm 'A', between 25-15 cm 'B', and less than 15 cm 'C'. than The overall weighted mean was calculated for 16 combinations and from the selection index so prepared the six combinations were selected. The selected combinations are sandal-casuarina with weighted mean of 35, sandal-pongamia with 31, sandal-albizia with 23 and sandal-terminalia, sandal-dalbergia and sandal-azadirachta with a mean of 21 each.

The present practice adopted by the Forest Department is to sandal with azadirachta as a host. Eventhough azadirachta grow found to be a good host of sandal species like casuarina, was pongamia, dalbergia, terminalia and albizia are found to be much better hosts from the experiment. Terminalia and dalbergia, which are native forest species are found to be at par with The host species identified are azadirachta. very much indigenous to Kerala.

Bhatnagar (1965) has pointed out that selection of hosts for sandal needs careful attention because short lived plants such as shrubs, would be unsuitable, simply because their death will cause the death of sandal. Further in the selection of sandal

host, an important aspect to be considered is the economic returns obtained, not only, from sandal but the host as well.

When the above mentioned aspects are taken into considerations, the combinations identified by the present study assume importance. If sandal is planted in association with host such as casuarina, dalbergia, pongamia, terminalia and albizia etc. the net return from the plantations can be much more productive.

Treatment	_		leaves	Shoot Biomass	Root Biomass	Shoot length	Root length	Tota
Sandal + Ailanthus	С	С	С	с	С	С	в	15
Sandal + Casuarina -	A	А	A	А	A	A	A	35
Sandal (Alone)	в	С	В	С	С	В	с	17
Sandal + Albizia	В	В	в	В	A	B.	В	23
Sandal + Emblica	С	С·	С	С	С	С	С	14
Sandal + Leucaena	С	C	.C	, C	В	С	в	16
Sandal + Pongamia	В	С	В	А	А	А	в	26
Sandal + Terminalia	A	В	А	А	А	A	В	31
Sandal + Lagerstroemi	a C	В	В	В	в	В	В	20
Sandal + Dalbergia	В	В	в	в	В	В	В	21
Sandal + Swietenia	С	С	С	в	С	С	В	16
Sandal + Acacia	С	С	С	В	с	С	В	16
Sandal + Azadirachta	В	С	В	В	A	В	· C	21
Sandal + Cassia	B ·	С	в	С	с	в	С	17
Sandal + Delonix	С	С	с	В	С	` C .	С	15
andal + Psidium	с	Ċ	с	С	C	С	в	15

Table 8. Selection Index based on seven growth parameters

Summazy

6. SUMMARY

The present investigation was undertaken with view to studying the biology of sandal seedling (<u>Santalum album</u> Linn). The main objective of the experiment was to study the influence of host plants on the development of sandal and to identify, promising sandal-host combinations to be recommended and adopted for large scale plantations.

Investigation was carried out during July 1990 to March 1991 the College of Forestry, Vellanikkara. The experiment was at laid out in a Completely Randomized Design with 15 different plant species as host (treatments). The experiment was conducted in pot culture so as to avoid interference of roots of other and grasses. Fortnightly observations were made on plants the growth parameters of sandal seedling, namely height, collar girth, number of leaves, shoot biomass, root biomass, root length and shoot length. A selection index was prepared from the above growth parameters. Anatomical studies mentioned seven were carried out, to observe the development of haustoria:

Following are the salient findings of the present investigations:

 Healthy and luxuriant growth of sandal was noticed when it was grown in association with casuarina, albizia, pongamia and terminalia.

- 2. Sandal shows a remarkable degree of host specificity and it attacks some host species extensively and others sparingly.
- 3. Growth of sandal was significantly influenced by the growth habit of host plant.
- 4. Exclusively developed root system of host plants, has little influence on the production of haustoria by sandal. Whereas an extensively developed sandal root system induces more number of haustorial connections.
- 5. Initiation of haustoria is the inherent capacity of sandal and it can produce haustoria even in the absence of host species.
- 6. From the study it has been revealed that the number of haustoria produced has a direct positive relation on the growth of sandal.
- 7. It was also noticed that some hosts have synergistic effect on sandal while some others have a sort of allelopathic effect or adverse effect on sandal.
- 8. Sandal when grown in association with certain hosts showed higher growth rate with regard to height. In the present study sandal-casuarina recorded the maximum height and sandal-emblica the minimum.

- 9. There was significant difference in the increment in girth among the different sandal-host combinations. With sandalcasuarina combination recording the maximum.
- 10. When sandal was associated with favourable hosts like casuarina, dalbergia etc. the number of leaves tended to increase.
- 11. Sandal when in association with desirable hosts showed higher production in the root biomass.
- 12. Association of sandal with casuarina terminalia, dalbergia, pongamia showed increasing trend in shoot biomass as against sandal-emblica association showed a decreasing trend as the investigation progressed.
- 13. Root and shoot length of sandal was found to have higher values when it was in association with host species like casuarina, dalbergia, terminalia, albizia etc. at the same time, association of sandal with emblica, delonix, acacia, gave comparatively lesser values of root and shoot length, in sandal.
- 14. The vascular connections between the host and sandal becomes so intimate, that, host root and parasite root become almost a morphological and physiological unit, catering to the nutritional requirement of sandal.

It has been conclusively proved that there is a great amount of variation in the parasitic habit of sandal when it grows in combination with differnt hosts. Sandal grows better, with abundant leaves and green foliage, when in association with some preferred host plants, while with some others, the growth is restricted. Similarly the root system is also well developed with numerous haustorial connections when grown in association with desirable hosts.

The selection index prepared clearly indicates that among the 15 hosts used casuarina is the most suitable host species for sandal followed by terminalia, albizia, dalbergia and pongamia. On the contrary emblica, ailanthus, cassia, delonix, psidium were found to be unsuitable for sandal growth.

From the study, five very promising hosts for sandal have been selected. If these identified hosts are used, instead of azadirachta, which at present is extensively used as host for sandal by the Forest Department, sandal plantations can be much more remunerative in future.

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Appendices

Appendix I. Summary of analysis of variance of height of sandalwood seedling at different stages of development after transplanting.

		Mean sum of squares							
Source/st de	ages of evelopment	Between treatments	Within treatments						
	DAT	19.82*	3.27						
60 I	DAT	45.24*	5.80						
90 I	DAT	105.27*	11.19						
120 1	DAT	270.65*	16.76						
150 1	DAT	657.57*	40.46						

* Significant at 5% level

Appendix II. Summary of analysis of variance of collar girth of sandalwood seedling at different stages of development after transplanting.

	Mean sum of	squares
Source/stages of development	Between treatments	Within treatments
، ہے خدی ہے خان کے تی او کے بار او کر نے کا بو کے بی رو		ہے کا کے کر اور کے بین اور کو بین ہونے ہیں ہے ہے ہے
30 DAT	0.093*	0.009
60 DAT	0.100*	0.021
90 DAT	0.057	0.066
120 DAT	0.131*	0.065
150 DAT	0.330*	0.027

* significant at 5% level

	III.	sandalwood	f analysis of var seedling at t after transplant	iance of leaf number of different stages of ing.
				of squares
Source/s d	tages levelop	of - ment	Between treatments	Within treatments
30	DAT		23.07*	5.52
60	DAT		86.09*	12.43
90	DAT		213.37*	22.67
120	DAT		347.94	43.09
150	DAT		901.18*	88.94
Appendi:	x IV.	sandalwood	of analysis of var: d seedling at nt after transplant	different stages of ting.
	 ,	sandalwood developmer	d seedling at ht after transpland	different stages of ting.
	stages	sandalwood developmer	d seedling at nt after transplan Mean su	different stages of ting.
	stages	sandalwood developmer	d seedling at nt after transplan Mean su	different stages of ting. n of squares
Source/s	stages develo	sandalwood developmer	d seedling at nt after transplan Mean su Between treatment	n of squares within treatments
Source/s	stages develo DAT	sandalwood developmer	d seedling at ht after transplan Mean sur Between treatment 4.753 [*]	different stages of ting. m of squares s Within treatments 0.794
Source/s 30 60 90	stages develo DAT DAT	sandalwood developmer	a seedling at ht after transplant Mean sur Between treatment 4.753 [*] 2.773	different stages of ting. m of squares s Within treatments 0.794 1.825

* significant at 5% level

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Appendix V. Summary of analysis of variance of root length of sandalwood seedling at different stages of development after transplanting.

		of squares
ource/stages of development	Between treatments	Within treatments
	*	
30 DAT	13.153*	1.250
60 DAT	25.187*	3.641
90 DAT	41.533*	14.031
120 DAT	93.580*	17.716
150 DAT	46.867*	4.866
		ی کے ساتھ ہو جو شرقہ میں بند کر اور جو ساتھ کے اور

Appendix VI. Summary of analysis of variance of shoot dry weight of sandalwood seedling at different stages of development after transplanting.

	Mean sum of	squares
Source/stages of development	Between treatments	Within treatments
ہ ہے جارہ ہے پر پر پر پر نے نے نے پر پر پر پر پر پر اور اور		
30 DAT	0.022261	0.1632
60 DAT	0.003741*	0.000902
90 DAT	0.098052*	0.005336
120 DAT	0.157364*	0.023013
150 DAT	0.08766*	0.001846
		ہ ہے جا جا سے اور اور کر کر کر اور اور اور اور اور اور اور اور اور او

* significant at 5% level

Appendix VII. Summary of analysis of variance of root dry weight of sandalwood seedling at different stages of development after transplanting.

	Mean sum of	squares
Source/stages of development	Between treatments	Within treatments
30 DAT	0.000557*	0.000072
60 DAT	0.002621	0.002268
	*	
90 DAT	0.014578	0.005667
120 DAT	0.012773*	0.004867
150 DAT	0.007799*	0.001225

* significant at 5% level

Appendix															opment.	
Stages								Treatme	ent							
of deve- lopment	Tl	^т 2	т3	т ₄	^т 5	^т 6	^т 7	^т 8	т ₉	^{'T} 10	^T 11	^T 12	^T 13	^T 14	^T 15	∫ ^T 16
	9.20	8.81	8.46		80.08	7.90	8.10	7.29	6.87	7.24	7.32	7.64	7.45	6.55	7.22	6.90
30 DAT	ac	a	ab	a	bđ	be	bcf	defg	gh	defhi	defhj	bdh	bdhk	gijkl	defhl	defhl
	19.60	10.46	27.78	20.51	9.93	10.13	19.48	25.48	14.10	16.04	10.34	13.53	17.07	16.62	15.71	15.81
150 DAT	cj	aj	k	dl	а	ae	cdf	k	g	gj	ah	egh	cgl	fgj	cgil	cgl
Common	letter	indicat	ces no s	signific	ant dif	ferenc	e among	treatme	ents							
							C 0 ⁻	11	ما سل م							
Annondiv								llar gi:	LTU							
whhenory	IX.	Treat	tment r	neans of	sanda]	l seedl		-		lants a	at dif:	ferent :	stages	of devel	lopment	•
	IX.	Treat	tment r	neans of	sanda.	l seedl		-	host p	lants a	at dif:	ferent :	stages	of devel	lopment	
Stages of deve- lopment	 T ₁	 T ₂	 Т _З	 Т ₄	 T ₅	 T ₆	ing alo	ng with Treatme	host p		at dif: 		stages T13	of deve:	lopment T_15	 T ₁₆
Stages of deve- lopment		т ₂	т _з		 ^т 5	T ₆	ing alon T7	ng with Treatmo T8	host p ent T ₉	T ₁₀	T	^T 12	T ₁₃	T14	 T,15	T16
Stages of deve- lopment		т ₂	т _з	т ₄	 ^т 5	T ₆	ing alon T7	ng with Treatmo T8	host p ent T ₉	T ₁₀	T	^T 12	T ₁₃	T14	 T,15	T16
Stages of deve- lopment	T1 0.184 cde	T ₂ 0.276 ab	T ₃ 0.320 ac	т ₄ , 0.300	T ₅ 0.263 bfg	T ₆ 0.220 ,dgh	Ing alon T7 0.240 bdi	ng with Treatmo T8 0.175 eh	host p ent T ₉ 0.160 e	T ₁₀	T ₁₁ 0.160 e	^T 12 0.158 e	T ₁₃ 0.171 eh	^T 14 0.173 eh	^T 15 0.173 eh	T ₁₆ 0.183

Common letter indicates no significant difference among treatments

Height

Number of leaves

.

Appendix	х.	Trea	tment	means of	5 sanda	l seedl	ing alc	ng with	host p	lants	at dif	ferent	stages	of deve	lopment	•
Stages		Treatment														
of deve- lopment	Tl	т ₂	т ₃	т ₄	т ₅	т ₆	. ^T 7	т ₈	т ₉	T ₁₀	T ₁₁	^T 12	^T 13	^T 14	^T 15	T16
	12.93	10.69	10.48	11.03	9.03	9.52	9.37	9.94	9.32	9.34	10.85	9.87	10.75	9.27	9.40	9.20
30 DAT		ab	ac	a 	с	cd	с	cbl	с	с 	aef	bcfg	adegh	с	ch	bcfh
	22.53	8.67	28.56	24.16	8.52	11.57	22.56	27.10	18.48,	18.58	12.21	14.03	18.00	16.14	10.14	14.38
150 DAT	cde	ab	с	cf	a	ag	dfh	ch	di	đj	ak	bgjik	l dkm	egijk	n almno	bgkio

							Sh	oot len	gth							
Appendix	x XI.	Trea	tment	means of	sandal	seedl	ing alc	ng with	host p	plants	at dif	ferent	stages	of deve	lopment	
Stages Treatment																
or deve- lopment	Tl	т ₂	т ₃	т ₄ .	т ₅	т _б	. ^Т 7	т ₈	т ₉	^T 10	T ₁₁	^T 12	^T 13	^T 14	^T 15	^T 16
	8.7	5.0	6.4	6.9	9 . 0	7.9	8.7	8.2	5.2	7.4	7.5	6.2	5.8	6.1	6.0	7.1
30 DAT	cd	a	ab	bef	с	ceg	ch	cei	a	bgij	bdghi	k ab	afl	afjk	afjm 	bgilm
150 DAT	15.1	7.2	31.2	19.5	11.0	10.3	24.5	25.7	15.3	18.0	13.7	13.3	18.7	16.0	10.7	12.0
	bcde	a		• f	abg	abh	i .	i	bfj	cf	cghij	cghl	dfk	dfglm	aej	aejm
0	3	·	 _			e										

Common letter indicates no significant difference among treatments

Stages						•		Treatme								
of deve- opment	тı	т ₂	 Т _З	· ^T 4	т ₅	т6	^т 7	-	^т 9	$^{\mathrm{T}}$ 10	Tll	T ₁₂	^T 13	T_{14}	^T 15	^T 16
	 5.67	4.70	7.40	6.43		7.80	9.60	8.20	4.40		10.10	8.10		9.07	7.37	8.27
O DAT	acef	ab	cd	acg	achi	дj	jkl	đg	behm	am	kn	dçl		djn	dfgi	dj
·	 8.10	10.47	22.77	12.43								12.80		9.53		10.10
L50 DAT	ad	abc		bde		bfg	bhi	j	bd	efhjk	bđ	bk	bđ	cdi	cd	cdgi
	XIII			means of				Treatm	ent.							
Stages								Treatm	ent 					·		
Lopment	Tl	^Т 2	-	^Т 4	-			^т 8					^T 13			^T 16
						0.052							0.063			
	0.019	0.030	0.054	0.000									-	a		a
	0.019 a	0.030 a	0.054 a	a	a	a	a .	a	a 	a 	a 	a 	a 		• a . 	
0 DAT		a	a 		a 						a 0.135		0.183			

Root length .

								Treatme	nt							
Stages of deve- Lopment	 T _l	 T ₂	 ^Т з	 T ₄	т ₅	т _б	 ^Т 7	т ₈	 Т ₉	T ₁₀	T ₁₁	T ₁₂	T ₁₃	T ₁₄	T ₁₅	т <u>1</u> 6
 \$	0.014	0.008	0.012	0.030	0.038	0.028	0.041	0.049	0.016	0.034	0.058	0.025	0.031	0.022	0.021	0.03
0 DAT	b	a	ab	cd	efg	ceh .	f		b	dg		ci	dhj	i	j	gj
	0.016	0.042	0.159	0.130	0.014	0.062	0.124	0.164	0.083	0.088	0.015	0.043	0.109	0.042	0.028	0.04
50 DAT	aef	abc	đ	deq	ah	bji	dk	đ	bklm	gikl	cfh	cfhjm	gk	cfhjm	cfhj	cfhj

Common letter indicates no significant difference among treatments

Root dry weight

BIOLOGY OF THE SEEDLING OF SANDAL WOOD

(Santalum album Linn.)

By

YAYATI B. TAIDE

ABSTRACT OF THE

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SUBMITTED IN PARTIAL FULFILMENT OF THE REQUIREMENT FOR THE DEGREE

MASTER OF SCIENCE IN FORESTRY KERALA AGRICULTURAL UNIVERSITY

FACULTY OF AGRICULTURE COLLEGE OF FORESTRY VELLANIKKARA - THRISSUR 1991

ABSTRACT

An experiment was conducted in the College of Forestry, Vellanikkara during July 1990 to March 1991. The investigation undertaken with a view to studying the biology of sandal was album Linn.). The main objective of seedling (Santalum the experiment was to study the influence of host plants on the growth and development of sandal and to identify promising sandal- host combinations to be recommended and adopted for large scale plantations. The experiment was laid out in a Completly Randomised Design with 15 host species as treatments.

The results revealed that sandal has host specificity and with certain host species sandal tended to put up better growth. Casuarina was found to be the best host among the 15 host species tried in the experiment. All the growth characters like height, collar girth, number of leaves, root and shoot biomass, root and shoot length were higher when sandal was associated with host species like casuarina, terminalia, albizia, dalbergia, pongamia. On the other hand some species like emblica, delonix, acacia, ailanthus, lucaena had an antagonistic or allelopathic effect. A multitrait selection index was developed for the 15 host species tried in the investigation and five sandal-host combinations have been identified for recommendation for large scale sandal plantations.