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Sandalwood Research Newsletter

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EDITOR'S NOTE

Farewell from Tanya Vernes as editor of SRN

After nearly five years with the Department of Conservation and Land Management in Western Australia I have taken up new employment with the World Wide Fund for Nature Australia. Having resigned from my position as Research Scientist on sandalwood and tropical forestry, I will no longer be editor of the SRN, and hand that responsibility over to Jon Brand who is a Research Scientist working on native sandalwood research in southern Western Australia. It has been a great experience as Editor of the SRN and I have enjoyed the contact I have had with researchers, land managers and community involved in sandalwood conservation and management. I have also been inspired by the work that is being carried on around the globe for improved management of sandalwood as a tree crop and also as a species in need of protection. The link between poverty, statutory laws and conservation of species is reinforced in the articles that have been published in the SRN to date.

This issue is the first for Jon Brand as the new editor of the SRN and he will continue to provide a quality forum for global sandalwood issues into the future. I would like to thank all those who have contributed to the newsletter to date and provided me with the feedback necessary to improve the newsletter and widen our readership. I also wish all our readers the best of luck with sandalwood research, management and conservation in the future. Keep up the good work!

Kind Regards

Tanya Vernes

Introduction from the new editor

Welcome to Issue 16 of the Sandalwood Research Newsletter. My name is Jon Brand, and I have a keen interest in sandalwood, having worked as a researcher in this field since 1991. I am delighted to take over as editor and I hope to bring you interesting articles from people studying sandalwood throughout the world.

In this issue, the three articles focus mainly on Indian sandalwood – Santalum album. Vernes and Robson give an account of the S. album industry in Australia. They present information on current levels of plantation establishment in Australia and predicted levels over the next ten years. They also forecast world demands for the valuable timber. Tue Man Luong compares S. album seedling performance when grown in pots with one, two, three and four host seedlings from the species, Alternanthera dentata. Angadi, Rajeevalochan, Ravikumar and Shankarayana also report on Peroxidase reagents that were used to identify S. album trees with high oil percentages in India.

Jon Brand

Indian Sandalwood Industry in Australia

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Introduction

Expansion of the Indian sandalwood plantation resource has been occurring in northern Australia over the past few years. The current annual level of investment is expected to make a significant contribution to the global sandalwood industry by maintaining a stable supply of a resource that is otherwise declining in its natural distribution. Opportunities are still expected to exist for further expansion of plantation grown sandalwood by both private companies and small-scale growers. This article gives a brief review of the current private resource of *Santalum album* in northern Australia and suggests what future influence it may have on the global market.

Utilization of sandalwood from native stands

Australia has been exporting sandalwood almost as long as its western existence. Just 15 years after its foundation in 1788, the first sandalwood was traded for tea in China (Statham 1990). Since then, the trade has made a significant contribution to Western Australia's economy and the native species, *Santa-*

lum spicatum has provided the basis of a small but important export industry (Shea *et al* 1998). Between 1892 and 1901, more than 50,000 tons of sandalwood was exported from Western Australia (Statham 1990). Today's harvest is currently about 2000 tonnes per year (Jones 2001). Considering that demand for sandalwood oil and products has not declined over the past century (Berney 1999), this puts supply potential in perspective.

In Western Australia, 49% of the present distribution of *S. spicatum* is protected from any form of harvesting. Government initiatives to create more conservation reserves will see protected areas increase over time while simultaneously maintaining the sandalwood industry by increasing plantation based resources (Jones 2001).

A long history of sandalwood trading exists in Queensland as well and *Santalum lanceolatum* from the north-western parts of the state has provided the basis for a small industry, which began in the 1920's. "*The Sandalwood Act of Queensland*" invoked by the state government in 1934 prohibited any harvesting without a licence. A monopoly licence was granted to the Australian Sandalwood Company (Western Australia), however harvesting declined and was effectively halted (Applegate *et al*, 1990). The revoking of the act in 1982 and introduction of a quota system has seen the gradual revival of the industry. Currently, the Queensland Department of Primary Industries - Forestry has imposed a sustainable harvest limit of 500 tonnes per year. Although it is envisaged that this policy will continue, there is increasing pressure from environmental groups to reduce the yearly harvest. An associated native forest management project developed by the Queensland Forestry Research Institute (QFRI) is investigating the ecology, growth and yield of *Santalum lanceolatum* in north-western Queensland.

Global supply and demand

The gap between sandalwood supply and demand has widened over time. During1970-1995, there was a decline in sandalwood heartwood and oil production in India, Indonesia, Fiji and Vanuatu due to overexploitation and lack of protection of the resource (Sekhar *et al* 2000, Radomiljac *et al* 1998). As production dropped, there was a simultaneous increase in price (Radomiljac 1995). In some areas of India, the rate of increase of sandalwood and oil prices has been exponential (Radomiljac 1995, Baruah 1999). Prices in Fiji for *S. yasi* followed a similar trend during 1985-1990 (Jiko 2000). Native stands of sandalwood are diminishing in India and it is estimated that India will only be able to maintain its current harvest of 1000 tonnes annually for the next 5 to 8 years (Anantha Padmanabha pers. comm.)

The world market demand is thought to stand at around 5000-6000 tonne (Radomiljac *et al* 1998). This figure incorporates demand for a number of different products, sourced from a variety of *Santalum* species.

Future demand will depend on many variables including the quality of the end product and the market specifications at the time. Synthetic substitutes are already present in the market but at a reduced cost compared to natural sandalwood oil. Some industries have been forced to obtain synthetic compounds where supply of natural sandalwood was not available (Anantha Padmanabha pers comm.).

> "The gap between sandalwood supply and demand has widened over time."

There is also the possibility of expansion within India due to recent changes in state regulations in Karnataka and Tamil Nadu providing incentives for large scale private plantation development, expected to boost the production levels of *S. album* from India in the future (Anantha Padmanabha pers comm.).

Indian sandalwood expansion in northern Australia

The last three years has seen a rapid expansion in Indian sandalwood (*Santalum album)* plantation development in the Ord River Irrigation Area (ORIA) of north Western Australia. The Department of Conservation and Land Management has been growing sandalwood since the early 1980's, and since 1993 the Department has had a research scientist investigating the silvicultural requirements of *S. album* in a flood irrigated system. These efforts coupled with industry enthusiasm saw the first private plantation of *S. album* established in the ORIA in 1998. Since then, a number of private companies and local landholders have been planting sandalwood on an increasing scale each year. There is also interest in growing sandalwood and other tropical trees on partially irrigated sites in the West Kimberley, although there is only one pilot plantation (6 ha) of *S. album* to date.

Currently, the area of *S. album* plantations is greatest in northern Western Australia where the resource currently stands at approximately 830 ha with an expected increase of 120-150 ha per year over the next ten years. This gives a total expected resource in 10 years of 2000-2300 ha for the ORIA alone. The current Northern Territory and Queensland plantation resource combined is less than approximately 55ha. In Western Australia, private companies have factored in an age of 15 years for the main harvest.

Queensland currently has no commercial sandalwood plantations. However, there is substantial interest by both private and investment companies in growing *S. album* in plantations. There has been some investment in a number of small research plots in south western and north Queensland. These research trials, conducted by QFRI, are investigating nursery silviculture and seed germination. Field trials are looking at species by host interactions, growth under natural and irrigated conditions, as well as a number of other silvicultural treatments such as open and under planting. QFRI has developed protocols for the clonal propagation of selected materials for several of the sandal species, including *Santalum album*. The development of propagated material from superior clones should encourage further expansion of commercial sandal plantations in the near future. By gauging current interest and anticipated future interest it can be estimated that a plantation industry will be established in Queensland within the next two years, with slow but increasing development over the next ten years.

Currently, the *S. spicatum* industry is worth approximately AUD$10 million

State	Current plantation resource 2001 (ha)#	Expected increase over 10 years (ha/yr)#	Predicted total 2011 (ha)
Western Australia	830	120-150	2030-2330
Northern Territory	35	Unknown	Unknown
Queensland	< 20*	50	500

* research trials

Figures supplied by Tropical Forestry Services, Integrated Tree Cropping, Exotic Timbers of Australia, Queensland Forestry Research Institute.

per annum (Forest Products Commission 2001). The WA Government has entered into a contract with a private company, Mt Romance to supply up to 1000 tonne of wood per year for the next ten years, which will be supplied from natural stands (Mt Romance 2001). In southern Western Australia *Santalum spicatum* is being trailed as a new tree crop within the wheatbelt region of Western Australia. Trials to date indicate that a stocking rate of 200-300 stems per hectare should yield two to three tonnes of commercial timber per hectare at age 20 years (Brand & Jones 1999).

Expected production levels of Australian plantations

The Department of Conservation and Land Management in conjunction with the Forest Products Commission is currently investigating the heartwood and oil content of trees of various ages and under different silvicultural regimes in research plots in the ORIA. However, there is no information yet available from this data on average growth rates or heartwood production. Private companies in the ORIA anticipate 27-38kg heartwood production per tree at age 15 years in their prospectus (East Kimberley Sandalwood Project No 1 Prospectus 1998, Ord River Sandalwood No 2 Prospectus 1999, TFS Sandalwood Project Prospectus 2000). Taking their mean figure of 32.5kg heartwood per tree and a stocking rate of 460 stems per hectare with a rotation length of 15 years the yield is 14.95 t per ha. Under these extrapolations, a plantation resource of around 335 ha per yr would be required to meet a global sandalwood demand of approximately 5000 tonnes. The annual planting program in northern Western Australia could only supply less than half of the current demand on proposed expansion rates.

Although there is some interest in the establishment of a plantation resource in Queensland, investors are treading cautiously. It is impossible to predict the expected or proposed plantation area. It can be assumed that any planting over the next ten years will be reasonably small.

However, the global demand for sandal may be greater than the current level of 5000 tonne. This figure may be reflecting the scarcity of the resource and masking any potential for expansion. It is also difficult to determine the annual demand due to the illegal trade of sandalwood occurring in many countries, and the large amount of stockpiled wood which merchants release when prices are high (Anantha Padmanabha pers. comm.). Thus, determining the potential for expansion will require more in depth marketing research than the information presented in this article.

Opportunities for small growers

The global demand of sandalwood coupled with its high prices and recent improvement in plantation technology has been a catalyst for the recent plantation in Western Australia. Large-scale growers have the advantage of being able to supply a constant amount of harvested wood, giving them some influence in price determination. Small-scale growers operating in isolation do not have this advantage. The task of gaining reliable information in global supply trends and prices is very difficult, and anecdotal evidence suggests buyers operate in a fragmented market, which allows them to maintain an advantage. To overcome this problem, small-scale growers within and across national boundaries must be organised to achieve a similar advantage. A coordinated sales approach, such as a cooperative, would assist individual sellers by strengthening their bargaining power.

Measures for small-scale growers to ensure high returns for their efforts are to maintain a silvicultural regime to promote good growth such as early pruning of sandal and hosts. In the future, planting genetically improved stock with characteristics such as early heartwood initiation or the development of high oil yielding heartwood are also likely to improve the returns from plantations.

Conclusion

The Indian sandalwood industry in Australia is still in its early stages. There are many unknowns in irrigated plantation development, which are currently being investigated, such as improved provenances, irrigation rates and host tree species. Expansion of the Australian sandalwood industry seems certain to continue in the near future, for both *S. album* and *S. spicatum*. However, expansion is not likely to flood the current market on the present development rates. More positively, plantation development is contributing to the conservation of species under threat in their natural environment and the sustainability of a global industry.

The work above has also been published in the PIFT Newsletter:
Vernes, T. and Robson, K. (2002). Indian sandalwood industry in Australia. Pacific Islands Forests and Trees Newsletter 4/01, 9-12.

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Competitive effects within and between *Santalum album* and pot host *Alternanthera dentata*

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The growth of Santalum album seedlings and the preferred pot host Alternanthera dentata under nursery conditions is the first important step in establishing this species in plantations. A 19 week pot trial was conducted in a glasshouse at Curtin University of Technology, Perth, Western Australia. The aim was to test whether an increase in host density improved growth of sandalwood seedlings. S. album seedlings had a tendency to grow better at lower densities of A. dentata (one or two hosts per pot), compared with higher densities (three or four hosts per pot). Seedlings with two hosts had greater heights, dry root and shoot weights and leaf area, while seedlings with one pot host had more leaves. There were no clear trends between number of haustorial connections made as host density increased. As host density increased, the leaf area, root and shoot weights of A. dentata declined. Both parasite and host were more affected by competition, however the host was more affected by intraspecific competition, indicated by large competitive responses to each other. S. album seedlings had less effect or response to density of A. dentata after 19 weeks, perhaps due to not being limited by the same resources as the host at this early establishment phase.

Introduction

Successful establishment of *Santalum album* L. in plantations requires the utilisation of appropriate pot hosts under nursery conditions (Radomiljac 1998). Early nursery growth of *S. album* is more rapid and secure when grown with a pot host (Fox 2000). The pot host: parasite relationship continues after plantation establishment and should persist until a second (intermediate) host plant is parasitised (Radomiljac 1998).

Alternanthera is a herbaceous, perennial ornamental shrub and has shown to be an excellent pot host (Fox *et al.* 1996). Various cultivated forms of *Alternanthera* can be planted as cuttings simultaneously with *S. album* (Fox 2000), and this has been adopted in Timor (Fox *et al.* 1995). *A. sessilis* (L.) DC. is known to promote good growth of *S. austrocaledonicum* in New Caledonia (Fox *et al.* 1995). In northern Western Australia, the herbaceous *A. nana* has been reported as an ideal host as it promotes high *S. album* survival and growth following field establishment (Radomiljac, McComb & McGrath 1999).

This paper examines the early growth of *S. album* seedlings with *Alternanthera dentata* (Moench) Scheygr. as a primary host. It specifically aims to determine whether an increase in the density of *A. dentata* improves or reduces early growth (height, leaf numbers, root and shoot mass, number of haustoria and attachments to host roots) of *S. album* seedlings.

Experimental Procedure

Experiments were conducted under a shaded glasshouse in the Field Trial Area at Curtin University of Technology, Perth, W. A. *S. album* seeds were sown in punnets filled with sterilised coarse sand and placed on 25°C heat beds. Moisture was provided five days per week. Germinants were transplanted into rectangular pots (9 x 9 x 15 cm) filled with one part coarse sand: one part fine sand: one part peat, with fertilisers.

Ten randomly selected *S. album* seedlings were grown at four levels of host densities (one, two, three or four per pot). Seedlings were 19-32 days old, between 4-13 cm in height, and had 3-6 leaves. A one-way analysis of variance confirmed no significant difference between *S. album* seedlings (height: $p=0.748$; leaves: $p=0.121$). The host plants used were nine-day-old, rooted *A. dentata* 'Ruby' cuttings which were introduced to seedlings. Seedlings were watered five days per week with rainwater. Pots were placed in a randomised block design and rotated weekly.

Height (cm) and leaf numbers of *S. album* and *A. dentata* seedlings were measured initially and weekly for 19 weeks, from December 2000 to April 2001. An initial harvest of one ran-

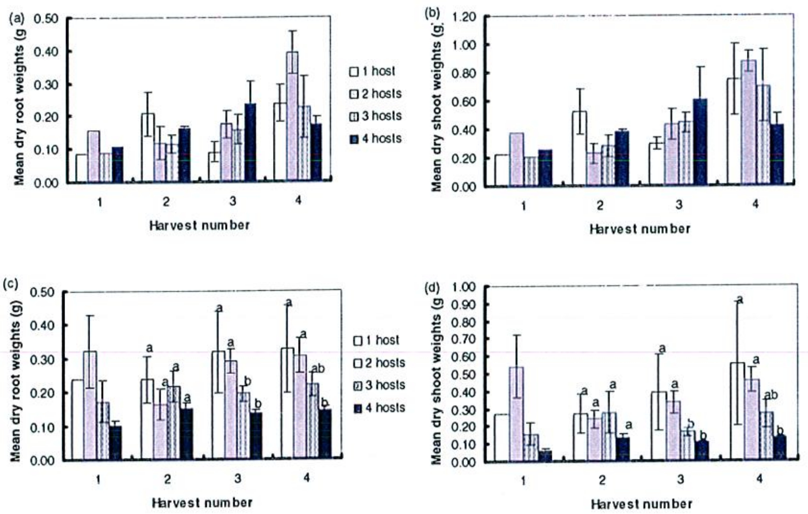

Figure 1. Mean dry root weights of (a) *S. album* and (c) *A. dentata*; and mean dry shoot weights of (b) *S. album* and (d) *A. dentata* at harvest (1=8 wk, 2=12 wk, 3=16 wk and 4=19 wk). SE bars shown. Similar letters indicate no significant differences (a=0.05) using Fisher's LSD test.

domly selected *S. album* (and host) from each treatment was taken at eight weeks. At subsequent harvests, three seedlings were randomly selected at 12, 16 and 19 weeks.

Harvest measurements recorded were:

1. Fresh shoot and root weights (g) of *S. album* and its host.
2. Dry shoot and root weights (72 hr at 60°C) of *S. album* and its host.
3. Leaf area (cm^2) of *S. album* and its host using an electronic planimeter (Otfoto 1.1 and NIH Image 1.62).
4. Number of *S. album* haustoria (at least 0.05 mm).
5. Number of haustorial connections on host roots.

Results

Differences in *S. album* heights were not apparent until after 12 weeks of growth with *A. dentata*. At 19 weeks, seedlings grown with two hosts were the tallest, followed by those growing with one, three and four hosts, with mean heights of 22, 21, 20 and 16 cm respectively. However, there were no significant differences over the whole trial.

Similar trends were observed in the number of *S. album* leaves present at 12 weeks, with seedlings grown at increasing densities of *A. dentata* having a lower mean leaf number, although differences were not significant. At 19 weeks, *S. album* grown with one host had the highest mean leaf number (20), compared with seedlings grown with two, three and four hosts (15-18 leaves).

Initially, there were no clear trends as to whether *S. album* root growth was influenced by the density of its pot host. At 16 weeks, there was a tendency for root growth of *S. album* seedlings to be greater as pot host density increased. However, by 19 weeks, a different trend was observed with *S. album* seedlings growing with two hosts having the best root growth, with a mean dry root weight of 0.39 g (Figure 1a). However, there were no significant differences.

Similar trends were observed in dry shoot weights of *S. album* (also not significant), with seedlings growing with two *A. dentata* hosts having the greatest mean dry shoot weight of 0.87 g, after 19 weeks of growth (Figure 1b). Leaf area of *S. album* seedlings followed the same pattern as dry shoot weights (no significant differences). *S. album* seedlings grown with two pot hosts had the greatest mean leaf area of 88 cm^2 at 19 weeks. In comparison, *S. album* seedlings grown with one, three and four hosts had mean leaf areas of 81, 68 and 44 cm^2 respectively.

Dry root and shoot yield of the pot host *A. dentata* appeared to be more affected by the density at which it was grown compared with *S. album* seedlings. Generally, there was a decrease in mean dry root and shoot weights as density increased. Dry root weights were significantly different at 16 weeks (p=0.001) and 19 weeks (p=0.011), with *A. dentata* grown singly with *S. album* seedlings having the heaviest mean dry root weights of 0.33 g at 19 weeks (Figure 1c). Similarly, there were significant differences in mean dry shoot weights at 16 weeks (p=0.002) and 19 weeks (p=0.012), with *A. dentata* grown singly with *S. album* seedlings having the heaviest mean dry shoot weights of 0.55 g at 19 weeks (Figure 1d).

A. dentata grown at greater densities had lower mean leaf areas, with significant differences present as time progressed at both 16 weeks (p=0.000) and 19 weeks (p=0.025). At 19 weeks, *A. dentata* grown singly had mean leaf area of 48 cm^2, compared with those grown with two, three or four hosts per pot (means of 45, 31 or 17 cm^2 respectively).

Haustoria began to form on roots of *S. album* seedlings grown with two, three and four hosts at eight weeks. At this

Table 1. Mean number of haustoria formed on *S. album* roots and mean number of haustorial connections with roots of *A. dentata* (± SD) at harvest 1= 8 wk (n=1); harvest 2= 12 wk (n=3); harvest 3= 16 wk (n=3); and, harvest 4= 19 wk (n=3). Significance tested using one-way ANOVA (a=0.05) and Fisher's LSD test (similar

	Haustoria numbers				Haustoria connections			
Week	8	12	16	19	8	12	16	19
1 host	0.0	15.3	18.3 c	18.3	0.0	1.7	1.7	4.3
	± 0.0	*± 12.5*	*± 16.3*	*± 18.3*	*± 0.0*	*± 2.9*	*± 2.1*	*± 7.5*
2 hosts	14.0	12.0	50.0 ab	33.7	2.0	0.0	4.0	7.0
	± 0.0	*± 2.6*	*± 8.9*	*± 18.6*	*± 0.0*	*± 0.0*	*± 5.2*	*± 6.1*
3 hosts	3.0	20.7	18.7 bc	22.7	0.0	3.0	5.7	9.7
	± 0.0	*± 2.9*	*± 2.3*	*± 27.2*	*± 0.0*	*± 2.6*	*± 4.0*	*± 10.7*
4 hosts	10.0	14.3	43.3 a	33.7	3.0	2.7	5.3	2.3
	± 0.0	*± 7.5*	*± 14.0*	*± 41.0*	*± 0.0*	*± 2.1*	*± 0.6*	*± 1.5*
F		0.71	5.98	0.235		1.11	0.82	0.58
P		0.574	0.019	0.869		0.401	0.516	0.643

time, only seedlings growing with two and four hosts began to form haustorial connections with the roots of *A. dentata.* By 12 weeks, all seedlings harvested had formed haustoria and haustorial connections. There were no significant differences in the amount of haustoria formed on *S. album* roots, regardless of the number of hosts present. However, at 16 weeks, seedlings growing with two hosts had a significantly higher number of haustoria (mean of 50), compared with those grown with one, three or four hosts (Table 1).

Discussion

A major factor influencing growth and survival of individual plants is competition from neighbours (Firbank & Watkinson 1985). The ability of a species to persist and prosper in a community is often determined by its competitive interactions with other species (Miller & Werner 1987). The growth of *S. album* seedlings was affected by varying densities of its host plant, although these differences were not significant. For the first eight weeks, host plant density had no affect on growth of *S. album.* Between 12 and 16 weeks, growth of *S. album* seedlings was greater as host plant density increased. The effects of competition were evident after 19 weeks, with *S. album* seedlings grown at lower host densities having attained better growth. It was more beneficial to grow *S. album* with two hosts. These provided greater shoot and root biomass, and presumably sufficient resources for greater height and leaf production. Seedlings grown with four hosts appeared to be restricted in growth. This was exaggerated by growth in close proximity and within a small pot environment. It is suggested that a constant increase in the biomass of competitors does not necessarily mean a constant decrease in the amount of limiting resources available to the individual plants. Initially when seedlings first emerge, there may be few competitive interactions. As the individuals grow, they occupy more space and require a greater amount of resources, thus the intensity of competition increases (Miller & Werner 1987).

In *Orobanche crenata,* the number of haustoria formed is affected by the density of host roots in the soil. It is suggested that there may be a positive association between host root weight and parasite weight, due to the increased probability of an attachment forming in a plant with a larger root system. The probability of forming attachments with hosts will differ due to the distribution of host roots in the soil, the susceptibility of the host root to formation of functional haustoria and the presence of host selection mechanisms in the parasite (Graves 1995). However, the increase in density of pot host generally did not influence the numbers of haustoria formed in *S. album* seedlings (except week 16) or number of haustorial connections made with the host.

Distances between individuals and differences in growth form and sizes will strongly influence the competitive effects and responses of species in a plant community (Miller & Werner 1987). Intraspecific competition can cause a reciprocal relationship between mean yield per plant and density (Firbank & Watkinson 1985). In this trial, the host *A. dentata* was more affected by competition within the same species, with significant differences in leaf area, root and shoot biomass at later stages of growth (16 to 19 weeks). Generally, as density increased, biomass of the host plant decreased. Thus, growing *S. album* seedlings with four hosts is not ideal as it not only reduces biomass, but the host plants cannot grow well to support the *S. album* seedlings, due to intraspecific competition.

Throughout the trial, *S. album* seedlings were less affected by competition than the host, *A. dentata.* It is suggested that successful species have either a low response to the abundance of other species and/or such a large effect that the abundance of other species is greatly reduced. Species that have little effect on and response to other species in the community are probably not limited by the same resources and will not be affected by changes in the abundance of other species in the community (Miller & Werner 1987). This could possibly explain the non-significant differences between *S. album* seedlings grown at various host densities. If species have both a large competitive effect on and a large competitive response to one another, the plants are possibly limited by the same resources (Miller & Werner 1987). This was observed between *A. dentata* plants in this trial.

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A note on Peroxidase reagents to distinguish between high and low yielders of Sandal (*Santalum album*) in the field

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A simple, less expensive peroxidase colour reaction has been developed to distinguish high and low yielders of Sandal in the field.

Peroxidase (donor: H_2O_2-Oxidoreductase, EC 1.11.1.7) enzyme which catalyses dehydrogenation of a large number of organic compounds such as Phenols, aromatic amines and hydroquinones has often been used as a marker enzyme by Plant Scientists (Van Huystee and Cairns 1980). In Sandal (*Santalum album* L), the enzyme peroxidase present in living bark tissue was used as a marker for oil bearing capacity (Parthasarathi *et al.* 1986) of the progeny.

Sandal (*Santalum album* L) is a small evergreen tree popularly known for its scented heartwood and oil content. In breeding studies or in selection of Candidate Plus Trees (CPTs) for Clonal Seed Orchards (CSOs), oil is one of the important criteria. Earlier methods of estimating oil content in sandalwood by steam distillation in Clevenger's apparatus and by rapid method (Shankaranarayana *et al.* 1997) were time consuming and non-field oriented. In view of this, an attempt was made to develop a simple field method to distinguish high and low yielders of sandal by modifying and simplifying the earlier methods (Parthsarathi *et al.* 1986).

For this purpose, twelve sandal plants (six high yielders - containing > 3% oil; and six low yielders - containing < 3% oil) were selected from among the experimental forest Institute of Wood Science and Technology, Bangalore. Twig samples (0.5 to 0.8 cm diameter) were taken from each plant and thin bark layers were peeled off and outer dead bark portions scraped off to get the living bark tissue. In each case, the living bark of 5 x 0.2 cm (1 x b) were selected: cut into small bits and placed into 50 ml conical flasks containing peroxidase reagents to study the colour reaction.

The following two peroxidase reagents were used to develop colour reaction:

a. Guaiacol peroxidase Reagent (GPR)

2ml of 1% Guaiacol in 50% ethanol + 2ml of 1% Hydrogen Peroxide + 6ml of distilled water – shaken well occasionally and allowed to develop for 15 to 20 minutes. Light brownish yellow developed for high yielders and reddish brown developed for low yielders.

b. Benzidine Peroxidase Reagent (BPR)

2ml of 1% Benzidine in 25% acetic acid + 2ml of 1% Hydrogen Peroxide + 6ml of distilled water – shaken well occasionally and allowed to develop for 30 to 45 minutes. Light brownish pink developed for high yielders and dark brownish pink developed for low yielders.

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The above observations were further confirmed by diluting 1 ml of each coloured solution with 5 ml of distilled water. In case of GPR, an almost colourless solution developed for high yielders, while the colour persisted for low yielders. For BPR, a similar pattern occurred for high and low yielders.

This method is simple, user friendly, less expensive and field oriented which could be useful for rapid screening of sandal plants in the field for selection of high yielders in breeding studies or in general use.

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