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GC-MS ANALYSIS OF ESSENTIAL OIL OF SHOREA ROBUSTA BAST

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GC-MS analysis of the essential oil of *Shorea robusta* bast (cambium + secondary phloem) has revealed the presence of twenty-eight compounds, of which nine compounds, constituting 48.79% of the oil, were identified as T-cadinol (16.75%), α -cadinol (16.45%), globulol (4.52%), α -copaene (3.79%), γ -cadinene (2.34%), viridiflorene (1.62%), β -elemene (1.54%), α -terpineol (1.33%) and γ -muurolene (0.45%). This is the first report on the volatile constituents of the bast which may be significant in influencing host location for *Hoplocerambyx spinicornis*, the most injurious heartwood borer of *Shorea robusta*.

Keywords: Shorea robusta; Bast; Volatile oil; Sesquiterpenoids

INTRODUCTION

Shorea robusta, Gaerti (Dipterocarpaceae) commonly known as "sal" is the most abundantly available timber yielding tree in India. Sal, a large sub deciduous tree and seldom quite leafless, is grown extensively in parts of North, East and Central India. Sal forests occupy 16.70% of the total forest area of the country. It is a multipurpose tree as its bark, resin, leaves and the seeds, available in considerable quantities, are exploited for their varied applications, including medicinal [1–4]. It also occurs in Ceylon, Burma and other South–East Asian countries. *Hoplocerambyx spinicornis* (Coleoptera: Cerambycidae) is considered to be the most injurious heartwood borer to the tree. Three outbreaks have occurred, in 1923, 1950 and 1997, in India during the last century. The last epidemic ravaged major areas of sal forests.

Volatiles released by the host tree are attractive to the borer [5]. Observations suggested the presence of these volatiles in the bast (cambium + secondary phloem), which when exposed broadcasts its attraction on all sides [6]. The orientation of phytophagus insects to their host plants involves perception of a variety of stimuli [7]. Olfactory stimuli play a major role in this process [8]. The identification of the volatile constituents of a host plant's odour is, therefore, an important step in the investigation of insect–plant interactions. There have been a few reports of volatile compounds produced by the tree [4,9]. Thus, studies on volatile

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constituents of different parts of the tree have been initiated by us. Recently, we have reported the essential oil constituents of heartwood and resin [10], and leaves [11]. The present study analysed the volatile constituents of the bast to search for olfactory cues that possibly determine the borer's response to the volatiles.

RESULTS AND DISCUSSION

GC-MS of the oil showed it to be a mixture of twenty-eight compounds, of which nine compounds, amounting to 48.79% of the oil, were identified (Table I). Nineteen compounds, constituting 51.16% of the oil, were unidentified. The identified compounds were sesquiterpenes (47.46%), except α -terpineol (1.33%). In the GC-MS, peak 3 represented α -terpineol (1.33%). Peaks 5, 6 and 8–10, amounting to 9.74% of the oil, were found to be sesquiterpene hydrocarbons and were characterised as α -copaene (3.79%), β -elemene (1.54%), γ -muurolene (0.45%), viridiflorene (1.62%) and γ -cadinene (2.34%), respectively. Oxygenated sesquiterpenes (peaks 14, 17 and 18) constituted the major portion of the oil (37.72%) and were identified as globulol (4.52%), T-cadinol (16.75%) and α -cadinol (16.45%), respectively. The data indicate that T-cadinol is the chief constituent of bast oil.

Both mono and sesquiterpenoids are known to be present as the volatile components of a plant's emitted odour [12]. These volatiles are important in influencing the host location for numerous phytophagus insects [13,14]. A number of mono [12,15,16] and sesquiterpenoids [17,18] and their oxygenated derivatives [19-21] have been shown to be effective attractants

Peak no.	Compound	Retention time (min)	Area (%)	KRI	Ident.
1	u.i.	4.58	4.79	718	_
2	u.i.	12.78	2.72	926	-
3	α-Terpineol	19.28	1.33	1196	MS, KI
4	u.i.	20.63	7.41	1264	-
5	α-Copaene	22.99	3.79	1392	MS, KI
6	β-Elemene	23.18	1.54	1404	MS, KI
7	u.i.	24.69	1.32	1492	-
8	γ-Muurolene	24.76	0.45	1496	MS, KI
9	Viridiflorene	25.08	1.62	1516	MS, KI
10	γ-Cadinene	25.45	2.34	1539	MS, KI
11	u.i.	25.85	1.37	1564	-
12	u.i.	26.33	0.43	1594	-
13	u.i.	26.48	6.99	1604	-
14	Globulol	26.61	4.52	1613	MS, KI
15	u.i.	27.03	1.87	1640	-
16	u.i.	27.23	1.46	1653	-
17	T-Cadinol	27.43	16.75	1666	MS, KI
18	α-Cadinol	27.63	16.45	1680	MS
19	u.i.	27.71	1.18	1685	_
20	u.i.	27.78	0.73	1690	-
21	u.i.	28.03	0.79	1707	-
22	u.i.	28.3	0.65	1725	-
23	u.i.	29.1	0.69	1781	-
24	u.i.	29.43	1.27	1804	_
25	u.i.	30.38	3.89	1873	_
26	u.i.	30.46	3.85	1879	-
27	u.i.	30.75	8.38	1900	-
28	u.i.	31.79	1.37	1978	_

TABLE I Composition (%) of essential oil of Shorea robusta bast

Total number of compounds identified 9 (48.79%). Total number of compounds unidentified 19 (51.16%). KRI = Kovats Retention Index; Ident. = Identification; MS = Mass Spectrum; u.i. = unidentified.

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to several phytophagus insects of Coleoptera. Further, monoterpene alcohols (*e.g.* (+)-*cis*-3-pinen-2-ol) and sesquiterpene alcohols (*e.g.* (+)-juniperol) have been reported to be highly attractive to the Cerambycid beetle (*Monochamus alternatus*), and have, therefore, been considered particularly important olfactory cues in the host location for the Cerambycid beetles [22,23].

Since the two sesquiterpene alcohols, namely T-cadinol and α -cadinol, constitute the major portion (37.72%) of the bast oil, these alone or in combination with other identified or unidentified constituents of the oil may possibly be involved in influencing host location for the heartwood borer.

Thus, the interaction between volatiles of the bast of *Shorea robusta* and *Hoplocerambyx spinicornis* may, very probably, involve specific compound blends or individual compounds as the important cues in influencing the host location for the borer; this is an interesting area for further investigation.

EXPERIMENTAL

General Experimental Procedures

GC-MS analysis of the bast oil was carried out on a HP 5890-Trio-1, equipped with a fused silica capillary column (BPX-5, $30 \text{ m} \times 0.32 \text{ mm}$; $0.25 \,\mu\text{m}$ phase thickness). Chromatographic conditions: helium carrier gas at a flow rate of $1-2 \text{ ml min}^{-1}$ and a head pressure of 10 psi; oven programme: 30° C for the first 5 min, 8° C min⁻¹ to 300° C and then held for 10 min. The column was coupled directly to the quadrupole mass spectrometer operated in EI mode at 70 eV, *via* an interface at 280°C. Mass spectra (*m*/*z* 33–650) were accumulated at the rate of 1 s^{-1} , with an acquisition time of 0.9 s, and an interscan time of 0.1 s. Data were collected and analysed using LAB BASE software, working under Quaterdeck extended memory management on a Dell 33 MHz, 80486 computer. Spectra were matched with the NBS library using the eight most abundant peaks and ranked according to reverse fit. The components were identified by comparing their mass spectra and Kovats retention indices (based on a series of n-hydrocarbons) with those from authentic compounds and with the published data [10,11,24–28].

Experimental Biological Material

A freshly felled log of healthy *Shorea robusta* was obtained from a wild population of sal forests growing in the Thano forest range of Dehra Dun in October 2000. The plant was identified by Dr Sas Biswas, Scientist-SF, Systematic Botany Branch, Forest Research Institute, Dehra Dun by comparing our specimen with the authentic one preserved in the herbarium of the Institute. A voucher specimen (Rameshwar Dayal 156633) was also deposited in the herbarium of Systematic Botany Branch, Forest Research Institute, Dehra Dun.

The bark was removed and the bast was peeled out from the log and cut into (approx. 0.5 cm^2) small pieces. The bast (1250 g) was hydrodistilled in a Clevenger apparatus for 6 h. The distillate was extracted with diethyl ether and dried over anhydrous Na₂SO₄. Removal of the ether in a gently heated water bath (30°C) yielded an oil (0.015%) on a moisture-free basis.

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