

Wood dimensional stability and extractives as reasons for termite and fungal resistance of the lesser known *Albizia malacophylla* Kenyan wood species

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Abstract Studies were carried out on termite and fungal resistance of the lesser known *Albizia malacophylla* Kenyan wood species. In addition wood dimensional stability, amount and chemical nature of heartwood extractives was also evaluated. Wood resistance against white rot tropical fungi was based on a laboratory soil bed test and subterranean termite resistance in the field according to American Wood Protection Association: E7-93 (1993) standard. Wood extractives were subjected to infra-red analysis using standard laboratory procedures. *Albizia malacophylla* heartwood is dimensionally stable (5.5 %) with a less dimensionally stable sapwood (9.6 %). Heartwood extractive content is high (9.7 %) in comparison to sapwood (4.6 %). *Albizia malacophylla* heartwood is resistant to fungi (8.1 %) mass loss and very resistant to termites (4.8 %) mass loss reported after 6 months exposure. Removal of extractives significantly lowered heartwood dimensional stability, termite and fungus resistance. Heartwood extractives were able to inhibit the growth of fungi under laboratory sterile conditions. Infra-red analysis of crude heartwood extractives indicated presence of aldehydes, ketones carbonyl compounds, esters, aromatic, carboxylic acids and aliphatic carbonyl compounds. Put together, the nature, amount of heartwood extractives and wood dimensional stability are at the origin of the found termite and fungus resistance of *A. malacophylla* wood.

Keywords *Albizia malacophylla* · Wood resistance · Fungi · Termites · Kenya

Introduction

Albizia malacophylla is a lesser known tropical hardwood species, characterized by a rough, pale brown grey bark with large irregular flat scales and attains a height of about 9 m at maturity (Eggeling and Dale 1951). The tree has lost large part of its expanse African wooded grassland due to overexploitation hence is currently endangered and facing threats of extinction (World Conservation Monitoring Centre 1998). In Kenya the remaining *A. malacophylla* is restricted to Teso, Bungoma County. Preliminary survey indicated that its wood is preferred for fencing posts, general construction, charcoal production and firewood over other local species, yet its wood properties are not well known. Wood properties of scarce and endangered species lead to its conservation through cultivation in plantations and possibly expand its utilization as a commercial wood (Silva et al. 2007).

Indeed, other tropical hardwoods such as Teak (*Tectona grandis*) possess good natural durability against decay and insect attack, are dimensionally stable and very resistant to surface checking and end-grain splitting (Williams et al. 2001). *Albizia adianthifolia*, *A. feruginea* and *A. zygia* have moderate to very durable heartwood, course texture, low shrinkage and little movements hence suitable for joinery and carpentry purposes (Kukakcha 1969). It is therefore important to carry out wood durability tests for better and professional utilization of this forest produce.

Evaluating reasons of natural durability of wood leads to more competitiveness and confidence as a building material (Acker et al. 2003; Calonego et al. 2010). Aspiration of bordered pits results in reduced permeability while high levels of gums limit the penetration of water (Schubert et al. 2011; Gerardin et al. 2004). Extractives contribute to wood color, fragrance, durability, pulping, drying adhesion, hygroscopicity and

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acoustic properties (Umezawa 2001; Baeza and Freer 2001). Flavanoids in wood contributes to diversity in wood colorations and have significant effect on its durability (Sirmah et al. 2011). Wood density is closely associated with its mechanical strength, shrinkage and heating value (Githiomi and Kariuki 2010). Cracking, twisting, shrinkage and swelling are attributed to undesirable dimensional changes (Sun et al. 2010).

This study reports termite and fungal resistance of *A. malacophylla* wood and some reasons that contribute to the found resistance.

Materials and methods

Wood procurement

From a household plantation in Teso Western Kenya, mature *A. malacophylla* were randomly selected, felled and quarter sawn into boards 1 m long \times 0.1 m wide \times 0.1 m thick. Defect free boards were sorted and air dried until they attained moisture content of <20 % before sawing further into appropriate sizes for termite, fungal and dimensional stability tests. Mature *Pinus patula* a perishable Kenyan species was similarly acquired for comparison purposes from Kaptagat forest plantation in Elgeyo Marakwet County, Kenya.

Wood resistance against fungi in a soil bed tests

Wood specimen preparation

This study was based on a modified AWWA: E23-07 (2008) (American Wood Protection Association) test standard. One hundred and sixty (160) heartwood specimens of *A. malacophylla*, 50 mm \times 10 mm \times 5 mm, (l \times r \times t) were prepared. To understand effects of extractives on wood resistance, thirty-two specimens were Soxhlet extracted separately in 150 ml of hexane, dichloromethane, acetone and water for 48 h at a rate of 6–10 cycles per hour. Thirty-two specimens of *P. patula* sapwood not solvent extracted were cut to the same dimensions as above. All the wood specimens were then conditioned at 60 °C in an oven to a constant mass (w_0) before they were subjected to fungi in a soil bed test. Solvent residue was evaporated to dryness and extractive yield expressed as percentage of initial weight of wood specimens (w_0).

Soil bed preparation and exposure of wood specimens

Plastic containers 250 mm \times 150 mm \times 170 mm (l, w, h) were filled with unsterile soil layers (bottom to top) as follows: 20 mm of gravel, 20 mm of sand and 130 mm of forest soil. The forest soil was collected from Kaptagat

Forest. Un-extracted, solvent extracted heartwood specimens of *A. malacophylla* and *P. patula* were exposed to the unsterile soil bed under room temperature (22–28 °C) and controlled soil moisture of about 70 % that is reported to optimize fungal wood decay. Soil beds were weighed weekly and sufficient water added to bring the weight of the container back to the original weight that gave the required moisture content. The specimens were labeled for identification purposes then buried randomly 20 mm apart so that \sim 1/3 length of each specimen protruded above the soil bed level.

Fungal wood decay resistance assessment

Fungal wood decay was assessed by evaluating the mass losses once every 4 weeks for 32 weeks. During each inspection a replicate set of 4 specimens was removed from the soil bed cleaned from any mycelium and soil particles which adhered to the surface. The specimens were then oven-dried at 102 ± 3 °C for 48 h and the mass loss evaluated as a percentage of the original mass before exposure using the formula below:

$$\text{Mass loss } \% = \frac{w_0 - w_1}{w_0} \times 100$$

where w_1 and w_0 is the weight of test specimen after and before exposure to fungal decay respectively.

Wood resistance against termites in a field test

Wood specimen preparation

This study was based on a modified American Wood Protection Association, AWWA: E7-93 (1993) method of determining resistance to subterranean termites. One hundred and twenty (120) heartwood specimens of *A. malacophylla*, 100 mm \times 10 mm \times 5 mm (l, r, t) were prepared. To understand effects of extractives on termites wood resistance, twenty-four specimens were Soxhlet extracted separately in 150 ml of hexane, dichloromethane, acetone and water for 48 h at a rate of 6–10 cycles per hour. In addition twenty-four non solvent extracted heartwood specimens and twenty-four *P. patula* sapwood were cut to the same dimensions as above. All the wood specimens were conditioned at 60 °C in an oven to a constant mass (w_f) before they were exposed to termites.

Termite nest preparation, exposure and attack evaluation

Termite nests were arbitrarily selected at Cheptebo, Kerio Valley in Elgeyo Marakwet County, Kenya and the test site cleared of plant life and any other cellulosic material. The test specimens were exposed to termites in a randomized

design at a distance of 30 cm from the termite nest with two-thirds of their lengths buried in the soil and a spacing of 20 cm apart. Every month, four replicate specimens were gently removed from the soil and soil particles on the surface of the specimens brushed off. The specimens were then dried until they attained constant weights (w_i) after which the extent of attack was evaluated by mass loss using the following formula:

$\%$ mass loss = $\frac{w_f}{w_i} \times 100\%$, where, w_f and w_i is the weight of test specimen before and after termite attack respectively.

Effects of heartwood extractives on fungal growth

Fungal mycelia initially isolated from attacked wood samples in the soil bed test described previously, were grown in 9-cm petri dishes filled with 20 ml of potato dextrose agar (PDA) medium containing none (control), 50 and 250 ppm of *A. malacophylla* heartwood extractives. Petri dishes were inoculated at the center by placing a 10 mm diameter fungi cut from the edge of an initially growing colony of fungus on PDA medium. The cultures were incubated in a sterile chamber at 22–24 °C for a period of 9 days. Fungal growth was evaluated daily by measuring two vertical diameters of the colony.

Each experiment was replicated three times. Growth was expressed as a percentage of the available space for growth according to Gerardin et al. 2004 as follows.

$$\text{Growth inhibition } \% = 100 \left(1 - \frac{d_1}{d_0} \right)$$

where d_0 and d_1 is the diameter of the untreated culture and culture with the extracts respectively.

Effects of extractives on wood dimensional stability

Twenty specimens from *A. malacophylla* heartwood and sapwood measuring 20 mm × 20 mm × 20 mm (dimensions measured to the nearest 0.01 mm) were cut. Sixteen heartwood specimens were Soxhlet extracted, four in each of the solvents (hexane, dichloromethane, acetone and water as previously described) while four were not extracted. Similar procedure was applied for sapwood specimens. The purpose of extraction was to test whether extractives influences wood dimensional stability. All wood specimens solvent extracted or not were oven-dried at 60 °C to constant weights and initial volumes (V_0) evaluated. All specimens were then exposed to hydrated copper (II) sulphate solution. The specimens were weighed after every 48 h until constant mass was reached, indicating that they have attained maximum moisture content. The dimensions of the specimens were measured again and wet

volume (V_1) determined. Swelling coefficient was calculated using the formula below:

$$\text{Swelling}(\%) = \frac{V_1 - V_0}{V_0} \times 100$$

The experiment was replicated three times.

FTIR analysis of heartwood extractives

1 µg of extract was mixed with a spatula end full of KBr, ground evenly in a mortar and pressed into disks. This was then examined in an FTIR between wavelength number ranges of 500–4,000 cm^{-1} .

Data analysis

Data was analyzed using Statistica version 7 for windows and analysis of variance (ANOVA). Pair wise comparison for dimensional stability was used to test for equality of means using two sample independent t tests. The analysis assumes that all the means comes from the same population.

Results and discussions

Wood dimensional stability

Figure 1 shown below reports the effects of solvent extraction of *A. malacophylla* wood on extractive yield and its dimensional stability.

A. malacophylla heartwood contains high amount of extractives (9.7 %) and is dimensionally stable (5.52 %) than sapwood, indicating strong hydrophobic nature of this wood species. Literature studies have attributed the dimensional stability of heartwood and high extractive content to wood natural durability (Kose and Taylor 2012; Neyra et al. 2004)). Heartwood and sapwood dimensional

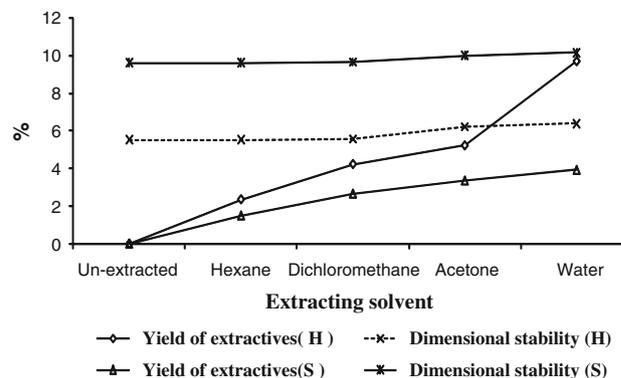


Fig. 1 Relationships of *A. malacophylla* heartwood (H) and sapwood (S) extractives and its dimensional stability

stability is significantly lowered ($p < 0.05$) through removal of extractives by different solvent extraction. The more polar solvents such as water and acetone extraction have more influence on both dimensional stability and extractive removal than the less polar hexane and dichloromethane ($p < 0.05$). This observation is in agreement with literature studies showing that polar solvents remove extractives located in the cell wall (Royer et al. 2010). Such extractives are known to bind to the polymeric cell wall by means of multiple hydrogen bonds making the wood dimensionally stable (Royer et al. 2010) and may be one of the factors contributing to its natural resistance.

Natural wood durability tests against fungi

Solvent extracted (water, hexane, dichloromethane, acetone, and unextracted *A. malacophylla* heartwood specimens were exposed alongside *P. patula* (a perishable species) to fungi in unsterile soil bed for a period of 32 weeks. The wood mass losses due to fungal degradation in the soil bed are reported in Fig. 2 below:

The first 4 weeks of exposure showed little variability in decay with percentage mass loss on all test specimens of about 2.3 %. This unclear decay pattern during initial period of wood exposure in soil bed test has been reported for other species (Acker et al. 2003). In the next 8 weeks, mass loss on solvent extracted specimens increased tremendously in comparison with the un-extracted specimens which still retained the least mass loss average of 2.6 %. Thereafter the mass loss increased immensely on the *P. patula* and the solvent extracted *A. malacophylla* wood specimens. In all the test specimens, mass losses due to fungal decay increased with exposure period with a significant difference amongst the treatments ($p < 0.05$). This is in agreement with observation that mass loss in softwoods is generally low in soil bed test (Acker et al. 2003; Machek et al. 2001). In our current experiment removal of extractives lead to significantly higher mass losses ($p < 0.05$), suggesting reduced decay resistance. It is therefore clear that *A. malacophylla* extractives contribute to wood natural resistance against fungi. Indeed, in their

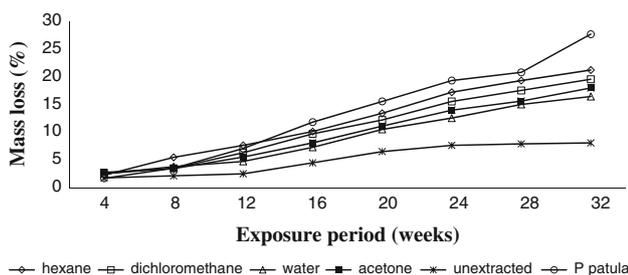


Fig. 2 Percentage mass losses of wood specimens in the fungal soil-bed

study on the decay resistance of Southern Asian timbers in sand block tests, Takahashi and Kishima (1973), observed that the non extracted specimens were more resistant to decay compared to solvent extracted specimens. Generally, the mass loss increased with decreasing solvent polarity implying that extractives soluble in low polar solvents such as hexane are mostly responsible for fungal decay resistance of *A. malacophylla*.

Fungal growth inhibition by *A. malacophylla* extractives

Unidentified tropical white rot fungus from the soil bed test experiment was cultured in PDA medium and inoculated with 0, 50 and 250 ppm of extractives. Fungal mycelium growth was measured daily from the centre of the petri dish and growth inhibition evaluated at the end of the experiment.

Figure 3 below shows percentage growth inhibition of hexane, acetone and water extracts of *A. malacophylla* heartwood at different concentration level.

Hexane extractives gave the highest inhibition activity against growth of fungi at 50 and 250 ppm concentrations in comparison with acetic and water extracts at the same level of concentration. Literature studies have reported similar growth inhibition properties of heartwood extractives of other tropical wood species (Neya et al. 2004; Mburu et al. 2007; Sirmah et al. 2008). The development of fungi on PDA treated with different concentration of hexane extracts as a function of time was measured daily. The results are presented in Fig. 4a and b.

Fungi started growing on the PDA medium treated to 50 and 250 ppm hexane extract after 2 and 4 days respectively. Similar trend of fungal activity was observed in acetic and water extracts (results not shown), suggesting fungistatic properties of *A. malacophylla* heartwood

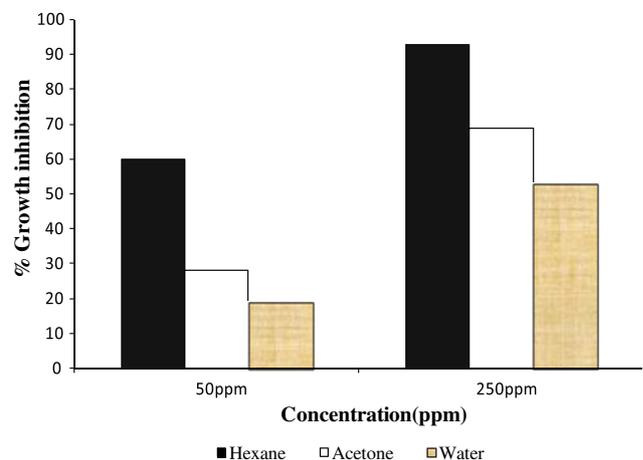


Fig. 3 Percentage growth inhibition of *A. malacophylla* heartwood extractives

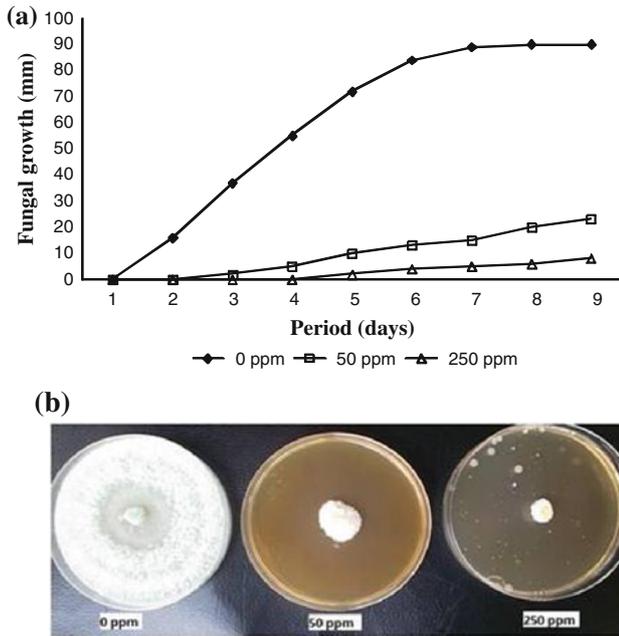


Fig. 4 Fungal growth on potato dextrose agar treated to different concentrations of hexane extractives. **a** Fungal growth as function of time in increasing hexane extract concentration. **b** Appearance of fungi after 9 days in increasing hexane extract concentration

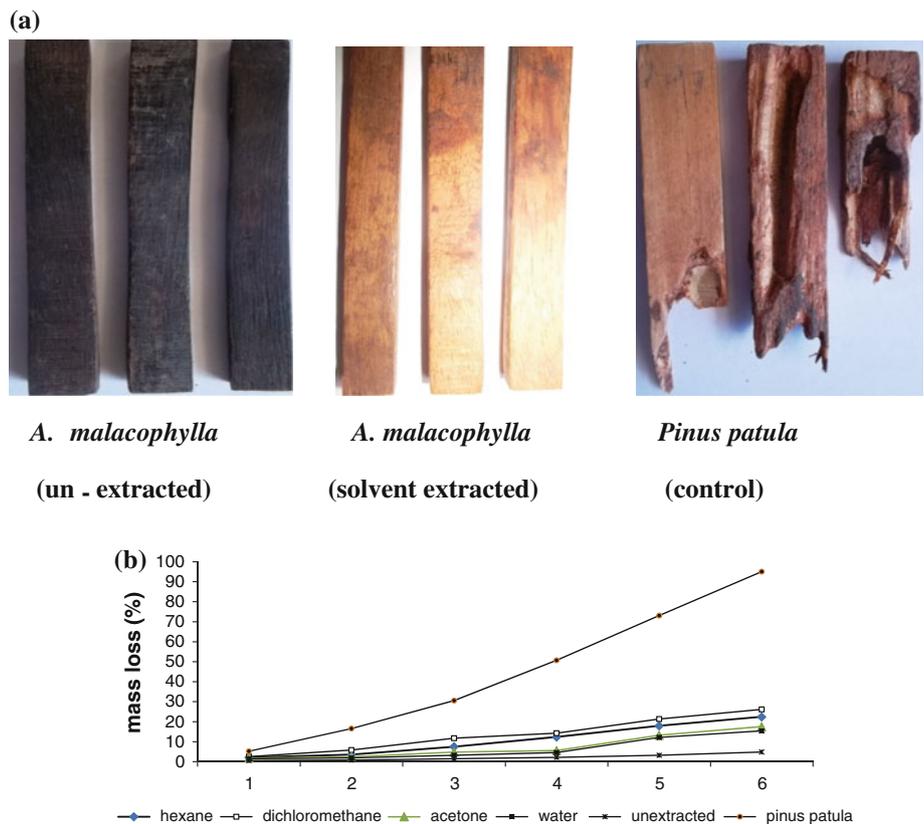
extractives even at low concentrations. This observation is similar for most tropical hardwood species reported in literature (Neya et al. 2004; Mburu et al. 2007; Sirmah et al. 2008). Indeed it can be inferred that heartwood extractives of *A. malacophylla* restricts the growth of fungus thus contributing to the reported wood natural durability. Similar observations have been made on extracts of other wood species (Mohareb et al. 2010; Sirmah et al. 2008; Mburu et al. 2007; Martinez-iningo et al. 1999).

Natural wood durability tests against termites

Figure 5a below shows the physical appearance of wood specimens that were subjected to termite attack in the field for a period of 6 months while Fig. b presents the % mass loss of wood specimens over the same period of time.

The highest mean mass loss was observed in heartwood specimens extracted with dichloromethane (26.1 %) followed by those that were extracted by hexane solvent (22.4 %). The un-extracted specimens showed the least mass loss of (4.8 %) while the *P. patula* controls were severely attacked and had mass loss of 95 % after 6 months of exposure. Heartwood specimens extracted

Fig. 5 **a** Extent of termite attack and **b** mass loss of wood specimens exposed to termites



with highly polar water solvent were least attacked by termites (15.4 %). It can be predicted that water would have removed polar extracts such as sugars and other soluble carbohydrates thus making the specimens less desirable to termite attack. Studies by Lukmandaru (2011) on the variability in the natural resistance of teakwood and its relationship with wood extractive content found that the lower the extractive content, the less severe the mass loss to termites. This was attributed to the removal of polar components that would have induced the termite activity in attacking and degrading the teakwood. In general, heartwood extractives of *A. malacophylla* especially those that are soluble in the less polar dichloromethane solvent contribute most to the natural resistance to termite attack.

FTIR analysis of heartwood extractives

FTIR Studies were carried out to understand the main functional groups present in the *A. malacophylla* heartwood extracts. Figure 6 below is an FTIR spectrum of dichloromethane heartwood extract.

The spectrum indicates an OH stretch at $3,055\text{ cm}^{-1}$ and a strong aliphatic stretch at $2,935\text{ cm}^{-1}$. The C=C group absorption is indicated by vibrations at $3,687\text{ cm}^{-1}$, typical

of either an alkene or an aromatic compound, confirmed by strong vibrations at $1,431$ and $1,608\text{ cm}^{-1}$ and C–H out of plane bending at 898 cm^{-1} . The strong carbonyl (C=O) absorption is evident at $1,724\text{ cm}^{-1}$, corroborated by C–H absorption at $2,862\text{ cm}^{-1}$. This indicates presence of aldehydes, ketones, carboxylic acids, esters or amines. The strong phenyl ring substitution band is evident at 736 cm^{-1} . Figure 7 below is an FTIR spectrum of acetonic heartwood extract.

The spectrum indicated a C=C group absorption at $3,525.6$ and $3,625.6\text{ cm}^{-1}$. The absorption at $2,927.7\text{ cm}^{-1}$ is attributed to the presence of aliphatic hydrogen while carbonyl group absorption at $1,720.4\text{ cm}^{-1}$ is confirmed to be an aldehyde at $2,788.9\text{ cm}^{-1}$. Presence of alcohol was indicated by spectrum absorption at $3,525.6$ and $3,602.8\text{ cm}^{-1}$.

Conclusions

A. malacophylla heartwood is dimensionally stable (5.52 %) with a less dimensionally stable sapwood (9.6 %), indicating its strong hydrophobic nature. Heartwood contains high amount of extractives (9.7 %) in comparison to

Fig. 6 FTIR spectrum of dichloromethane extractives

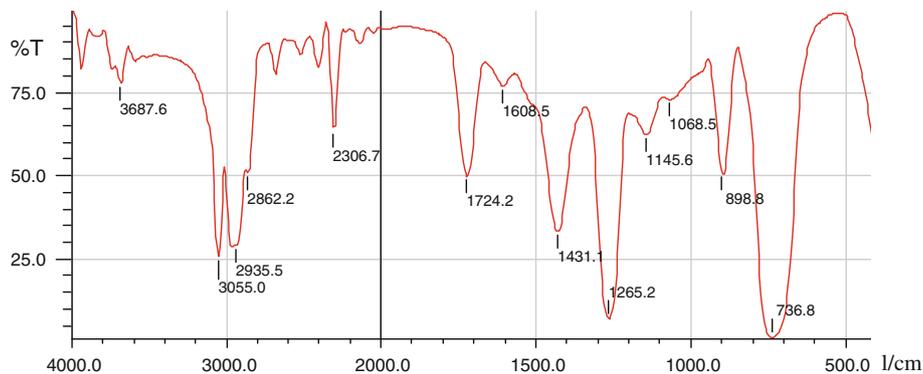
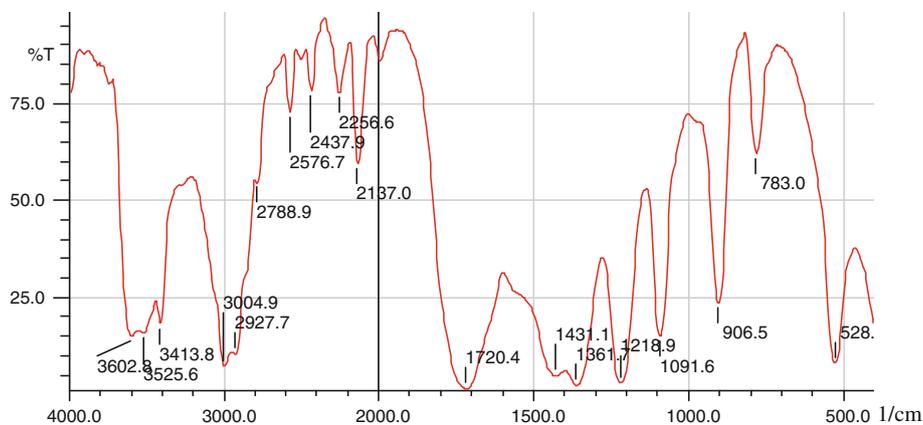


Fig. 7 FTIR Spectrum of acetone extractives



sapwood (4.6 %). Most of these extractives are soluble in the polar water solvent (9.7 %) while small amount are soluble in a non polar hexane solvent (2.36 %). Removal of extractives by solvent extraction makes this wood less dimensionally stable, less hydrophobic and prone to degradation by fungi and termites. *A. malacophylla* wood is durable against decay fungi in soil bed (8.1 %) mass loss and very resistant to termites in field (4.8 %) mass loss but in both cases resistance is lowered by solvent removal of extractives. Heartwood extractives inhibit the growth of fungi to some extent at different levels of concentration. Hexane extracts are effective as fungal growth inhibitors even at low concentration levels in comparison with water and acetone extracts. Infrared analysis indicated presence of aldehydes, ketones carbonyl compounds, esters, aromatic, carboxylic acids and aliphatic carbonyl compounds in the different extracts. Put together, the nature, amount of heartwood extractives, wood dimensional stability and its strong hydrophobicity are at the origin of the found termite and fungus resistance of *A. malacophylla* wood.

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