

The Natural Termite Resistance of Teak Wood Grown in Community Forest

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Abstract

The objective of this study was to evaluate the antitermitic activities of young teak wood through wood extracts and wood blocks samples method. Correlation between extractive content and its antitermitic properties was also studied. The disc samples from the bottom part of the five 8 years old and four 22 years old trees were sawn and samples from outer sapwood, inner sapwood, outer heartwood, and inner heartwood were prepared. Extractive contents were determined through secluded cold extraction of the wood meal using *n*-hexane, ethyl acetate (EtOAc) and methanol (MeOH). Force-feeding method using *Reticulitermes speratus* Kolbe termites was used for termite resistance test. A significant interaction between tree age and radial direction factors existed with regards to the *n*-hexane and EtOAc extractive content. The tree age factor did not affect the mass loss levels in the extract samples. The most susceptible to termites was the sapwood of the 8 years old trees. The *n*-hexane and ethyl acetate extracts of the inner and outer heartwood significantly influenced the mortality rate, but in contrary to that of tree age. The EtOAc extracts of the outer heartwood was the most active antitermite. The extractive content of heartwood did not correlate to antitermite properties.

Key words: antitermitic activities, community forest, extractive, *Reticulitermes speratus*, *Tectona grandis*

Introduction

Teak (*Tectona grandis* L.f) has been recognized for its versatile uses due to its high durability. The present increasing demand of teak wood has encouraged the use of less durable teak wood obtained from younger tree planted in community forests. However, the quality of wood from community forest has been questioned. The woods contain a large proportion of sapwood and immature wood, whose properties may differ from those of older trees. A more rational use of naturally durable timber can be realized when its properties are well understood. Therefore, the present research was aimed to evaluate the

resistance of teak wood from younger tree of community forests against termite and decay fungi.

Bhat and Florence (2003) previously reported that a 5 year old juvenile wood was less decay resistant than a 13 year old and mature teak wood from forest plantations. The lower durability of young plantation teak and inner heartwood of older trees were also observed by some researchers (Da Costa *et al.* 1958, 1961, Bhat *et al.* 2005, Kokutse *et al.* 2006). Even though natural durability of wood has been related to extractives, however, the influence of extractive content on natural durability of teak wood has not been done to a greater extent. In addition,

various methods have been used to assess the natural durability of wood either based on natural solid wood or based on isolated extractives. It has been well known that lower extractive content in several other woods correlated with reduced termite and fungal resistance (Hillis 1987, Hashimoto *et al.* 1997). Proceeding our previous works (Lukmandaru & Takahashi 2008, Lukmandaru 2011), the purpose of the present study was to evaluate the natural termite resistance of 8 and 22 years old teak woods grown in community forests based on no-choice feeding method and to relate it with the extractive contents. Comparison between wood block and wood extractives methods in assessing wood resistance against termite were also carried out.

Materials and Methods

Sample preparation

Trees of the 8 years old (5 trees, diameter at breast height (dbh) of 8-13 cm, sapwood proportion of 66-84%) and 22 years old (4 trees, dbh of 23-30 cm, sapwood proportion of 36-44%) were felled from community plantation forests

of Jogja Province. A 5 cm thick disc was taken from the trees at approximately breast height. The 8 years old wood discs were divided into three parts (Figure 1), i.e. the outer sapwood (OS), inner sapwood (IS), outer heartwood (OH), while those from the 22 years old wood were divided into four parts, i.e. the outer sapwood (OS), inner sapwood (IS), outer heartwood (OH), and inner heartwood (IH). From each part, blocks were tangentially (perpendicular to the radii) sawn across the diameter and then converted into wood meal (40–60 mesh size) to determine the content and chemical composition of the extractives. For the termite resistance test, blocks ca. 5.0 (L) × 0.8 (T) × 0.8 (R) cm were obtained in a similar pattern to those of extractives analysis sampling. The blocks were then dried at 100 °C for 3 h, after which they were cooled and weighed. The meals from two opposite radii were then combined to form a single sample in order to minimize any variation between radii. For comparative purposes, 5 of 51 years old trees were felled from Perhutani Plantation, Randublatung, Central Java as well as susceptible pine sapwood blocks.

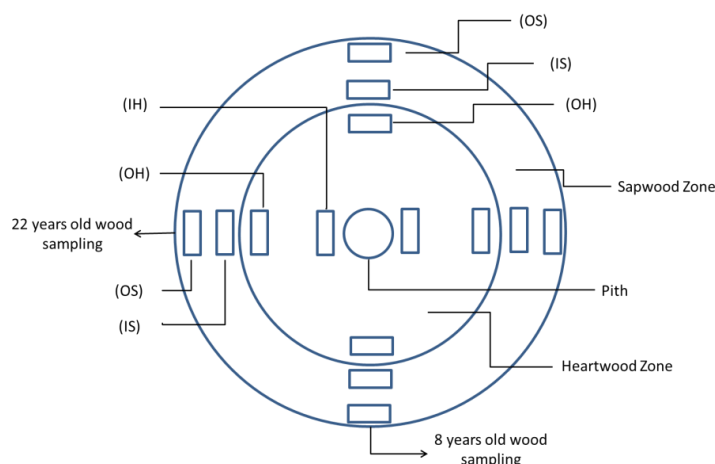


Figure 1 Sampling position on a cross-section of teak trunk (OS = Outer sapwood; IS = Inner sapwood; OH = Outer heartwood; IH = Inner heartwood).

Determination of extractive content

Extractives were obtained through cold extractions of 2 g wood meal for 24 hours in *n*-hexane, ethyl acetate (EtOAc), and methanol (MeOH). After evaporating the solvent, the extractives were removed, dried and weighed to determine the percentage of extractive content based on moisture-free wood meal. No extraction was conducted to the controls (the 51 years old trees).

Termite resistance testing

Wood extract

No choice anti-feedant bioassay test was carried out in this research. A petri dish (diameter of 9 cm and height of 2 cm) containing 20 g moistened and sterilized sea sand was used as a container test. Paper discs of 8 mm diameter (Whatmann International) were impregnated with chloroform solution containing each extract of the test fractions. No extracts from the control trees were tested. The treatment retention was 5% (w/w) per disc and 5 duplicates were applied for each sample. After drying at 60 °C for 2 h, followed by drying in a vacuum desiccator for 24 h, they were put on a petri dish. The control discs were impregnated with chloroform only and dried with the same manner. Fifty worker *Reticulitermes speratus* Kolbe termites were introduced into the petri dish. The petri dishes were placed in a dark chamber at 27 °C and 80% relative humidity. After 10 days the discs were taken out, dried in the same manner and the weight loss was determined. Dead termites were counted at the end of observation.

Wood block

For each test, an air-dried wood block (moisture content of 10–12%) was

inserted into a plastic cup (5.0×6.0) cm², and placed on 20 g of sterilized sand. The sand was moistened with distilled water regularly to retain a constant relative humidity. Fifty worker termites were added to each cup. Included in the tests for comparative purposes were controls (51-year-old trees). Three replicates were measured for each sample. The cups were placed in the environmental chamber for 14 days. To measure the termiticidal activity, surviving termites were counted at the end of observation. The blocks were then dried at 100 °C for 3 h, after which they were cooled and weighed. The mass loss was determined to quantify the extent of the termite attack on the wood.

Data analysis

The effects of tree age and radial position on extractive content, survival rate and mass loss were calculated by analysis of variance (ANOVA) GLM procedures followed by Duncan's multiple range test ($p = 0.05$). The relationships between the independent variables were studied with a Pearson's correlation analysis. The termite survival rates (percentages) were transformed by the arcsine function for analysis but were presented as untransformed values to facilitate interpretation. All statistical calculations were conducted using SPSS-Win 10.0

Results and Discussion

Factorial analysis of variance results for the various properties measurements are summarized in Table 1. There are significant interactions between tree age and radial direction factors in regards to *n*-hexane and EtOAc extractive content. Both factors did not significantly affect termite mortality in *n*-hexane and MeOH extracts.

Table 1 Factorial analysis of variance results for extractive content, mass loss, and termite mortality of three different extracts (*n*-hexane, ethyl acetate (EtOAc), and methanol (MeOH))

Source of variation	df	ss	MS	VS	Fpr
(a) <i>n</i> -hexane extractive content					
Tree age	1	2.312	2.312	33.317	<0.01**
Radial direction	3	5.061	1.687	24.309	<0.01**
Tree age x radial direction	2	0.515	0.257	3.710	0.038*
(b) Mass loss in <i>n</i> -hexane extract					
Tree age	1	21.757	21.757	2.498	0.126 ^{ns}
Radial direction	3	138.915	46.305	5.316	0.005**
Tree age x radial direction	2	8.010	4.005	0.460	0.636 ^{ns}
(c) Termite mortality in <i>n</i> -hexane extract					
Tree age	1	0.0314	0.0314	1.047	0.316 ^{ns}
Radial direction	3	0.230	0.076	2.551	0.078 ^{ns}
Tree age x radial direction	2	0.0079	0.003	0.133	0.876 ^{ns}
(d) EtOAc extractive content					
Tree age	1	3.795	3.795	24.557	<0.01**
Radial direction	3	30.704	10.235	66.228	<0.01**
Tree age x radial direction	2	1.248	0.624	4.037	0.03*
(e) Mass loss in EtOAc extract					
Tree age	1	5.391	5.391	0.643	0.43 ^{ns}
Radial direction	3	143.451	47.817	5.704	0.004**
Tree age x radial direction	2	2.853	1.426	0.170	0.844 ^{ns}
(f) Termite mortality in EtOAc extract					
Tree age	1	2925.714	2925.714	6.359	0.019*
Radial direction	3	8746.445	2915.482	6.337	0.003**
Tree age x radial direction	2	80.647	40.323	0.088	0.916 ^{ns}
(g) MeOH extractive content					
Tree age	1	21.757	21.757	2.498	0.126 ^{ns}
Radial direction	3	138.915	46.305	5.316	0.005**
Tree age x radial direction	2	8.010	4.005	0.460	0.636 ^{ns}
(h) Mass loss in MeOH extract					
Tree age	1	1.764	1.764	0.167	0.686 ^{ns}
Radial direction	3	331.869	110.623	10.478	<0.01**
Tree age x radial direction	2	23.358	11.679	1.106	0.345 ^{ns}
(i) Termite mortality in MeOH extract					
Tree age	1	136.896	136.896	0.183	0.673 ^{ns}
Radial direction	3	3412.214	1137.405	1.522	0.237 ^{ns}
Tree age x radial direction	2	169.936	84.968	0.114	0.893 ^{ns}

Note: ns = not significant, ** Significant at 1 % level, * significant at 5 % level

Tree age factor significantly affected termite mortality in EtOAc extract while the radial direction affected the rest parameters.

Extractive content

Generally, the extractive content increased with the polarity in every part of the wood (Table 2). MeOH extractive contents gave the highest amounts although the values obtained here were

lower than the published data (Da Costa *et al.* 1958, Lukmandaru & Takahashi 2008, Lukmandaru 2011, Narayanamurthi *et al.* 1962) of young teak wood. Theoretically, MeOH extracts both non-polar and polar extractives. The lower values were due to cold extraction was used in this experiment instead of using reflux or soxhlet extraction as the early reports were. In each solvent, the OH showed significantly higher values

than those of sapwood. There were significant differences in the extractive content levels between IS and OS as well as between IH and OH depend on the solvents. The average of *n*-hexane extractive contents in the IS was almost twice as those in the OS. The increasing of *n*-hexane and EtOAc extractives content from the inner to outer heartwood was also found. Those trends are interpreted as the increasing of non-polar substances along with the increasing the age of the wood in radial direction.

Mass loss

In the form of wood block, the tree age factor significantly affected the mass loss levels, in that 51 year old tree gave the lowest values in each part. The level of activity of the OS of the 51 years old trees was even comparable to that of the OH of 8-year-old trees. The striking differences were found in the sapwood part in that the 8 years old tree showed the most susceptible ones. It is noticed, except in the OS part, the mass loss levels trees were not statistically different between 22 and 51 years old trees. The termite susceptibility in the sapwood must be taken into consideration, since the percentage of

sapwood was relatively high in trees younger than 51 years old.

In the form of extracts, the EtOAc extracts tend to give lower values of mass loss than other extracts. In line with our previous study (Lukmandaru 2011), by successive extraction, EtOAc the greater the EtOAc soluble extractives (EEC) then the higher the termite resistant (lower mass loss), and conversely the greater the MeOH-soluble extractive (MEC) then the lower the termite resistant (greater mass loss). It seems that not all of these components are equally important in determining natural durability as heartwood extractives within a piece of wood can range from low molecular weight volatile compounds to large polymers.

The ANOVA showed that tree age did not affect the mass loss of wood on contrary to radial direction. As expected, the outer heartwood part showed significant higher activities although it did not significantly differ between outer and inner heartwood in the MeOH extracts. No significant differences were also found between outer and inner sapwood as well as between the outer heartwood of 8 and 22 years old trees in all extracts.

Table 2 Extractive content (% oven dried mass m^{-1}) of cold extraction *n*-hexane, EtOAc, and MeOH in teakwood by tree age and radial position

Tree age	Radial position	Extracts		
		<i>n</i> -hexane	EtOAc	MeOH
8 years old	Outer sapwood	0.32 (0.16) a	0.71 (0.21) f	2.13 (0.67) j
	Inner sapwood	0.63 (0.28) b	0.93 (0.28) f	2.31 (1.10) j
	Outer heartwood	1.05 (0.34) c	2.61 (0.43) h	4.00 (0.56) k
22 years old	Outer sapwood	0.53 (0.06) b	0.96 (0.21) f	1.73 (0.62) j
	Inner sapwood	1.31 (0.27) d	1.60 (0.72) g	2.14 (0.73) j
	Outer heartwood	1.89 (0.28) e	3.90 (0.41) i	3.69 (0.55) k
	Inner heartwood	1.34 (0.32) d	2.43 (0.35) h	3.14 (0.36) k

Note: Mean of 5 trees (8 years old) and 4 trees (22 years old), with the standard deviation in parentheses

Compared to the patterns in wood blocks, the similarity was found in 8 years old trees in that an increase in natural termite resistance from sapwood to heartwood in all extracts was evidenced. The pronounced differences in the patterns between the wood blocks and extracts were detected between the OH and IH in the 22 years old trees.

Mortality rate

In the form of wood blocks, sapwood of 8 years old trees showed the lowest activities while no systematic differences in the heartwood parts were found among the three ages (Table 4). It was also noted that the mortality rate in sapwood of 22 years old trees were not statistically different than those of the heartwoods. The patterns in the mortality rate of the 8 years old trees were in line with those of mass loss levels in the same tree age. However, the insignificance of mortality rate among the parts in the 22 and 51 years old trees differed from those of mass loss levels.

In the extracts, most OS parts gave lowest values of mortality rate. The IS parts showed significantly higher activities than OS in the EtOAc of 8 years old trees. In the heartwood, significant differences between inner and outer heartwood were measured in the *n*-hexane and EtOAc extracts. The OH in EtOAc extracts showed the most active against termites. No significant effects were found after treating with MeOH extracts although it still showed activities compared to untreated controls.

This fact indicated that the most active substances were that soluble in EtOAc. On the other hand the MeOH soluble weakened the termite resistances.

The patterns were similar between the wood blocks and the extracts in the 8 years old trees samples. On the contrary, those trends were not found in the 22 years old trees as no significant differences were measured in every parts of the wood block samples.

Table 3 Mass loss (mg) of teakwood against *R. speratus* by tree age and radial positions

Tree age	Radial position	Extracts ^a			Wood block ^b
		<i>n</i> -hexane	EtOAc	MeOH	
8 years old	Outer sapwood	6.55 (2.68) b	5.95 (4.08) c	8.84 (2.88) e	27.68 (8.05) j
	Inner sapwood	5.90 (3.54) b	6.67 (1.72) c	8.82 (3.46) e	31.81 (14.78) j
	Outer heartwood	2.74 (1.97) a	1.37 (0.83) d	2.45 (1.08) f	7.45 (6.39) h
22 years old	Outer sapwood	8.97 (2.07) b	5.48 (3.09) c	11.67 (2.80) e	14.52 (6.78) i
	Inner sapwood	8.45 (2.81) b	4.92 (1.01) c	7.22 (4.43) e	7.84 (6.23) h
	Outer heartwood	2.99 (1.63) a	1.00 (0.83) d	2.74 (1.01) f	4.07 (3.32) gh
	Inner heartwood	6.08 (4.90) b	5.58 (3.30) c	3.80 (2.93) f	5.69 (4.36) gh
Control (untreated)		20.62			
Control (51 years old)	Outer sapwood				6.30 (4.34) gh
	Outer heartwood				1.39 (1.22) g
	Inner heartwood				4.72 (3.46) gh
Control (pine sapwood)					52.41 (5.69)

Note: a = 10-day observation, b= 14-day observation. Mean of 5 trees (8 years old) and 4 trees (22 years old), with the standard deviation in parentheses. The same letters in the same column indicates a non-significantly different at $p < 5\%$ by Duncan's test

Table 4 Mortality rate (%) of teak wood against *R. speratus* by tree age and radial position

Tree age	Radial position	Extracts ^a			Wood block ^b
		<i>n</i> -hexane	EtOAc	MeOH	
8 years old	Outer sapwood	13.00 (7.58)a	11.00 (9.61)c	23.00 (17.58)g	44.15 (14.68)h
	Inner sapwood	10.00 (5.72)a	6.25 (4.78)c	46.80 (24.47)g	52.50 (19.33)h
	Outer heartwood	27.50 (16.41)b	45.00 (32.78)e	53.00 (11.80)g	62.44 (13.10)i
22 years old	Outer sapwood	15.00 (4.24)a	28.75 (24.95)d	25.00 (18.66)g	75.67 (18.56)i
	Inner sapwood	20.00 (5.77)a	32.50 (11.90)d	32.50 (15.24)g	75.82 (22.60)i
	Outer heartwood	36.25 (8.87)b	65.00 (29.43)f	38.50 (13.30)g	71.19 (21.09)i
	Inner heartwood	18.75 (3.76)a	20.00 (17.79)cd	39.25 (18.67)g	74.19 (13.51)i
Control (untreated)			7.00 (6.70)		
Control (51 years old)	Outer sapwood				76.27 (15.15)i
	Outer heartwood				72.80 (13.81)i
	Inner heartwood				68.67 (14.57)i
Control (pine sapwood)					11.33 (2.30)

Note: a = 10-day observation, b= 14-day observation. Mean of 5 trees (8 years old) and 4 trees (22 years old), with the standard deviation in parentheses. The same letters in the same column indicates a non-significantly different at $p < 5\%$ by Duncan's test

The different patterns were also found between the mass loss and mortality rate in the EtOAc and MeOH extracts. Those facts were interpreted as the complexity in natural resistance as well as in heartwood extractives that some factors could affect the results.

Relationship between termite resistance and extractive contents

The correlation between antitermitic properties and extractive content was described in Table 5. If the data in the sapwood and heartwood were combined, significant moderate correlations were measured in the MeOH and EtOAc extracts that means the variation in antitermitic properties between those

parts could be explained partly by extractive contents. The positive correlation between the mortality rate and extractive content indicates that the higher extractive content, the higher the mortality rate. In contrary, a negative correlation between the mass loss and extractive content occurred.

This result is reasonable since the extractive content in the heartwood was higher than that in the sapwood. If the data were divided, unexpectedly, the correlation was found in the sapwood part between the termite mortality rate and EtOAc extractive content levels. On the other hand, no significant correlations were detected in the heartwood part.

Table 5 Pearson's correlation coefficients between natural termite resistance parameters and extractive component contents

Extracts	Antitermitic properties					
	Mass loss			Mortality rate		
	Total	Sapwood	Heartwood	Total	Sapwood	Heartwood
<i>n</i> -hexane extract	-0.23	0.07	0.17	0.30	0.16	0.04
EtOAc extract	-0.53**	-0.14	-0.38	0.64**	0.58*	0.52
MeOH extract	-0.50**	-0.01	-0.08	0.47*	0.48	0.29

Note: ** Significant at 1 % level, * significant at 5 % level

Quinones and their derivatives have been detected to inhibit termite and fungal attacks (Haupt *et al.* 2003, Rudman & Gay 1961, Sandermann & Simatupang, 1966, Sumthong *et al.* 2006, Thulasidas & Bhat 2007). The *n*-hexane extracts non-polar substances such as fats, oil, resin, and waxes as well as some quinones (Windeisen *et al.* 2003).

The EtOAc, a moderate polar solvent, extracts non-polar substances as well as some phenolics whereas the MeOH, a polar solvent, extracts non-polar substances, phenolics and sugars. The significant correlation was measured in the sapwood indicates that this part contained some toxic components. Although the *n*-hexane extractives showed antitermitic activities, no significant relationship was found. As the mass loss was moderately correlated with *n*-hexane extractive content obtained by soxhlet extraction in previous communication (Lukmandaru & Takahashi 2008), this fact indicated that toxic quinones were not thoroughly extracted by cold extraction in this experiment. By cold extraction, it was thought that EtOAc extracted most toxic quinones bringing about higher mortality rate. Even though MeOH extracted toxic quinones also but some non-quinones such as other phenolics or sugars were also extracted which were conversely responsible for lowering termite resistance (lower mortality rate). The variation in the extractive content also could not explain the variation in the antitermitic properties in the heartwood. This could be assumed that there is synergistic or antagonistic relationship among extractives with regard to antitermitic properties. Furthermore, the weak correlations were due to the toxic component concentrations were independent to the pattern of extractive

content. A subsequent study will describe the quantity of toxic components of each extract to explain this discrepancy.

Conclusions

The heartwood and sapwood of all younger trees tested showed antitermitic activity both in the form of wood blocks and wood extracts. The mortality rate and mass loss in the wood block samples of sapwood and heartwood of 8 years old trees are significantly lower than those of 22 years old trees. The similar patterns were also observed in the EtOAc extract samples. Different results were observed between wood blocks and extracts in force-feeding method to termites, as well as the patterns between mass loss and mortality rate which suggest that the relationship between heartwood extractives and heartwood durability is complex. Variations in extractive contents were moderately correlated with the anti-termite properties in the ethyl acetate and methanol soluble but weakly correlated in the *n*-hexane extracts. However, no significant correlations were detected in the heartwood part alone between the extractive content by cold extraction and antitermitic properties.

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