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ORIGINAL

Ex-situ performance of extracts from naturally durable heartwood species and their potential as wood preservatives

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Abstract

To avoid the use of toxic synthetic chemicals due to their potential environmental impacts, the feasibility of using heartwood extracts of *Tectona grandis* and *Cedrus deodara* as wood preservatives against the subterranean termite, *Reticulitermes flavipes* and two basidiomycete decay fungi, *Trametes versicolor* and *Rhodonia placenta*, was investigated in laboratory experiments. There were no significant differences in feeding for *R. flavipes* fed solvent-extracted and non-extracted *T. grandis* in choice and no-choice tests with 100% mortality. *Reticulitermes flavipes* ignored non-extracted *C. deodara* (mass loss 1.93%) in choice tests and consumed significantly more solvent extracted *C. deodara* (mass loss 33.4%) with 64.9% mortality. Complete termite mortality (100%) was observed after exposure to non-extracted *C. deodara* versus 53% mortality when fed on extracted *C. deodara* in a no-choice test. When extracted and non-extracted blocks of each wood species were exposed to decay fungi, durability of both heartwood species was reduced post extraction. Extracts removed from wood shavings via Soxhlet extraction were used to treat non-durable southern pine and cottonwood. Both extracts imparted termite resistance to the non-durable species. Weight losses of both non-durable species were reduced at the highest extract concentration tested (10 mg ml^{-1}), and were inversely related to extract concentrations and retentions. Significantly higher termite mortality was observed at the maximum extract concentration tested for either extract. Water leaching of non-durable wood species treated with *T. grandis* extract did not reduce termite resistance, and no significant difference between mortality of termites on leached versus non-leached samples was observed. Conversely, the weight loss of wood treated with *C. deodara* extract was significantly greater post leaching. *T. grandis* and *C. deodara* extracts showed no protective effects at tested concentrations against decay fungi when applied to non-durable southern pine or cottonwood.

1 Introduction

The natural resistance of wood to biological attack is a complex phenomenon. Some wood species have developed the ability to resist attack by biotic agents. Resistance to termites or decay fungi is attributed generally to the synthesis of toxic compounds in heartwood as a tree grows (Kityo and Plumptre 1997; Kirker et al. 2013; Kadir et al. 2014; Hassan et al. 2017). Chemical compounds/extracts in heartwood are non-structural components present at varying levels. Toxic compounds are found in abundance in heartwood compared to sapwood, the latter being more susceptible to decay and termite attack because of this deficiency. Thus, the presence of toxic heartwood extracts affects resistance to biological attack (Hinterstoisser et al. 2000). Heartwood extracts have been found to exhibit antifeedant, antioxidant, antiviral, bactericidal and fungicidal properties (Walker 2006; Ragon et al. 2008, Hassan et al. 2018a) and can protect wood by direct toxicity to termites (Walker 2006; Hinterstoisser et al. 2000; Schultz and Nicholas 2002; Hassan et al. 2016a, b, 2017, 2018a, b, 2019). Other factors such as wood density, specific gravity, hardness and lignin content also affect biological activity of heartwood compounds and natural resistance of wood (Schultz and Nicholas 2002; Arango et al. 2006; Kirker et al. 2013; Owoyemi and Olaniran 2014). Previous studies indicate that natural durability of wood against termite attack could be linked to increased specific gravity (Esenther 1977; Rasib et al. 2014). However, other

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studies found that specific gravity and wood density were poorly correlated with fungal and insect resistance (Peralta et al. 2004; Arango et al. 2006). This argument has been strengthened by a plethora of studies showing that removal of heartwood components via solvent extraction results in increased biological attack of the solvent-extracted wood (Ohmura et al. 2000; Taylor et al. 2006; Oliveira et al. 2010; Kirker et al. 2013; Mankowski et al. 2016a; Hassan et al. 2016a, b, 2018b, c). Relationships between wood properties and extracted heartwood compounds have been evaluated showing they contribute to the natural durability of wood (Schultz et al. 1990; Reyes-Chilpa et al. 1998; Chang et al. 1999; Morimoto et al. 2006).

Tectona grandis L.f. (teak) is a tropical hardwood species native to Southeast Asia (Bhat et al. 2005). It is well-known for its natural resistance to termite and fungal attack (Ngee et al. 2004, Thulasidas and Bhat 2007, Lukmandaru and Takahashi 2008, Dungani et al. 2012; Lukmandaru 2017; Hassan et al. 2016b, 2017, 2018c). *Cedrus deodara* (Lamb.) G. Don (Lamb.) (Himalayan or Deodar) is listed as moderately resistant to decay (Scheffer and Morrell 1998) and is an important softwood in Pakistan. Wood from these species is widely used in Southeast Asia. In Pakistan, cedar wood is used in carpentry and construction as plywood.

Depending on the targeted service life of wooden material, naturally durable wood species with antitermitic properties may be considered an alternative to commonly used non-durable commercial timbers like southern pine (*Pinus* spp.) and cottonwood (*Populus* spp.) (Hassan et al. 2017, 2018b, c). Heartwood extracts of resistant wood species are valuable sources of antifungal and antitermitic compounds that could potentially be removed and applied to more susceptible wood species to prevent termite and fungal attack. Due to environmental concerns regarding chemical wood preservatives, the use of naturally produced chemicals from heartwood or plant extracts for the management of wood destroying organisms could be of some use in the field of wood protection. Recent studies showed that extractive compounds were directly toxic and/or repellent to several termite species and had fungicidal properties (Thevenon et al. 2001; Lukmandaru and Ogiyama 2005; Asamoah et al. 2011; Ragon et al. 2008; Dungani et al. 2012; Tascioglu et al. 2012; Kirker et al. 2013; Kadir et al. 2014, 2015; Mankowski et al. 2016b; Hassan et al. 2016a, b, 2018b, c). Moreover, these compounds were also detrimental to symbiotic protozoa in the guts of *R. flavipes* and *Heterotermes indicola* (Hassan et al. 2017).

Extracts from the selected wood species have shown antitermitic activities in previous studies (Mankowski et al. 2016b). However, very few studies have been conducted on the impregnation of these compounds to non-durable wood in order to improve their durability. The objective of this study was to evaluate the toxic potential of heartwood extracts from two naturally durable wood species (*Tectona grandis* and *Cedrus deodara*) against termites and decay fungi.

2 Materials and methods

2.1 Wood sample preparation

Heartwood from *Tectona grandis* Linn (teak) and *Cedrus deodara* (Lamb.) G. Don (Himalayan cedar) was selected. Defect free logs of *C. deodara* and cottonwood (*Populus* sp.) were purchased from a timber market located in Faisalabad, Pakistan, and shipped to United States. Marine grade *T. grandis* was acquired from a supplier in the United States (McIlvain, Pittsburg, PA, USA). Non-durable southern pine (*Pinus taeda*) and cottonwood (*Populus* sp.) were selected to test the effectiveness of extracted heartwood components as wood preservatives. Wood was taken from single trees that ranged in age from 15 to 25 years old. Logs were air dried, cut into 5–6 boards ($500 \times 150 \times 25$ mm^3), and then cut to 100 blocks measuring $19 \times 19 \times 19$ mm^3 according to AWPA Standard E1-17. For the heartwood component extractions, a section of *T. grandis* or *C. deodara* was converted into shavings (1000–1200 g) with approximate size of 1.56 mm thick × 3 mm wide × and 5–10 mm long using an electric planer (DEWALT DW733-QS) (Hassan et al. 2018b).

2.2 Preparation of extracts and solvent-extracted wood

Air-dried shavings of *T. grandis* or *C. deodara* were placed in 12-g batches in each of several Soxhlet extractors and processed according to ASTM standard D1105-96 with minor modifications (ASTM 2014). Shavings were added to ~ 20 Soxhlet apparatuses with a small pad of cotton below and above the shavings and extracted for 6 h with ethanol:toluene (2:1 v/v). The resulting aliquot was evaporated to dryness at reduced pressure by using a rotary evaporator (BUCHI, R-114) in a tared round bottom flask, and extraction yield was calculated per gram of wood shavings. A stock solution of 10 mg ml^{-1} was prepared by re-solubilizing the dried extract in ethanol:toluene (2:1) based on the dry weight of the extract and stored at 4 °C in a 1-l glass jar.

Solvent-extracted wood was prepared by following ASTM standard D1105-96 (ASTM 2014). Conditioned blocks ($19 \times 19 \times 19$ mm^3) (33 °C, 62 ± 3%) were numbered and weighed prior to placement in a Soxhlet apparatus and extracted for 6 h using 300 ml ethanol:toluene (2:1 v/v). Blocks were then washed by dipping in ethanol to remove excess toluene, returned to the Soxhlets and extracted for 6 h in ethanol (95%) alone. Ethanol-extracted blocks were air dried overnight and then boiled for 6 h in 1 l of distilled water with hourly water changes. Blocks were conditioned again at 33 °C and 62 ± 3% RH. These blocks were considered solvent extracted and were assumed to have had their soluble chemical components removed. They were used in the tests with non-extracted blocks for comparison.

2.3 Termite

Workers and soldiers of *R. flavipes* from a single colony were collected from fallen logs and dead trees at Sam D. Hamilton Noxubee National Wildlife Refuge (Mississippi) and maintained in the laboratory on southern pine at 25 °C in buckets (Hassan et al. 2017).

2.4 Choice and no-choice tests on solvent-extracted and non-extracted durable wood against termites

Solvent-extracted and non-extracted heartwood blocks of each durable wood species were subjected to choice and no-choice feeding tests according to the AWPA standard E1-17 (AWPA 2017a). Screw top jars were filled with 150 grams sand along with 27 ml distilled water and held for 2 h to equilibrate moisture. For the no-choice test, solvent-extracted and non-extracted blocks were weighed after conditioning and placed on top of the dampened sand with one block in each jar. For the choice test, each jar received one solvent extracted and one non-extracted block. Both experiments were replicated five times per wood. A total of 400 termites (396 workers and 4 soldiers) were released in each jar. Jars were incubated at 27 ± 2 °C and 75 ± 1% RH for 28 days. At the end of 28 days, the number of live termites was counted. Blocks were brushed to remove sand, conditioned for 1 week and re-weighed to determine weight loss. All blocks were visually rated using a 0–10 scale as described in the AWPA standard E1-17.

2.5 Termite bioassay with non-durable wood pressure-treated with heartwood extracts

Non-durable wood blocks were treated using the method described by Hassan et al. 2018b. Weighed and conditioned (33 °C, 62 ± 3% R.H.) southern pine and cottonwood sapwood blocks (19 × 19 × 19 mm^3) were treated with three different concentrations (2.5, 5 and 10 mg ml^{-1}) of extract from either durable wood species separately in a vacuum pressure chamber. Extracts were diluted in an ethanol:toluene (2:1 v/v) solution to the desired concentration. A vacuum (91.4 kPa) was held for 30 min, and then pressure (275.8 kPa) was applied for 60 min. Blocks treated with solvent only (ethanol-toluene) or treated with distilled water were included as control treatments. Blocks were weighed before and immediately after treatment to determine solution uptake to calculate net extract retention. The termite bioassay was conducted according to the no-choice option of AWPA standard E1-17 with some modifications and repeated five times as mentioned above in Sect. 2.4. At the end of 28 days, the number of live termites was counted. Blocks were brushed to remove sand, conditioned for 1 week and re-weighed to determine weight loss. All blocks were visually rated using a 0-10 scale as described in the AWPA standard E1-17.

2.6 Decay resistance of extracted wood and their extracts against *Rhodonia placenta* and *Trametes versicolor*

Soil bottle decay tests were conducted according to the AWPA standard E10-16 (AWPA 2017b) using either *Rhodonia placenta* (Fr.) MJ Larsen and Lombard (Mad-698-R) or *Trametes versicolor* L. (Loyd) (Mad-697). Blocks (19 × 19 × 19 mm^3) of southern pine and cottonwood were treated with extracts (2.5, 5.0 and 10 mg ml^{-1}) from the durable wood species as described in the termite bioassay and compared to non-extracted (untreated), water-treated and solvent-treated controls. Southern pine feeder strips were used to inoculate soil bottles with *R. placenta*, while red maple (*Acer rubrum* L) feeder strips were used to initiate growth of *T. versicolor*. Non-extracted blocks of the two durable wood species were compared with solvent-extracted blocks in assays where the hardwood (*T. grandis*) was challenged with the white rot fungus and the softwood (*C. deodara*) was challenged with the brown rot fungus. Soil block tests were conducted for 12 weeks for *T. versicolor* and 8 weeks for *R. placenta* at 27 °C in a 70% RH. Treatments were repeated five times. Blocks were weighed before and after experiments following conditioning at 27 °C and 30% RH to calculate weight loss. Durability indices (*DI) of 0–0.14 (very durable), 0.15–0.29 (durable), 0.30–0.59 (moderately durable), 0.6–0.89 (slightly durable, 0.9 and greater (not durable) were calculated from each test by comparing the mean weight loss of test blocks to that of non-durable blocks as referenced in EN 350-1 (1994).

2.7 Leaching resistance of extract-treated non-durable wood against termites

The effect of leaching on the activity of extracts from the treated non-durable wood blocks was assessed following AWPA standard E11-16 (AWPA 2017c) with some modifications. After conditioning, five treated blocks were submerged in 300 ml of deionized water in a 500 ml vessel and then, subjected to a 30-min vacuum (91.4 kPa) to impregnate the blocks. The vessel was subjected to mild agitation by shaker with water changes after 6, 24 and 48 and thereafter at 48-h intervals for 14 days. Blocks were conditioned

(33 °C, 62 ± 3%) for 2 weeks and a no-choice test with *R. flavipes* was conducted as described above. At the end of the test, blocks were brushed to remove sand, conditioned for 1 week (33 °C, 62 ± 3%) and re-weighed to determine weight loss All blocks were visually rated using a 0–10 scale as described in the AWPA standard E1-17. In all tests, each conditioning period shows the conditioning time required for the mass stabilization of wood blocks.

2.8 Statistical analysis

Weight loss and mortality data in the no-choice test were analyzed using 2-way ANOVA with wood type and extraction as factors, assuming blocks as independent variable. Data on the effect of leaching on no-choice termite tests were analyzed using a 3-way ANOVA with type of extract, type of non-durable wood and concentration of extracts as factors. Data from the transferable durability test were analyzed using a one-way ANOVA. However, weight loss data from the choice test were analyzed using a split-plot design with block pairs within each wood type. Termite mortality in the choice test was analyzed using Exact Wilcoxon two-sample test and Median test in SAS software. The adjusted p-values were determined via SAS simulation method. Means of treatments were separated using Tukey HSD test at the 5% level of significance.

3 Results

3.1 Choice and no-choice termite tests of solvent-extracted and non-extracted durable wood

Bioassay results for weight loss from the no-choice test showed that there was no significant interaction effect between wood type and extraction ($p = 0.07$), a significant wood type effect ($p = 0.05$), and there was also a significant extraction effect ($p = 0.00$) on wood weight loss for both wood species. Termite ignored non-extracted *C. deodara* blocks, consuming more of the solvent-extracted blocks (Fig. 1a). Weight loss for solvent-extracted *C. deodara* was significantly greater (37.13%) than non-extracted blocks (0.27%) with average termite damage rating of 8.2 and 9.2, respectively. However, the difference between wood consumption for solvent-extracted vs non-extracted *T. grandis* was not significant with average termite damage rating of 9.4 and 9.5, respectively. There was a significant interaction effect ($p = 0.00$) between the *C. deodara* extracted and non-extracted (adj. $p = 0.00$), *C. deodara* extracted and *T. grandis* non-extracted (adj. $p = 0.00$), and *C. deodara* extracted and *T. grandis* extracted (adj. p value $= 0.00$) blocks for percent mortality in the no-choice test. Complete mortality was observed in *R. flavipes* after feeding on solvent- extracted and non-extracted blocks of *T. grandis*. However, the differences between wood consumption for solvent-extracted and non-extracted *C. deodara* were significant (Fig. 1a).

Wood weight loss in choice tests showed no significant interactions between wood type and extraction ($p = 0.59$), a marginal effect for wood type ($p = 0.05$) and a significant effect for extraction ($p = 0.02$) on wood weight loss of both woods. *R. flavipes* ignored non-extracted blocks of *C. deodara* (mass loss and rating were 1.93% and 9.5, respectively) but consumed significantly more solvent-extracted *C. deodara* blocks (33.4%) with average termite damage ratings of 8.5. Weight losses for solvent-extracted and non-extracted *T. grandis* did not differ significantly from each other with 9.2 and 9.5 average termite damage ratings, respectively. Significantly higher termite mortality was observed in choice test after feeding *T. grandis* (100%) compared to *C. deodara* (64%) (Fig. 1b).

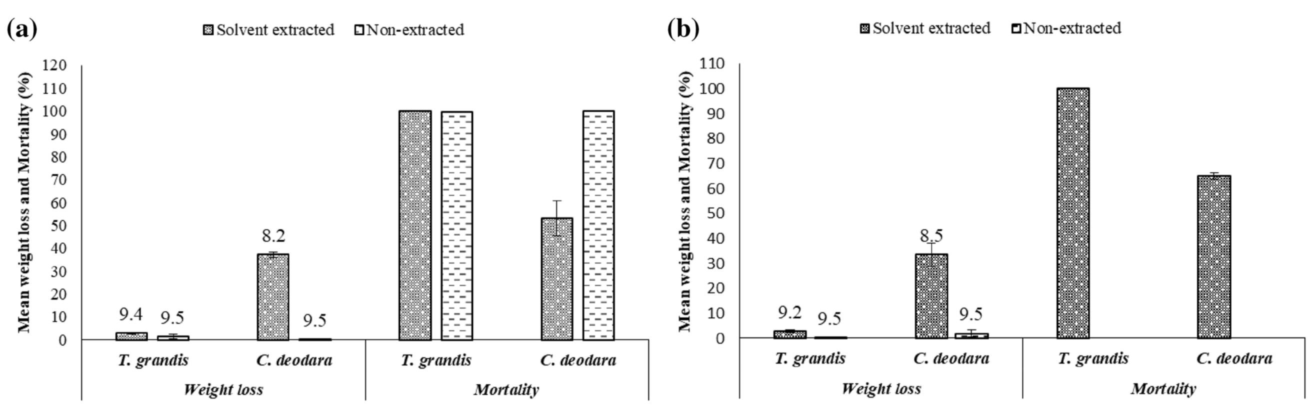

Fig. 1 Mean weight loss (%) for solvent-extracted and non-extracted blocks of two durable wood species and mortality of *R. flavipes* under no-choice (**a**) and choice tests (**b**). Values on the bars show average termite damage ratings

3.2 Termite bioassay on southern pine and cottonwood pressure-treated with extracts

Mean weight loss, retention and AWPA E-1 termite damage ratings for solvent-treated, water-treated and *T. grandis* extract-treated southern pine and cottonwood blocks exposed to *R. flavipes* are presented in Table 1. Solvent- and water-treated southern pine control blocks lost more mass with minimum termite damage ratings. Conversely, *T. grandis* extract-treated non-durable wood blocks showed substantially lower weight losses of 5.48 and 12.36%, for southern pine and cottonwood, respectively, at the highest tested extract concentration (10 mg ml^{-1}). Weight loss was inversely related to extract concentration. Average termite damage ratings at the maximum retentions for southern pine and cottonwood were 8.90 for both wood species. Weight losses in the southern pine control treatments differed significantly compared to all other treatments except for 2.5 mg ml^{-1}. Termite mortalities were not significantly different for southern pine for the rest of the treatments except at the maximum tested extract concentration of 10 mg ml^{-1} (82.8% mortality), whereas mortalities on cottonwood were significantly more in blocks treated with 10 mg ml^{-1} of extract compared to the rest of the treatments including the control (Table 1).

Mean weight loss, retention and AWPA E-1 termite damage ratings for *C. deodara* extract-treated, solvent-treated and water-treated southern pine and cottonwood exposed to *R. flavipes* are shown in Table 2. Southern pine and cottonwood treated with *C. deodara* extract showed significantly lower weight losses at the highest extract concentration (10 mg ml^{-1}) compared to the other treatments. Average termite damage ratings at the maximum retentions for southern pine and cottonwood were significantly higher compared to controls and extract treatments. *Cedrus deodara* extract was toxic to termites at the lowest concentration tested (2.5 mg ml^{-1}) with average termite damage ratings of 6.2 and 5.8 for southern pine and cottonwood, respectively. Mortality at the maximum extract concentration tested was significantly higher in southern pine with all other concentrations except for 5.0 mg ml^{-1} (Table 2).

3.3 Decay resistance of extracted wood and their extracts against *Rhodonia placenta* and *Trametes versicolor*

The hardwood, *T. grandis*, was compared to cottonwood as a non-durable control while the softwood, *C. deodara*, was compared to the southern pine controls. Cottonwood lost 47.60% of its original weight when exposed to *T. versicolor*. Non-extracted *T. grandis* was found to be highly durable with 2.9% weight loss (0.06 DI) compared to the reference material (1.0 DI) (DI = durability index as calculated in EN350). Extracted *T. grandis* was only durable (0.20 DI) with 9.53% weight loss (Table 4). The southern pine controls lost 58.29% of their original weight after exposure to *R. placenta*, while non-extracted *C. deodara* lost 8.62%. Durability indices (DI) for non-extracted *C. deodara* were 0.15 (very durable); however, durability was reduced when extracts were leached (DI = 0.84, slightly durable) and weight loss increased to 48.94% (Table 3).

Neither of the extracts tested improved decay resistance of southern pine or cottonwood. (Fig. 2a, b). Pairwise

Table 1 Mean mortality, weight loss, retention and damage rating of southern pine and cottonwood treated with *T. grandis* extract and exposed to *R. flavipes*

Woods	Conc. (mg ml^{-1})	Mortality (%)	Weight loss (%)	Retention (kg m^{-3})	Rating (avg)
Southern Pine	Water	24.85 ± 0.78^b	26.54 ± 0.81^a	–	4.6^c
	Solvent	25.25 ± 0.84^b	25.12 ± 0.39^a	–	3.6^c
	2.5	26.35 ± 1.45^b	22.90 ± 1.3^a	13.47 ± 0.24^c	6.0^c
	5.0	39.15 ± 2.78^b	18.03 ± 0.71^b	26.65 ± 0.32^b	8.0^b
	10.0	82.8 ± 10.1^a	5.48 ± 1.16^c	48.70 ± 5.7^a	8.9^a
	F	27.07	82.27	28.22	13.22
	p	0.00	0.00	0.00	0.00
Cottonwood	Water	32.10 ± 5.08^b	37.54 ± 2.00^a	–	1.6^d
	Solvent	36.20 ± 2.29^b	36.45 ± 1.05^a	–	1.6^d
	2.5	37.95 ± 7.08^b	33.08 ± 2.89^a	15.23 ± 0.26^c	5.4^c
	5.0	70.60 ± 7.48^a	20.01 ± 2.23^b	30.19 ± 0.47^b	7.8^b
	10.0	82.05 ± 7.37^a	12.36 ± 3.42^b	58.20 ± 2.08^a	8.9^a
	F	13.67	20.64	309.47	29.56
	p	0.00	0.00	0.00	0.00

Mean ± SE sharing same letters in columns for each wood species are not significantly different from each other at $p > 0.05$

Conc. concentration, *F* F value, *p* *p* value

Table 2 Mean mortality, weight loss, retention and damage rating of southern pine and cottonwood treated with *C. deodara* extract and exposed to *R. flavipes*

Woods	Conc. (mg ml^{-1})	Mortality (%)	Weight loss (%)	Retention (kg m^{-3})	Rating (avg.)
Southern Pine	Water	24.85 ± 0.78^{c}	26.54 ± 0.81^{a}	–	4.6^{bc}
	Solvent	25.25 ± 0.84^{c}	25.12 ± 0.39^{a}	–	3.6^{c}
	2.5	47.9 ± 13.2^{b}	19.57 ± 4.78^{a}	13.31 ± 0.15^{c}	6.2^{ab}
	5.0	60.5 ± 14.20^{a}	20.48 ± 3.93^{a}	27.57 ± 0.30^{b}	8.0^{a}
	10.0	58.0 ± 10.4^{a}	14.42 ± 2.53^{a}	53.24 ± 0.80^{a}	8.5^{a}
	F	2.95	2.56	1605.95	11.99
	p	0.04	0.07	0.00	0.00
Cottonwood	Water	32.10 ± 5.08^{d}	37.54 ± 2.00^{a}	–	1.6^{b}
	Solvent	36.20 ± 2.29^{cd}	36.45 ± 1.05^{a}	–	1.6^{b}
	2.5	58.45 ± 1.90^{bc}	26.28 ± 1.48^{b}	15.11 ± 0.29^{c}	5.8^{a}
	5.0	67.00 ± 8.87^{ab}	20.21 ± 2.63^{bc}	29.30 ± 0.78^{b}	6.8^{a}
	10.0	89.35 ± 7.69^{a}	15.35 ± 2.24^{c}	57.01 ± 2.23^{a}	7.8^{a}
	F	15.95	24.85	239.58	18.40
	p	0.00	0.00	0.00	0.00

Mean ± SE sharing same letters in columns for each wood species are not significantly different from each other at p > 0.05

Conc. concentration, *F* F value, *p* p value

Table 3 Mean weight loss of extracted and non-extracted durable wood species exposed to *T. versicolor* and *R. placenta* in a soil bottle assay

Wood species	Wood type	Mean weight loss (%)	DI*	Class	Fungi
Cottonwood (control)	Non-extracted	47.60 ± 1.85^{a}	1.00	Non-durable	*T. versicolor*
T. grandis (hardwood)	Non-extracted	2.90 ± 0.16^{c}	0.06	Highly durable	
	Extracted	9.53 ± 0.33^{b}	0.20	Durable	
Southern pine (control)	Non-extracted	58.29 ± 1.05^{a}	1.00	Non-durable	*R. placenta*
C. deodara (softwood)	Non-extracted	8.62 ± 1.7^{c}	0.15	Durable	
	Extracted	48.94 ± 5.12^{b}	0.84	Slightly durable	

*DI = durability index as calculated in EN350-*DI: 0–0.14 (very durable), 0.15–0.29 (durable), 0.30–0.59 (moderately durable), 0.6–0.89 (slightly durable, 0.9 and greater (not durable)

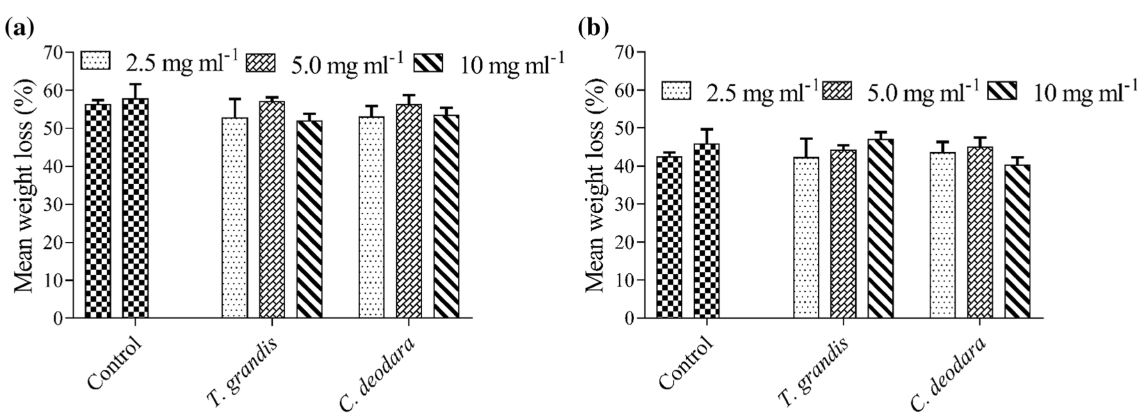

Fig. 2 Mean weight loss of southern pine blocks exposed to *R. placenta* (**a**) and cottonwood blocks exposed to *T. versicolor* (**b**) after treatment with extracts of *T. grandis* and *C. deodara*

comparisons through Tukey's HSD revealed that extract treatments did not differ significantly from one another. Extracts from the durable reference wood, *T. grandis*, did not improve durability of southern pine or cottonwood test blocks. The concentrations tested may have been below the thresholds or not effective against the fungi tested.

Table 4 Mean mortality of *R. flavipes*, weight loss and rating of leached and non-leached extract-treated southern pine and cottonwood in no-choice tests

Treatments	Mean mortality (%)				Mean weight loss (%)			
	Southern pine		Cottonwood		Southern pine		Cottonwood	
	Leached	Non-leached	Leached	Non-leached	Leached	Non-leached	Leached	Non-leached
Solvent	27.10 ± 0.21^{f}	25.25 ± 0.24^{f}	$38.60 \pm 0.88^{d–f}$	32.10 ± 5.07^{ef}	27.38 ± 0.61^{c} (4.4)	25.12 ± 0.39^{c} (3.6)	28.94 ± 1.18^{bc} (2.4)	37.53 ± 2.00^{a} (3.60)
Water	22.63 ± 0.70^{f}	24.85 ± 0.78^{f}	$37.92 \pm 0.81^{d–f}$	$36.20 \pm 2.29^{d–f}$	25.15 ± 0.55^{c} (4.80)	26.54 ± 0.81^{c} (4.6)	36.69 ± 0.86^{a} (3.2)	36.45 ± 1.05^{ab} (4.60)
T. grandis extract	$76.68 \pm 1.28^{a–c}$	82.80 ± 10.13^{a}	81.59 ± 1.33^{ab}	84.17 ± 6.53^{a}	7.65 ± 0.57^{ef} (8.10)	5.47 ± 1.16^{f} (8.9)	$9.68 \pm 0.63^{d–f}$ (7.6)	$12.35 \pm 3.41^{d–f}$ (8.90)
C. deodara extract	24.80 ± 0.46^{f}	$57.95 \pm 10.41^{b–d}$	$53.51 \pm 2.96^{c–e}$	89.35 ± 7.68^{a}	27.90 ± 0.64^{c} (4.0)	14.41 ± 2.52^{de} (8.5)	$30.41 \pm 1.30^{a–c}$ (6.6)	15.35 ± 2.23^{d} (7.80)

Values in parenthesis show mean rating for leached and un-leached southern pine and cottonwood after exposure to *R. flavipes* in no-choice tests

3.4 Leaching resistance of extract impregnated non-durable woods against termites

Mean percent termite mortality and weight losses for southern pine and cottonwood in the no-choice test for non-leached and leached extract-treated wood specimens after exposure to *R. flavipes* are shown in Table 4. Results of a 3-way ANOVA showed that there were significant interactions for wood weight losses between treatments and leaching ($p = 0.00$), treatments and type of non-durable wood ($p = 0.00$), and leaching and type of non-durable wood ($p = 0.00$). Several other interactions (treatment, leaching and type of non-durable wood) were not significantly different ($p > 0.05$). Similar results were observed for termite mortality after feeding on leached and non-leached wood. Leaching significantly affected termite resistance in both southern pine and cottonwood specimens treated with *C. deodara* extract. Increased feeding and lower mortality were observed in *R. flavipes* on leached samples compared to non-leached samples. Termite mortality from *C. deodara* extract-treated non-leached southern pine was 57.95% compared to 24.80% for leached samples. Southern pine and cottonwood treated with *T. grandis* extract did not differ significantly in mortality or weight loss between leached and non-leached samples. Termite mortality on leached and non-leached *T. grandis* treated cottonwood and southern pine differed significantly from *C. deodara* treated leached, non-leached southern pine and cottonwood. AWPA E-1 termite damage rating results showed a significant difference between leached and non-leached *C. deodara* treated southern pine and cottonwood when exposed to termites in the no-choice test. No significant differences were observed in ratings for *T. grandis* treated leached or non-leached southern pine and cottonwood after exposure to *R. flavipes* (Table 4).

Evaluation of the leached blocks against decay fungi was not conducted since extract-treated wood did not improve decay resistance.

4 Discussion

Tectona grandis showed no significant differences in feeding or mortality in *R. flavipes* when exposed to solvent-extracted and non-extracted blocks in either test. Continued durability of the blocks after solvent extraction indicated that not all of the toxic heartwood components were removed using this particular solvent system. However, sample sizes and duration of extraction have resulted in incomplete removal of extracts. A significant difference was observed for extract amount removed by using ethanol:toluene from *T. grandis* (5.51%) compared to *C. deodara* (9.67%). Lukmandaru (2011) also observed the solubility and amount of compounds removed by using different solvents. That study observed 1.9–2.7% average ethyl acetate-soluble extract content and 1.8–3.7% n-hexane soluble content from the heartwood of *T. grandis*. Bhat et al. (2010) found 9.7% extract content in the outer and 13.1% in the inner portion of *T. grandis* heartwood using ethanol as a solvent. These differences were most likely due to solvent selection and highlight the variability in recovery of extracts in different solvents.

Teak durability can be attributed to other factors such as its high density and hardness as it has been observed to remain durable after compound removal (Peralta et al. 2004; Arango et al. 2006; Kirker et al. 2013). The opposite results were observed in *C. deodara*, where there was a significant difference in weight loss and termite mortality after feeding on solvent-extracted and non-extracted blocks. Similarly, when solvent extracted and non-extracted blocks were exposed to two decay fungi, *T. grandis* (non-extracted) was reduced a durability class from highly durable to durable after extraction. Similarly, *C. deodara* was reduced from durable to slightly durable after solvent extraction. Taylor et al. (2006) also observed that removal of methanol soluble heartwood components of *Thuja plicata* Donn ex D. Don and *Chamaecyparis nootkatensis* (D. Don) reduced its resistance to fungal and termite attack. The current results mirror other studies showing that removal of extracts from heartwood decreased resistance to termites and fungi (Yamamoto and Hong 1989; Hwang et al. 2007; Kim et al. 2009; Mohareb et al. 2010; Syofuna et al. 2012; Kibet et al. 2013; Zaharin 2013; Hassan et al. 2016a; Mankowski et al. 2016a; Roszaini et al. 2016; Hassan et al. 2018b, c).

Extract-treated non-durable southern pine and cottonwood showed increased resistance to termites compared to non-extract treated wood of the same species. Weight losses of either non-durable wood were reduced at the highest extract concentration tested with an average rating > 8. The current results are in agreement with earlier work where *T. grandis* extracts were shown to improve the durability of non-durable wood against biological attack (Thevenon et al. 2001; Lukmandaru and Ogiyama 2005; Lukmandaru and Takahashi 2008; Asamoah et al. 2011; Mankowski et al. 2016a, b; Ismayati et al. 2016; Brocco et al. 2017; Hassan et al. 2017, 2018c). Akhtar (1981) studied the effects of wood, wood extracts, and essential oils from *Dalbergia sissoo* (Roxb.), *Pinus wallichiana* A.B. Jacks, and *C. deodara* against *Coptotermes heimi* (Wasmann) and found that hexane, water, and acetone extracts of *C. deodara* were more toxic to termites. Akhtar and Jabeen (1981) showed similar results on the feeding responses of *Bifiditermes beesoni* (Gardner) on wood and wood extracts of three species, i.e. *D. sissoo*, *C. deodara*, *P. wallichiana* in the laboratory.

Previous chemical analyses have shown high concentrations of anthraquinone and squalene in the extracts of *T. grandis* and three sesquiterpenes, cuprenene, himachalene and cedrene, in extracts of *C. deodara* heartwood (Hassan et al. 2017). These compounds have strong biological activity against insects and other organisms. Anthracenedione (anthraquinone) and squalene have biocidal activity against termites and decay fungi. Similarly, sesquiterpenes have been found to be antifeedant, repellant and also illicit behavioral responses in subterranean termites (Lukmandaru and Ogiyama 2005; Bhat et al. 2010; Zhu et al. 2010; Mankowski et al. 2016b; Hassan et al. 2017). The majority of active compounds identified in heartwoods are also powerful antioxidants and inhibited glutathione S-transferase activities in the gut of *Heterotermes indicola* (Hassan et al. 2018a).

Tectona grandis and *C. deodara* extracts showed no protective effects on non-durable southern pine or cottonwood at the extract concentrations evaluated in the present decay test. This may be due to tested extract concentrations being below the required threshold to be effective against the two decay fungi tested (Mankowski et al. 2016a). Previous studies showed that *T. grandis* extracts were effective against decay fungi (Sumthong et al. 2008; Adegeye et al. 2009; Astiti and Suprapta 2012; Brocco et al. 2017; Lukmandaru 2017). However, few studies have examined the antifungal properties of wood extracts from *C. deodara*. Antifungal properties derived from extracts of *C. doedara* have been reported to be effective against *Laetiporus sulphureus*, *Trametes versicolor*, *Aspergillus fumigatus* and *Candida albicans* (Chowdhry et al. 1997; Cheng et al. 2005; Parveen et al. 2010; Gupta et al. 2011). Leaching of *T. grandis* extract-treated southern pine and cottonwood specimens did not reduce resistance against termites, since there were no significant differences for termite mortality or wood weight loss between leached and non-leached samples. Conversely, leaching significantly affected termite resistance against both southern pine and cottonwood treated with *C. deodara* extract. There was a significant difference in feeding rates and mortality for *R. flavipes* on southern pine and cottonwood treated with *C. deodara* extracts. Reduced post leaching performance might be due to hydrolysis of extract compounds during leaching process and/or physical attachment of extracts with susceptible wood instead of chemical bonding (Lupsea et al. 2012; Yeniocak and Suleyman 2018), but it is a future research objective.

5 Conclusion

Transferring durability using toxic *T. grandis* and *C. deodara* heartwood extracts to non-durable wood species improved the resistance against termites but not against decay fungi at tested concentrations. Future work will need to focus more on solubility, fixation and leachability of the individual components of these extracts in order to predict their utility as wood protectants. Future studies will also examine single extract component isolates to determine whether they are effective alone or act synergistically with other heartwood components.

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Compliance with ethical standards

Conflict of interest All authors declare no conflict of interest.

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