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COMBINED EFFECTS OF NEEM (*Azadirachta indica*) AND SESAME *(Sesamum indicum)* OIL AS A WOOD PRESERVATIVE ON SUBTERRANEAN TERMITES IN THE FIELD

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ABSTRACT

The current study was aimed to evaluate the effect of neem and sesame oils on the improvement of poplar (*Populus* spp.) wood resistance to subterranean termite in the field. Concentrations of 10 %, 20 %, and 30 % of both oils alone in toluene and their different combinations along with heat treatment were bio-assayed against termites. Results showed that both oils protected woods against termites differently; however, neem oil was more protectant than sesame oil. Moreover, neem oil, when combined with sesame oil, significantly improved the wood resistance compared to separate oils treatment. Heat treatment alone was not enough to enhance resistance against subterranean termites. The treatment at 200 ºC for 6 h combined with 75 % neem and 25 % sesame (3:1) appears to be the best treatment condition in this investigation. It is concluded that neem oil can be useful as a wood preservative against subterranean termites combined with sesame oil and heat treatment.

Keywords: Heat treatment, poplar, plant oils, subterranean termites, wood resistance, wood preservative.

INTRODUCTION

Subterranean termites are serious pests of structural wood and wood-based products in tropical and sub-tropical countries. The termite fauna of Pakistan consists of 53 species. Of these 13 subterranean species are reckoned pests of agriculture, buildings, and forestry (Hassan 2017). The magnitude of damage by different species has not been enumerated; however, the most damaging species are *Coptotermes heimi* (Wasmann), *Odontotermes obesus* (Rambur), *Heterotermes indicola* (Wasmann) and *Microtermes obesi* Holmgren. These are the most common infestation species in the urban area that readily attack structures and buildings due to their aggressive behavior (Manzoor *et al*. 2010, Rasib *et al*. 2017).

The concept of wood protection against biodegradation is based on the principle of toxicity. This includes the impregnation of wood with traditional synthetic biocides such as creosote, copper chromium arsenate (CCA), and pentachlorophenol (PCP) to prevent degradation by insect and decay fungi (Jones and Brischke 2017). However, the use of traditional wood preservatives is nowadays subject to environmental and consumer constraints. Some of these chemicals have been limited by the commercial markets around the world (Ahmed *et al.* 2020, Koski and Ahonen 2008). Wood treated with common preservatives is classified as hazardous waste in some countries. These contemplations have stimulated academics and the wood preservation industry to look for new eco-friendly alternatives (Hassan *et al*. 2020, Hassan *et al.* 2017, Himmi *et al*. 2013).

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Various domestically produced vegetable oils have been used for wood impregnation against termites and decay fungi. Still, those of importance are neem, linseed, rapeseed, tall oil, and their derivates, palm, soybean, and coconut oil (Panov *et al*. 2010). Vegetable oils have been used for rapid heat transfer in wood during vacuum pressure treatment (Archer and Lebow 2006, Ibach and Rowell 2001, Kaur *et al*. 2016). These are not only suggested as a heating medium but also acts as toxic chemicals against termites (Hassan 2017, Hassan *et al*. 2020, Lee *et al.* 2018, Sousa *et al.* 2019). Pressure impregnation of wood with linseed, neem, jatropha, and jojoba oils significantly reduced wood weight loss of *Acacia nilotica*, *Dalbergia sissoo,* and *Pinus wallichiana* and increased termite mortality (Ahmed *et al*. 2020). Oil from the nut of the kukui plant (*Aleurites moluccana*) acts as a feeding deterrent rather than a toxic agent. Non-toxic vegetable oils decrease water uptake of wood, but penetration is not enough without heat (Kartal *et al*. 2006). Surface application and cold immersion of wood in vegetable oil have been demonstrated to reduce weight loss by subterranean termites with or without heating (Abbas *et al.* 2016, Ahmed *et al.* 2014).

However, the high rate of photodegradation, leaching, short stability time, and slow killing rates of neem oil restricts its replacement for conventional pesticides (Campos *et al.* 2016). Neem oil was ineffective as a wood preservative against termites and decay fungi due to leaching in the soil (Machado *et al.* 2013). Sesame oil is highly absorbent and protects the wood against weathering (Ozgenc *et al*. 2013). To complement the benefits of oils in combination for outdoor wood preservation, the mixture of oils or organic acid has been applied to several wood species against rot fungi and termites (Hussain *et al*. 2013, Teaca *et al.* 2019). Still, given the fact that oils only fill the wood cavities by capillarity, not being chemically bonded to the wood cell walls, it is necessary to ensure a high ability of oil retention to acquire the required protection (Demirel *et al*. 2016). Therefore, present studies were carried out to determine the efficacy of the combination of oils to prevent termites from attacking the wood.

MATERIALS AND METHODS

Wood species and oils

Defect-free logs of Poplar (*Populus* spp.) were purchased from the timber market located at Jhang Road, Faisalabad. Sapwood and heartwood were visually distinguished and separated from each other using an electric saw. Sapwood samples were cut into smaller specimens with the dimension of 10 cm × 4 cm × 2 cm (longitudinal × radial × tangential). The initial moisture contents of wood were determined according to the method described in Ahmed *et al.* (2014). The wood specimens were pooled and randomly allocated into treatment groups. Before treatment, all samples were conditioned (33 °C; 62 % ± 3 % RH) to constant mass.

Seeds of neem (*Azadirachta indica* A. Juss) and sesame (*Sesamum indicum* L.) were collected within the vicinity of Faisalabad, Pakistan (latitude 31,4504° N, longitude 73,1350° E), a region with average 375 mm annual rainfall, very hot and humid summers and dry cool winters. Oils were extracted by cold pressing from plant material collected and were used unmodified.

Treatment of wood

Experiment No 1: Wood specimens were labeled using marking engraved on the aluminum sheet, which was nailed on them. Neem and sesame oils were separately applied to wood specimens by dipping in the treatment solution for 24 h. Treatment solutions consisted of three concentrations of 10 %, 20 %, and 30 % of both oils separately in toluene. The specimens were removed, blotted to remove excess solution, weighed to determine solution uptake according to Ahmed *et al.* (2020), and wrapped in plastic bags before conditioning. The same procedures were used with test specimens that were submitted to solvent control treatments. Samples were then conditioned at 33 °C; 62 % ± 3 % RH to a constant weight.

Experiment No. 2: In another experiment, neem and sesame oils at 30 % concentration were applied in combination in the following order respectively; 1:1, 1:2, 1:3, 2:1, and 3:1 in a similar way as described above.
Experiment No. 3: For the oil-heat treatment, the specimens were heated at three temperatures (100 °C, 150 °C, and 200 °C) in an oil bath containing neem and sesame oil (30 %). On the oil reaching the desired temperature, the wood specimens were immersed in it for 2 h, 4 h, and 6 h, and then cooled off for 15 minutes in the bath. The rest of the procedure is the same as described above.

Experiment No 4: Specimens were heat-treated with oil (3:1) as described above.

The list of treatments and retention levels in each experiment is given in Table 1.

Termite filed test

The field test was carried out at Post Graduate Agricultural Research Station, Faisalabad. Treated and untreated specimens were placed in an underground concrete pit previously designed for termite experiments (Ahmed *et al.* 2020, Ahmed *et al.* 2014, Fatima *et al.* 2015, Zulfiqar *et al.* 2020). A soil layer was spread in the pit along with attracting materials to gain access to termites to the pit. With the start of the foraging of termites, stakes were placed in five replicates for 60 days. At the end of the bioassay, test specimens were removed from the pits and cleaned. Test specimens were then oven-dried under the same conditions used to obtain the initial masses. After cooling and weighing the test specimens, the final and initial mass was compared to determine the percentage of mass loss using Equation 1.

$$Weight\ loss(\%) = \frac{W_1 - W_2}{W_1}\ x\ 100 \quad (1)$$

Where
W_1= Initial weight of stakes
W_2= Final weight of stakes.

Statistical analysis

Analysis of variance (ANOVA), with a significance level of 5 percent, was used to verify the effect of treatments on the weight loss of the wood. Treatment means were separated using Tukey's HSD in GraphPad Prism.

Table 1: List of controls, treatment formulations, and retentions of neem and sesame oil used to treat stakes.

	Treatments	Retention (kg/m^3)
Experiment #1	Solvent treated	0
	Untreated	0
	Neem oil (10 %)	17,90
	Neem oil (20 %)	18,01
	Neem oil (30 %)	18,99
	Sesame oil (10 %)	21,23
	Sesame oil (20 %)	21,38
	Sesame oil (30 %)	22,21
Experiment # 2	Solvent treated	0
	Untreated	0
	Neem oil: Sesame oil (1:1) at 30 %	20,39
	Neem oil: Sesame oil (1:2) at 30 %	21,27
	Neem oil: Sesame oil (1:3) at 30 %	23,24
	Neem oil: Sesame oil (2:1) at 30 %	19,98
	Neem oil: Sesame oil (3:1) at 30 %	20,45
Experiment # 3		
	Heat-treated without oil (120 °C, 150 °C, 200 °C) for 2 h, 4 h, and 6 h	0
	30 % Neem oil (100 °C); 2 h	23,39
	30 % Neem oil (100 °C); 4 h	24,10
	30 % Neem oil (100 °C); 6 h	24,90
	30 % Neem oil (100 °C); 2 h	25,10
	30 % Neem oil (150 °C); 4 h	26,30
	30 % Neem oil (150 °C); 6 h	26,60
	30 % Neem oil (200 °C); 2 h	26,01
	30 % Neem oil (200 °C); 4 h	26,20
	30 % Neem oil (200 °C); 6 h	27,00
	30 % Sesame oil (100 °C); 2 h	25,80
	30 % Sesame oil (100 °C); 4 h	26,20
	30 % Sesame oil (100 °C); 6 h	26,50
	30 % Sesame oil (150 °C); 2 h	27,70
	30 % Sesame oil (150 °C); 4 h	28,10
	30 % Sesame oil (150 °C); 6 h	28,23
	30 % Sesame oil (200 °C); 2 h	29,20
	30 % Sesame oil (200 °C); 4 h	29,90
	30 % Sesame oil (200 °C); 6 h	30,20
Experiment #4	Heat-treated without oil (100, 150, 200 °C)	0
	3:1 Neem oil: Sesame oil (100 °C); 2 h	24,99
	3:1 Neem oil: Sesame oil (100 °C); 4 h	25,41
	3:1- Neem oil: Sesame oil (100 °C); 6 h	26,49
	3:1- Neem oil: Sesame oil (150 °C); 2 h	26,40
	3:1- Neem oil: Sesame oil (150 °C); 4 h	26,27
	3:1- Neem oil: Sesame oil (150 °C); 6 h	27,28
	3:1- Neem oil: Sesame oil (200 °C); 2 h	28,10
	3:1- Neem oil: Sesame oil (200 °C); 4hrs	29,19
	3:1- Neem oil: Sesame oil (200 °C); 6 h	30,10

RESULTS AND DISCUSSION

Mean weight losses caused by the termites to poplar wood test specimens after treatment with different concentrations of neem and sesame oils are presented in Figure 1a. Termites caused extensive damage to solvent treated and untreated test specimens, with weight losses ≥ 80 %. In comparison, the specimens treated with oils had significantly fewer weight losses. Weight losses of test specimens significantly decreased with the increasing concentrations of both oils (sesame oil: F=13,34; p=0,03; neem oil F=123,45; p: 0,001). Those treated with 30 % concentration have significantly reduced attack of termites compared to the other two concentrations. However, specimens treated with neem oil at each concentration were more resistant to termites than sesame oil concentrations. Minimum weight loss of specimens treated with 30 % neem oil was 21,3 % compared to 38,8 % weight loss after treatment with 30 % sesame oil.

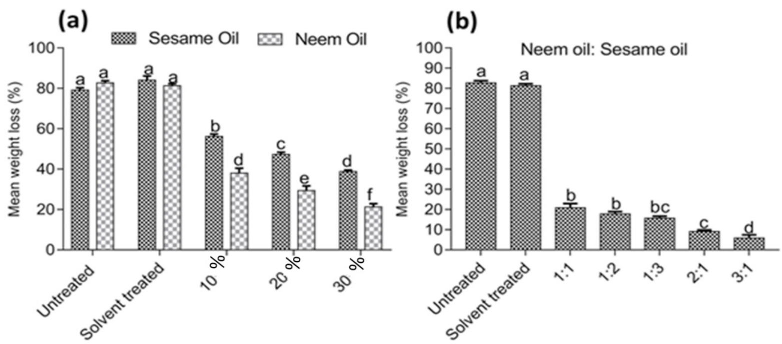

Figure 1: Mean weight losses in poplar wood samples treated with different concentration of neem and sesame oil (a) and their combination (b) subjected to bioassay against termites in the field.

Results of the bioassay with poplar wood treated with neem and sesame oils at 30 % concentration when applied in different combinations are presented in Figure 1b. Solvent treated and untreated specimens lost >80 % weight compared to other treatments (F= 129,29; p= 0,004). Mixtures of oils perform differently at each oil combination. Test specimens treated with a mixture of oils containing 75 % neem oil and 25 % sesame oil (3:1) showed more resistance to termites with 5,90 % weight loss than the rest of the treatments. Results showed that different oil combinations with an increasing proportion of sesame oil (1:1, 1:2, 1:3) had a non-significant effect on the resistance of wood compared to combinations with the increasing neem oil proportion (2:1, 3:1).

Results of the bioassay with poplar specimens treated with heat, 30 % sesame, and neem oil showed that only heat treatment also has the same effect on resistance against termites (Figure 2). Controls specimens heated at 150 °C for 2 h, 4 h, and 6 h showed higher weight loss (>70 %) compared to controls heated at 200 °C for 4 h and 6 h (<65 %). Wood heated at different temperatures along with sesame and neem oils showed significantly more resistance to termites compared to all control treatments (sesame oil: F=112,32; p=0,03; neem oil F=98,45; p: 0,02). Mean weight losses of specimens treated with hot neem oil for 6 h at 200 °C showed significantly lower weight loss (8,1 %) than the rest of treatments with this oil. Similarly, test specimens treated with hot sesame oil at 200 °C for 6 h showed minimum weight loss (12,3 %). Neem hot oil-treated specimens had much lower weight losses than sesame oil-treated samples at the thermal treatment of 100 °C, 150 °C, and 200 °C for different time intervals.

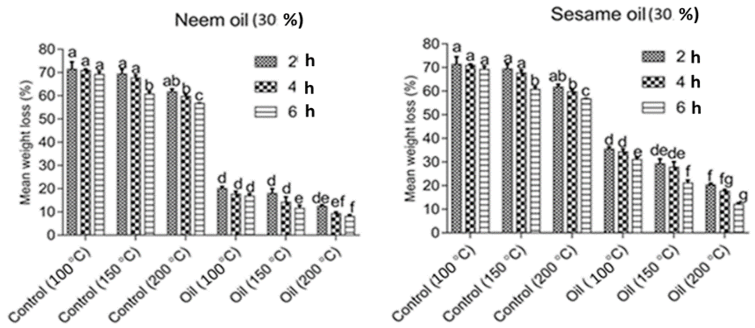

Figure 2: Mean weight losses in poplar wood samples treated with 30 % neem and sesame oil along with different levels of heat treatment for three-time durations subjected to bioassay against termites in the field.

Results showed that the dipping time of test specimens and temperature both significantly affected wood weight loss. Exposure of the samples to higher temperature levels and more dipping time increased the termite resistance compared to lower levels.

Weight loss of specimens treated with 75 % neem and 25 % sesame oil (3:1) heated at different temperatures (100 °C, 150 °C, and 200 °C) for 2 h,4 h, and 6 h was significantly lower than control specimens (F= 235,89; p=0,008). Minimum weight loss of specimens was observed after treatment with hot oil (3:1) at 200 °C when samples were dipped for 6 h before exposure to termites (Figure 3).

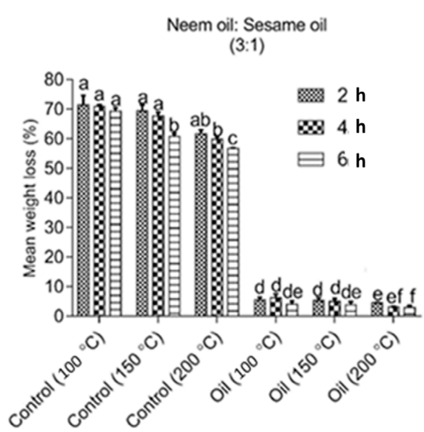

Figure 3: Mean weight losses in poplar wood samples treated with neem and sesame oil (3:1) along with different levels of heat treatment for three-time durations subjected to bioassay against termites in the field.

Increasing eco-friendly directives and public health apprehension concerning the toxicity of synthetic wood preservatives has resulted in efforts to the development of less toxic wood preserving methods based on natural compounds (Hassan *et al*. 2020). In the current study, we tested the combined effect of sesame and neem oil with or without heat treatment to protect wood against subterranean termites in the field. Non-durable poplar wood showed increased resistance to termites after treatment with sesame oil, neem oil, and with mixtures of sesame and neem oil, compared with non-oil–treated wood. Similarly, hot oil treatment of both oils alone and in the mix also increased resistance against termites compared to heat-treated only wood. However, wood samples were more resistant to termites after treatment with a mixture of neem and sesame oil with heat or without heat treatment compared to the rest of the treatments. Previous studies showed that neem oil is ineffective as a wood preservative when used alone against termites and decay fungi due to leaching or low doses (Grace and Yates 1992, Machado *et al.* 2013). Many authors proposed combining neem oil with other chemicals/compounds to produce less leachable compounds or increased wood protection. Neem oil when combined with castor bean oil or copper and cashew nutshell liquid produced protection against the termites (Paes *et al.* 2012, Venmalar and Nagaveni 2005). Therefore, we combined neem with sesame oil to determine their combined effect. The combination of sesame oil with neem oil significantly improved the resistance of wood compared to neem oil alone. Unlike neem oil, sesame oil is non-toxic and not has been used against termites until this study. However, it is highly absorbent and protects the wood against weathering (Ozgenc *et al.* 2013). In the current study, it provided some resistance to polar wood against termites. One possible mechanism of non-toxic oils could be creating a hydrophobic barrier that resulted in the displacement of water in woods with oil and prevented termite feeding (Ahmed *et al.* 2020, Lyon *et al.* 2007).

Plant oils are considered suitable as heat transfer media for the heat treatment of wood since the boiling temperature of numerous oils is higher than 260 °C (Bazyar 2012, Gunstone 2011). In the current study, plant oils combined with heat treatment led to more significant improvement in the resistance of wood to subterranean termites than heat treatment alone or only oil treatment without heat. Several studies have reported that oil heat treatment improved resistance to decay fungus (Bazyar 2012, Candelier *et al.* 2016, Hao *et al.* 2018, Manalo and Garcia 2012). However, most studies on oil heat treatment of susceptible wood relate only to physical and mechanical properties, and very few on resistance against termites (Manalo and Garcia 2012). Current study the first report in which the combined effect of two oils and heat treatment is used to determine the resistance of wood against subterranean termites in the field.

CONCLUSIONS

The 8-week field termite exposure test was conducted to determine the effects of oil heat treatment using neem and sesame oil. Results showed that treatment of poplar wood with a combination of oils and heat treatment could significantly improve the resistance of wood against termites. Results also indicated that increasing the heat treatment process from 2 h to 6 h has significant effects on the resistance of poplar wood.

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