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Termicidal Activity and Chemical Components of Wood Vinegar from Nipah Fruit against *Coptotermes curvignathus*

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ABSTRACT

The termicidal activity and chemical components of wood vinegar from two sources of biomass, nipah fruit shells (NFS) and a mixture of shells and fiber (MSF), were evaluated against *Coptotermes curvignathus*. A no-choice test was carried out to evaluate their termicidal activity using filter paper samples treated with 2.5%, 5.0%, 7.5%, 10.0%, and 12.5% NFS or MSF vinegar. Both wood vinegars exhibited antitermitic activity against *C. curvignathus*. The results show that increased concentrations of NFS and MSF vinegar significantly increased termite mortality. In particular, the NFS vinegar caused complete mortality and the lowest filter paper mass losses at 2.18% when treated with 12.5% wood vinegar. The most abundant chemical compounds of NFS vinegar were cyclopropanecarbonyl chloride, 2,5-dichlorophenol, 2-propanone, acetic acid, propanoic acid, benzenesulfonic acid, 3,7-dimethyl-6-octenal, and trans-geraniol. Meanwhile, the main compounds in the MSF vinegar were 1,2-ethanediol, formic acid, acetic acid, ethanoic acid, 2-furancarboxaldehyde, phenol, 2-methoxy phenol, and 4-methyl phenol.

Keywords: antitermitic activity, *Coptotermes curvignathus*, mortality, nipah fruit, nipah fruit fiber, wood vinegar

1. INTRODUCTION

The preservation of wood using synthetic chemicals has long been used to protect wood from destructive organisms. However, the toxicity and risks to human health as well as the environmental impact of the use of synthetic chemicals have limited their use in many countries (Coles *et al.*, 2014). Grewal *et al.* (2018) suggested that natural products from bio-renewable resources need to be developed as alternative protective agents. Chemicals, such as wood vinegar, liquid smoke, and bio-oil, are produced as the result of wood or other biomass burning at a high temperature. In addition, burning wood in the absence of oxygen produces by-products like charcoal, tar, and other gaseous chemical components. Due to their complex chemical compounds, wood vinegar is hypothesized to protect wood from fungal and termite attacks. There are numerous studies that have tested their effectiveness against termites (Adfa *et al.*, 2020; Arsyad *et al.*, 2020; Kadir *et al.*, 2022; Subekti and Yoshimura, 2020), other insects (Urrutia *et al.*, 2021), fungi (Adfa *et al.*, 2020; Oramahi *et al.*, 2020a; Sharip *et al.*, 2016),

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and bacterial attack (Misuri and Marri, 2021; Suresh *et al.*, 2019).

Sharip *et al.* (2016) reported that condensates from superheated steam pre-treatment of oil palm mesocarp fiber at 190℃–240℃ showed antifungal activities. Antifungal tests against *Aspergillus fumigatus* and *Trichoderma asperellum* using the agar dilution method at 10% (10 mL wood vinegar: 90 mL Potato Dextrose Agar /PDA solution), and against *Ganoderma boninense* using the spore germination method at a 1:1 ratio (spore suspensions of 10^6 with PDA solution) showed promising results. Wood vinegars from a mixture of softwoods exhibited antibacterial activity against *Escherichia coli*, *Enterobacter aerogenes*, *Pseudomonas aeruginosa*, *Listeria monocytogenes*, and *Esterococcus faecalis.* The principal antibacterial compounds of these wood vinegars were acetic acid, vanillin, and furfural (Suresh *et al.*, 2019). Wood vinegar from sunflower seed hull waste demonstrated insecticidal activity against *Sitophilus orizae*, *Lasioderma serricorne*, and *Tribolium castaneum* (Urrutia *et al.*, 2021).

Temiz *et al.* (2013) revealed that giant cane (*Arundo donax* L.) contains acids, ketones, furans, benzene, phenols, sugars, and guaiacols, which all exhibited antitermitic activity against *Reticultermes flavipes.* Adfa *et al.* (2020) reported the antifungal and antitermitic activities of wood vinegar from *Cinnamomum parthenoxylon* stem wood against *Schizophyllum commune*, *Fomitopsis palustris*, and *Coptotermes curvignathus*. Recently, Kadir *et al.* (2022) reported that wood vinegar from *Dyera costulata* produced at 500℃ showed antitermitic activity. Termites are important bio-deterioration agents of wood and wood products. Therefore, the development of alternative bio-renewables is needed to protect wood and wood products from termites, *C. curvignathus*.

To the best of our knowledge, there are no reports on the chemical characterization of wood vinegar from shells or a mixture of shells and nipah fruit fiber, in terms of antitermitic activity. The aim of this study was to evaluate the termicidal activity and chemical components of wood vinegars from shells and a mixture of shells and nipah fruit coir fiber against the subterranean termite, *C. curvignathus*.

2. MATERIALS and METHODS

2.1. Material preparation and wood vinegar production

The preparation of nipah palm shells (NFS) and a mixture of shells and fiber (MSF) was carried out at the Wood Workshop Laboratory of the Faculty of Forestry, Tanjungpura University, Pontianak. The raw materials (NFS and MSF) were obtained from Kubu Raya Regency, West Kalimantan, Indonesia. NFS and MSF were ground into particles using a 425 micrometer sieve and retained 250 micrometer. The no-choice bioassay procedure was carried out at the Wood Technology Laboratory, Faculty of Forestry, Tanjungpura University. The wood vinegar was made using the pyrolysis method, according to methods outlined in previous studies (Darmadji and Triyudiana, 2006; Oramahi *et al.*, 2021), which was carried out at the Engineering Laboratory, Faculty of Agricultural Technology, Gadjah Mada University, Yogyakarta. This included pouring approximately 5 kg NFS and MSF particles into an aluminum reactor, setting the aluminum condenser circuit, and heating the furnace to 420℃ for 150 min.

2.2. Characterization of wood vinegar

The chemical components of NFS and MSF wood vinegar were analyzed using gas chromatography with mass spectrometry (GC-MS; QP-210S, Shimadzu Manufacturing, Kyoto, Japan). The GC-MS conditions were as follows: Stabilwax-DA Capillary Column (silica fusion) 30 m × 0.25 mm; injection temperature, 250℃; column temperature, 60℃–200℃, increasing by 10℃/min; and helium flow rate, 40.0 mL/min. The GC-MS was utilized in the electron ionization mode at 70 eV with an interface temperature of 200℃. Eleven samples were injected into the column and maintained at 60℃–200℃ with an increase of 5℃ per min. Compound identification was carried out by comparing MS spectral data with standard library data (Mun and Ku, 2010) and quantified using the integrated peak areas.

2.3. Determination of total phenol

Total phenol was determined for each wood vinegar using the Folin-ciocalteu reagent assay (Theapparat *et al.*, 2019). The absorbance was read with a spectrophotometer (UV-1601 UV-VIS Spectrophotometer, Shimadzu Manufacturing) at a wavelength of 750 nm. The total phenol content of the sample solution was calculated based on the standard curve obtained from the pure phenol solution.

2.4. Determination of total acidity

The total acidity in NFS and MSF wood vinegar was carried out according to the AOAC (1990). NFS and MSF wood vinegar (1 mL) were diluted with distilled water (total volume 100 mL). The solution was titrated with standard 0.1 N NaOH solution until the pH reached 8. The total acid content is expressed in percentage weight of acetic acid.

2.5. Effect of wood vinegar on anti-termite activity testing

Mature workers and soldiers of *C. curvignathus* Holmgren were collected from infected trees in the Ambawang area, Kubu Raya district, West Kalimantan, Indonesia. An optional test procedure (Ganapaty *et al.*, 2004) was used to evaluate the termicidal qualities of NFS and MSF wood vinegar. Diluted wood vinegar (2.5%, 5.0%, 7.5%, 10.0%, and 12.5%, v/v) was pipetted and dripped onto filter paper (Whatman No. 1, 55 mm diameter). The treated filter paper was placed in a Petri dish and 30 workers and 5 soldiers of *C. curvignathus* Holmgren were added to each. Filter paper treated with DI alone was used as a control. All destilat water and vinegar mixtures were diluted to 0.3% (v/v) and the test dishes were then covered and placed in an incubator with approximately 70%–85% humidity in the dark at 27 + 2℃. The number of dead termites was calculated every day for 21 days. Termite mortality and filter paper mass loss were calculated on mean and expressed as a percentage (%). Each measurement was replicated three times, and the filter paper mass loss was calculated at the end of the experiment.

3. RESULTS and DISCUSSION

3.1. GC analysis results

The chemical compounds in NFS and MSF vinegar are shown in Tables 1 and 2, respectively. The most abundant compounds in NFS vinegar were cyclopropanecarbonyl chloride, 2,5-dichlorophenol, 2-propanone, acetic acid, propanoic acid, benzenesulfonic acid, 3,7-dimethyl-6-octenal, and trans-geraniol. Meanwhile, the main compounds in the MSF vinegar were 1,2-ethanediol, formic acid, acetic acid, ethanoic acid, 2-furancarboxaldehyde, 2-methoxy phenol, 4-methyl phenol, and phenol.

Oramahi *et al.* (2020a) characterized the main components of wood vinegar made from *Shorea leavis* at 400°C; these were acetic acid, 1-hydroxy-2-propanone, 2-furancarboxaldehyde, 3-pentanone, phenol, 2-methoxy phenol, 2-methoxy-4-methyl phenol, 2,6-dimethoxy phenol, and 1,2,4-trimethoxybenzene. Aguirre *et al.* (2020) used GC-MS to analyze wood vinegar made from feedstock, such as pine, poplar, forest pine residues, and urban pruning waste; the main constituents were acetaldehyde, acetic acid, 2-propanone, propanoic acid, 1,2-

Table 1. Chemical components of wood vinegar made from nipah fruit shells

No	Retention time (minute)	Wood vinegar compounds	Peak area (%)
1	1.905	Cyclopropanecarbonyl chloride	13.54
2	2.568	2,5-Dichlorophenol	5.61
3	3.560	2-Propanone	32.96
4	7.393	Acetic acid	18.08
5	9.366	1-Hydroxy-2-propanone	2.21
6	11.617	Propanoic acid	2.88
7	14.150	1-Hydroxy-2-butanone	3.28
8	17.236	2-Furancarboxaldehyde	2.11
9	18.794	2-Furanmethanol	2.19
10	26.849	Benzenesulfonic acid	3.60
11	28.450	Octadecanoic acid	1.92
12	30.987	3,7-Dimethyl-6-octenal	5.99
13	35.314	Trans-geraniol	5.63

ethanediol monoacetate, 1-hydroxy-2-butanone, succindialdehyde, furfural, 2-cyclopenten-1-one, 2(5H)-furanone, phenol, 2-methoxyphenol, cresol, 2,6-dimethoxy phenol, and levoglucosan. The main compound of wood vinegar from NFS dan MSF were same with Oramahi *et al.* (2020a) and Aguirre *et al.* (2020) results ie : acetic acid, 2-methoxy phenol, and phenol.

Maliang *et al.* (2021) reported on GC-MS analysis of bamboo tar, which found that the main components were 2,6-dimethoxyphenol, 2- or 4-ethylphenol, 2- or 4-methylphenol, phenol, 4-ethylguaiacol, dimethoxyphenol, 4-methylguaiacol, 4-propenyl-2,6-dimethoxyphenol, and 2,4-dimethylphenol. Baharom *et al.* (2020) carried out GC-MS on wood vinegars made from *Cocos nucifera*, *Averrhoa carambola*, and *Mangifera indica;* these were different in chemical composition and active compounds. The main compounds identified in *C. nucifera* shells were furfural, phenol, benzofuran, acetic acid, hexanal, ethanone, and formic acid. In *A. carambola*, the abundant compounds are furfural, imidazole, 3-pyridinecarboxaldehyde, benzaldehyde, phenol, benzofuran, indene, acetic acid, indazole, naphthalene, cyclohexanecarboxylic acid, palmitamide, palmitic acid, heptadecanenitril, and sterylamide. Meanwhile the dominant components in *M. indica* are toluene, furfural, imidazole, annulene, benzaldehyde, phenol, carbamic acid, acetic acid, naphthalene, heptadecanenitril, and stearylamide. Lu *et al.* (2019) characterized the main organic components in wood vinegar from *Cunninghamia lanceolata* (Lamb.) Hook. waste as acid, phenols, alcohols, ketones, aldehydes, and esters. Wood vinegar obtained from *Acacia mangium* contains acetic acid, methanol, phenol, o-cresol, furfural, and cyclohexane (Nurhayati *et al.*, 2005).

The main compounds found in liquid smoke from *Dyera costula* were benzyl alcohol, guaiacol, cresol, 2,6-dimethyl phenol, cathechol, vanillin, aceto vanillone, and syringaldehyde. The most abundant chemical components were phenols, followed by ketones and acetic acid (Kadir *et al.*, 2022). Desvita *et al.* (2021) stated that the primary compounds in wood vinegar from cacao pod shells (*Theobroma cacao* L.) are phenol and its derivatives, such as phenol; 2-methyl-p-cresol; phenol;

Table 2. Chemical compounds in wood vinegar made from a mixture of shells and fiber

No	Retention time (minute)	Wood vinegar compounds	Peak area (%)
1	2.288	Cyclohexylethylamine	0.67
2	2.542	1,2-Ethanediol	10.18
3	3.079	Formic acid	3.03
4	3.547	2-Propanone	1.63
5	3.875	Methyl ester	1.18
6	5.554	2,3-Butanedione	0.56
7	5.787	2-Butanone	0.27
8	7.740	Acetic acid	32.54
9	7.977	Ethanoic	13.94
10	9.434	Hydroxyacetone	1.64
11	11.394	3-Hydroxy-2-butanone	0.16
12	11.753	Propanoic acid	1.06
13	11.942	2-Propenoic acid	0.39
14	14.186	Propanoic acid	0.30
15	14.670	Cyclopentanone	0.44
16	15.954	Butanoic acid	0.47
17	17.271	2-Furancarboxaldehyde	10.14
18	18.819	2-Furanmethanol	0.59
19	19.189	1,2-Ethanediol etylene glycol	0.43
20	20.528	2-Cyclopenten-1-one, 2-methyl	0.56
21	20.938	Ethanone	0.42
22	23.342	Butyrolactone	0.43
23	23.525	Propanoic acid	0.75
24	23.769	5-Methyl	0.90
25	24.428	3-Metyl-2-cyclopenten-1-one	0.35
26	25.143	Butanoic acid	0.33
27	26.880	Benzenesulfonic acid	6.96
28	29.375	2-Methoxy phenol	2.54
29	10.508	4-Methyl phenol	1.17
30	30.842	2-Cyclopenten-1-one	0.37
31	31.058	6-Octenal	0.48
32	31.980	Pentanal	0.53
33	33.703	Phenol	1.25
34	35.337	Geraniol	0.38
35	36.441	2-Methyl-3-heptanol	0.30
36	37.083	2-Methoxy-4-methylphenol	0.46
37	37.918	Hydrazine	0.17
38	38.126	1,2-Benzenediol	0.53
39	40.272	2,6-Dimethoxyphenol	1.09
40	44.151	1,2,4-Trimethoxybenzene	0.42

3-methyl-; 1-propanol; 2-amino-1-octadecanamine; n-methyl-2-amino-1-propanol; and 4-ethyl-phenol; 4- ethyl. Recently, Aly *et al.* (2022) reported that the main components of wood vinegar from *Ficus benjamina* were syringol (48.98%); 4,5 dimethoxy-2-methyl phenol (4.16%); guaiacol-4-ethyl (3.05%); mequinol (2.61%); estragole (33.09%); benzene; 1,2,5-trimethoxy-3-methyl (2.65%); and butylated hydroxytoluene (1.69%). In addition, Oramahi *et al.* (2022) reported that the dominant components of vinegar made from mabang wood (*Shorea panchyphylla*) were 1,2-ethanediol, fluoromethane, formic acid, 2-propanone, acetic acid, acetol, furfural, 2,4-hexadecanoic acid, and guaiacol.

3.2. The total phenol and total acid content in the nipah fruit shells (NFS) and mixture of shells and fiber (MSF) wood vinegars

In this study, the performance of NFS and MSF exhibited several significant differences. Total phenol and total acid of NFS and MSF wood vinegar are shown in Table 3.

Total phenol and total acid contents in the NFS wood vinegar were 1.09% and 11.78%, respectively, while the total phenol and acid contents in the MSF wood vinegar were 1.84% and 9.18%, respectively. The total acid content of NFS was greater than that of MSF, resulting in higher antitermitic activity and lower loss of filter paper mass.

Table 3. Components of NFS and MSF wood vinegar

Wood vinegar source	Components	
	Phenol (%)*	Acid (%)*
NFS	1.09	11.78
MSF	1.84	9.18

* Measured in triplicate.
NFS: nipah fruit shells, MSF: mixture of shells and fiber.

The maximum total phenol and total acid contents of wood vinegar from oil palm empty bunches were 2.98% and 10.04%, respectively (Oramahi *et al.*, 2019). The abundant chemical components of vinegar are strongly influenced by the chemical nature of the biomass of origin, including cellulose, hemicellulose, lignin, and temperature of pyrolysis (Abnisa *et al.*, 2013; Demiral and Ayan, 2011). In this research, we focus only on two kinds of wood vinegar.

3.3. Anti-termite properties of nipah fruit shells (NFS) and and mixture of shells and fiber (MSF) wood vinegar

In this study, the daily termite mortality *C. curvignathus* treated with NFS and MSF wood vinegar was determined for 21 days using a no-choice feeding test; the results are presented in Table 4. Increasing concentrations of wood vinegar were associated with significantly increased mortality of *C. curvignathus* and decreased mass loss of the filter paper (Table 4); the highest termite mortality was at the highest concentration (12.5%) of wood vinegar. Statistically significant differences in filter paper consumption were observed for diluted wood vinegar between the control and treated samples. This trend is similar to Oramahi *et al.* (2020b) wherein *Coptotermes formosanus* died after 21 days of exposure.

Termicidal activity was consistent with the total concentration of acid in the wood vinegars (Table 1), which is in agreement with previous results. For example, Oramahi and Yoshimura (2013) examined wood vinegar from *Vitex pubescens* Vahl and found that it exhibited termicidal activity against *C. formosanus* and *Reticulitermes speratus.* The largest component of vinegar that contributed to high termicidal activity was total acid.

Overall, wood vinegar contributed significantly to termite mortality and the effectiveness of wood vinegar against *C. curvignathus* increased with the concentration

Table 4. Termicidal performance of two types of wood vinegar at different concentrations against *Coptotermes curvignathus* in a no-choice test with treated filter paper

Treatment		Termite mortality (%)	Mass loss after 21 days (%)
Source biomass	Concentration of treatment solution (%)		
NFS	0	19.19 ± 14.95[a]	33.40 ± 4.53[a]
	2.5	57.58 ± 33.75[abc]	26.90 ± 3.79[ab]
	5	53.54 ± 28.64[abc]	14.34 ± 5.43[cd]
	7.5	70.20 ± 33.28[abc]	15.32 ± 9.81[cd]
	10	88.89 ± 14.32[bc]	9.52 ± 8.04[cd]
	12.5	100 ± 0[c]	2.18 ± 0.31[d]
MSF	0	20.20 ± 16.69[a]	44.68 ± 3.83[a]
	2.5	30.30 ± 6.06[ab]	26.65 ± 2.53[bc]
	5	70.71 ± 34.99[abc]	22.86 ± 2.56[bc]
	7.5	74.24 ± 30.26[abc]	13.30 ± 1.41[cd]
	10	91.92 ± 7.00[bc]	17.40 ± 11.77[bcd]
	12.5	100 ± 0[c]	9.98 ± 5.10[cd]

Means (N = 4) ± SD of 33 termites per replicate.
[a–d] Means in the same column with the same letters are not significantly different (p < 0.05) according to Duncan's multiple range test.
NFS: nipah fruit shells, MSF: mixture of shells and fiber.

of the wood vinegar. Wood vinegar made from the shells of the nipah fruit lost more filter paper mass than wood vinegar made from a mixture of shells and nipah fruit coir fiber at a concentration of 12.5%. We hypothesize that the chemical compounds found in the nipah fruit coir fiber vinegar, including acetic acid, propanoic acid, benzenesulfonic acid, and octadecanoic acid, together with its characteristic phenol derivatives may cause this antitermitic activity. Similarly, Kadir *et al.* (2022) concluded that wood vinegar from *D. costulata* was effective against *C. curvignathus* Holmgren. Furthermore, Sunarta *et al.* (2011) stated that wood vinegars from oil palm fruit had the potential to be an environmentally low-impact wood preservative for preventing attacks by *Criptotermes* spp.

4. CONCLUSIONS

Wood vinegars made from the shells of the nipah fruit shells (NSF) and with mixture of shells and fiber (MSF) have anti-termite potential, especially NFS vinegar against *C. curvignathus*. The main compounds found in the NFS vinegar were cyclopropanecarbonyl chloride, 2,5-dichlorophenol, 2-propanone, acetic acid, propanoic acid, benzenesulfonic acid, 3,7-dimethyl- 6-octenal, and trans-geraniol. Meanwhile, in the MSF vinegar, the abundant compounds were 1,2-ethanediol, formic acid, acetic acid, ethanoic, 2-furancarboxaldehyde, phenol, 2-methoxy phenol, and 4-methyl phenol.

CONFLICT of INTEREST

No potential conflict of interest relevant to this article was reported.

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