Mahogany Wood-Waste Vinegar as Larvacide for *Spodoptera litura*

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The carbonization of lignocelullosic waste to obtain wood vinegar (WV) was investigated in this work. WV was used as a botanical insecticide against armyworm (*Spodoptera litura*), which is known as a major pest of soybean crops in Indonesia. This study includes the following: (i) the assessment of potential use of lignocellulosic waste from mahogany wooden-sandal home industry; (ii) the determination of the yield of various components of carbonization process, from each unit of the waste; and (iii) the application of the produced WV as larvicide on *S. litura* larvae in the laboratory. The experiments were arranged in a completely randomized design, and the observed variables included mortality and anti-feedant activity of *S. litura* larvae. The data were analyzed using analysis of variance with Duncan's multiple differences test. The results showed that the amount of wood waste generated at wooden-sandal craftsman level was 16.12%. Carbonization of 1,000 g of the wood waste yielded WV, tar, bio-oil, and char in quantities of 442.68 g, 36.5 g, 4.04 g, and 251 g, respectively. The treatment using WV concentration of 1.5% to 3.0% showed low larvacidal action, which gave LC_{50} value of 12.82%, but it had adequate anti-feedant activity.

Keywords: Wood vinegar; Lignocellulosic waste; Carbonization; Larvacidal; Antifeedant

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INTRODUCTION

Mahogany (*Swietenia mahagoni*) is the most widely used wood as a material for wooden sandals in Tasikmalaya City, Indonesia, because it has a good texture and there is a sufficient supply of logs in the surrounding area. Generally, the wood processing industry continually produces lignocellulosic waste. Incineration and landfilling practices to reduce lignocellulosic wastes from the furniture and handicraft industry is not appropriate to meet the environmental and sustainability requirements. Lignocellulosic waste incineration will increase CO_2 emissions, which will in turn contribute to global warming. Similarly, the landfilling of waste will involve a process of anaerobic decomposition that generates methane gas (CH_4), which will have a strong impact on global warming (Tiilikkala *et al*. 2010; Prodest 2012).

The waste biomass can be converted to produce useful materials by several methods. The thermochemical conversion methods include gasification, pyrolysis, and carbonization (Donate 2014). Biochemical conversion or processes include composting, making of silage, biomethanation, and bioethanol fermentation (Yokoyama 2008; Rahmat *et al*. 2014a).

Through the carbonization process, biomass can be converted to charcoal, wood vinegar (WV), tar, and bio-oil. WV utilization has been widely studied for pest control. Mahogany WV is expected to be effective as a larvicide, since previous reports on the use of other WV have demonstrated its effectiveness as a pesticide. Wititsiri (2011) revealed that WV possessed the most effective termiticidal activity against termite workers (*Odontotermes* sp.). A similar high pesticidal activity was also found among three wood vinegars against striped mealy bugs (*Ferrisia virgata*). The termiticidal and pesticidal properties of these wood vinegars can be attributed to the mode of action of their active components. Wagiman *et al*. (2014) reported that the application of WV from coconut shell at a concentration of 12% was recommended for the control of brown planthopper (*Nilaparvata lugens*) during rice cultivation.

In this study, the application of WV was evaluated in an effort to overcome the low production of soybean (*Glycine max*) in Indonesia, primarily caused by the presence of pests. One of the most important pests of soybean is *Spodoptera litura* Fabricius (Lepidoptera: Noctuidae), which is commonly called armyworm, cottonworm, or tobaccoworm. Farmers usually controlled this pest with the use of synthetic pesticides (Suprapto and Pujiharti 2012; Javar *et al*. 2013). However, the negative effects of these synthetic pesticides need to be considered. In particular, the employed pesticide should meet the requirements for sustainable agriculture and environmental safety (Marwoto and Suharsono 2008).

The aim of this study was to generate wood vinegar from waste mahogany shavings and to assess its larvicidal effect on *Spodoptera litura*.

EXPERIMENTAL

Materials

Preparation of mahogany wood-waste vinegar

Mahogany-wood chip waste was obtained from four different locations (2 kg from each) representing the home industries that prepare wooden-sandal handicrafts in the industrial center of the Cibeureum subdistrict, Tasikmalaya city, Indonesia. The waste was mixed homogeneously and then dried until it reached a humidity of 20%, while the feedstock for the pyrolysis was processed in a kiln.

Preparation of soybean leaves

Soybean (*Glycine max*) of the Grobogan variety was cultivated in the experimental station of the Agriculture Faculty, Siliwangi University, Tasikmalaya, Indonesia. The leaves of the crop were used as feed for the larvae of *S. litura*. The cultivation area of 6 m x 6m was screened with cages to avoid pest infestation.

Preparation of Spodoptera litura larvae

S. litura larvae collected from various host crops were reared on their respective host crops, which were maintained at Lembang Horticulture Research Station Bandung (Indonesia) during the study period (January-March 2015).

Furthermore, the pupae were reared following the procedure of Tukaram *et al.* (2014). The pupae were kept separately for moth emergence in a plastic jar of 12-cm diameter, wherein the pupae were placed on moist sand. To facilitate egg laying, fresh soybean leaves were kept inside the jar and a few drops of 10% honey solution were added as moth feed. Then, each jar was covered with muslin cloth. Eggs collected from each host crop were disinfected with 1% sodium hypochlorite solution to avoid any entomopathogenic contamination. The eggs collected from each jar were kept for incubation at 27 ± 1 °C and 70% to 80% relative humidity. The larvae culture were placed in air-conditioned room.

The newly hatched larvae, reared in plastic boxes of size 30 cm x 20 cm x 15 cm, were given fresh soybean leaves moistened with wet sponges to maintain leaf turgidity. The second and third instar larvae were taken and reared in a plastic basin measuring 25 cm in diameter, covered with muslin cloth.

Methods

Wood waste potential assesment

Many wood beams were processed within one week from four selected craftsmen, and the wastes were collected. In each of the craftsmen, the weight of sawdust, chips, and wood pieces were determined

Generation of WV and monitoring of parameters

The WV-generating process was carried out following the procedure of Rahmat *et al*. (2014b). Wood chip waste (1,000 g) with a water content of 20% was used as feedstock per unit of WV produced. The chips were heated to 450 °C for 45 min, in the absence of air or oxygen, in the airtight kiln, which was connected to a water-cooled condenser. The WV production parameters considered were the production rate of WV during the process and the final quantity of the products (WV, tar, bio-oil, and biochar). The crude distillate contained WV, bio-oil, and the remaining tar, all collected in the condenser outlet. This distillate was decanted for two weeks to form a separate factions. The biochar was weighed at the end of the process after the kiln was cooled. The experiment was repeated three times.

Experimental setup for testing the efficacy of WV

The experiment aimed to test the effectiveness of six concentration treatments of WV on the mortality and feeding intensity of the *S. litura*. The concentration of WV was varied as k_0 (0.0% as control), k_1 (1.0%), k_2 (1.5%), k_3 (2.0%), k_4 (2.5%), and k_5 (3.0%), respectively.

The experiment was applied in Bioassay Method using leaf dip technique. Fresh soybean leaves (2 g) were dipped in each concentration for 2 min, then wind-dried for 10 min. Futhermore, the dried leaves were placed in a plastic jar 12 cm in diameter and 8 cm in height and used to rear 10 third instar larvae of *S. litura*, respectively.

The experiment was arranged in a complete randomized design with four repetitions conducted. The data were analyzed using analysis of variance (ANOVA) and the Duncan's multiple range tests (Gomez and Gomez 1983).

Response variables were measured based on (i) the number of dead larvae, taken after seven days of incubation; and (ii) the average weight of residual leaves obtained after six days of incubation. The experiment was performed for seven days in the laboratory at 27 ± 1 °C.

Experimental parameters

(1) Larvicidal action

Larval mortality was recorded after seven days of treatment. The percent mortality (M) following Abbott in Baskar *et al*. (2011) was calculated:

$$M = \frac{\%\ \text{mortality in treatment} - \%\ \text{mortality in control}}{100 - \%\ \text{mortality in control}} \text{x} 100 \quad (1)$$

LC_{50} was determined by a series of concentrations of WV solution to the mortality of larvae of *S. litura* on the seventh day. Furthermore, the correlation of concentration and mortality was determined using a Probit Anaysis by Stat-RIV 2.0 software (Moekasan and Prabaningrum 2001).

(2) Antifeedant action

The larvae consumption of the treated leaf and control for seven days after incubation was recorded using an analytical balance. The antifeedant activity (A) was calculated:

$$A = \frac{Leaf\ weight\ before\ incubation - Leaf\ weight\ after\ incubation}{Leaf\ weight\ before\ incubation} \times 100\% \quad (2)$$

RESULTS AND DISCUSSION

Potential Quantity of Wood Waste

The quantity of mahogany wood-waste derived from each unit of prepared wood beams was processed to determine the quantity of lignocellulosic waste. Samples from the four craftsmen were acquired, and the data obtained are shown in Table 1.

Table 1. Quantities of Mahogany Wood-Waste from Wooden Sandal Industry

Sample of Crafstmen	Wood beams (kg)	Wood waste (kg)			Total
		Saw dust	Wood chips	Wood pieces	
1	167	8.45 (5.06%)	6.35 (3.80%)	12.02 (7.20%)	
2	376	20.31 (5.40%)	14.06 (3.74%)	27.45 (7.30%)	
3	286	13.73 (4.80%)	10.84 (3.79%)	19.45 (6.80%)	
4	460	24.84 (5.40%)	18.86 (4.10%)	32.66 (7.10%)	
Average	-	5.16 %	3.86 %	7.1 %	16.12%

There are several solutions for converting the huge amount of lignocellulosic waste into useful material. The method may be selected to convert the lignocellulose waste, including: (i) hydrolysis for ethanol and biogas production (Taherzadeh and Karimi 2007); (ii) pyrolysis for vinegar and/or biochar production (Tiilikkala et al. 2010); and (iii) wood board and block production (Purwanto 2011). Apparently, pyrolysis applications are simpler, because the plant is simpler and almost no pretreatment of the feedstock is required in the process. However, the selection of the application depends on the purpose of obtaining desired product.

Generation of WV

The measured rates included (i) volume of tar that was released from the outlet of heavy fraction and (ii) the volume of crude distillate measured in a 5-min period during the process (Fig. 1).

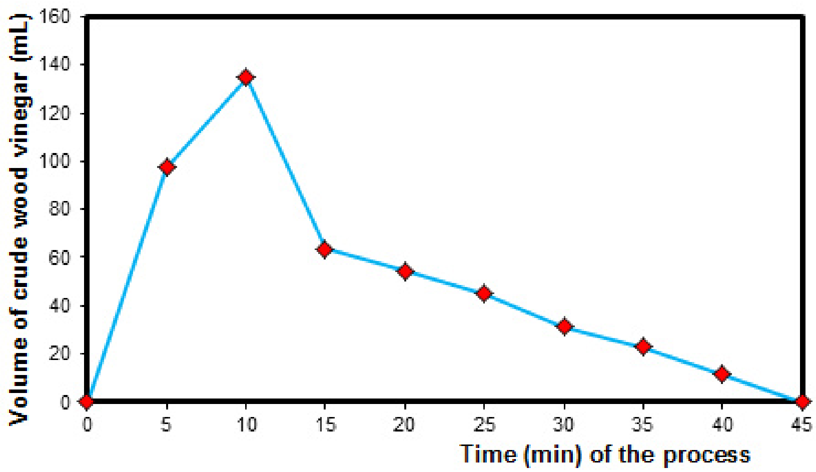

Fig. 1. Generation of crude wood vinegar with time

In the first phase, the condensate containing WV was produced and reached maximum production at the 10th min (132.67 mL). Then, the yield of WV decreased over time and reached almost zero at the 45th min. The increase of distillate yield in the initial period was due to the temperature of 100 °C, at which the water started to evaporate. The temperature of the kiln was gradually increased, which led to the degradation of organic compounds in the feedstock. As stated by Yokoyama (2009), the pyrolysis of wood is initialized with the degradation of hemicellulose at 200 to 260 °C, then cellulose at 240 to 350 °C, and lignin at 280 to 500 °C.

Bridgwater (2004) stated that the quantity of the products depends on the process. Therefore, to obtain the optimal results, the temperature should be maintained at the carbonation range (not exceeding 500 °C) and the time should be lengthened. Furthermore, the reported composition of the carbonization products was as follows: gas (35%), liquid (30%), and solid (35%).

If the temperature exceeds 500 °C or even reaches 800 °C, gasification will occur, which will yield gas (85%), liquid (tar, 5%), and solid (10%). Danarto *et al.* (2010) stated that pyrolysis can be defined as the thermal decomposition of organic material in its inert condition (without the presence of oxygen), which may induce the formation of volatile compounds. Generally, pyrolysis was started at 200 °C and kept at a temperature of 450 to 500 °C.

Quantities and Properties of Components

Carbonization of 1,000 g of wood chip waste produced WV, bio-oil, tar, and biochar with average quantities of 442.68 g, 4.04 g, 36.5 g, and 251 g, respectively (Table 2).

Table 2. Yield of Pyrolysis Products from 1,000 g of Mahogany-wood Waste

Batch	Yield of components			
	Biochar (g)	Wood vinegar (g)	Tar (g)	Bio-oil (g)
1	274	442.68	38	4.32
2	223	430.44	37	3.72
3	256	454.92	35	4.08
Average	251	442.68	36.5	4.04
%-wt	31.37	55.33	4.56	0.40

Wijaya *et al*. (2009) reported that the pyrolysis yields of WV at a temperature of 110 to 500 °C for 5 h of sawdust for teak wood, pine wood, and bamboo were respectively 55.20%, 58.33%, and 62.35%. Identification of the WV component of palm bunches showed that most of the phenol and acetic acid existed at all pyrolysis temperatures (350, 400, and 450 °C). The quantity of phenol and acid were increased with increasing temperature (Indrayani *et al*. 2011).

The amount of obtained bio-oil increased in the range of 300 to 350 °C and slightly decreased in the range of 350 to 500 °C. On the other hand, the charcoal yield decreased with increasing temperature. These results showed that bio-oil yield was inversely correlated with that of charcoal. In the range of 300 to 350 °C, the effect of thermal cracking led the forward reaction rate higher. Pyrolysis is an endothermic reaction that occurs at temperatures higher than 350 °C. The decrease in bio-oil yield was caused by the occurrence of continuous secondary thermal cracking. During secondary thermal cracking, solids such as charcoal were continuously pyrolyzed into non-condensed gas, leading to a reduction of bio-oil and charcoal yields and an increase in gas yield (Chaiya 2011).

The wood vinegar produced had the physical properties shown in Table 3.

Table 3. Physical Properties of Wood Vinegar from Mahogany-wood Waste

Parameter	Results
pH	3.4
Density (ρ)	1.020 g/mL
Color	Yellowish-brown

These physical properties of WV from mahogany wood were almost the same as those found by Yashimoto (1994). Hence, our first research priority was to test the effectiveness of WV in the agrochemical application.

Larvacidal Activity of Wood Vinegar

Based on observations during the seven days after incubation, the cumulative mortality between the treatment WV concentration k_3 (2.0%), k_4 (2.5%), and k_5 (3.0%) showed differences with the control (Table 4). However, the mortality rate was relatively low, as indicated by the LC50 value of 12.82%. This is due to the fact that the level of concentration of WV was still low when compared to previous studies. For instance, Chalermsan and Peerapan (2009) reported that WV concentrations of 5%, 10%, and 15% had a significant effect in reducing egg-laying and the number of damaged seeds by the cowpea beetle (*Collosobruchus maculatus*). Wagiman *et al*. (2014) showed that neutralized WV from coconut shell gave LC_{50} values for the mortality of brown planthopper (*Nilaparvata lugens*), after 24, 48, and 72 h, respectively, of 11.94%, 10.73%, and 9.94% for direct exposure and of 36.36%, 27.99%, and 26.15% for indirect exposure.

Table 4. Effect of WV on *S. litura* Larvae after Seven Days of Incubation

Treatments	Mortality (%)	Antifeedant activity (%)	Number of cannibalized larvae *
k_0 (0.0%)	00.0±0.00 a	13.72±0.76 a	1.00±0.00 a
k_1 (1.0%)	10.0±0.00 ab	18.34±1.28 ab	1.25±0.43 a
k_2 (1.5%)	15.0±8.66 bc	20.28±4.35 b	1.54±0.37 a
k_3 (2.0%)	17.5±8.29 c	20.10±4.33 b	1.74±0.46 a
k_4 (2.5%)	17.5±4.33 c	21.08±2.22 b	1.49±0.52 a
k_5 (3.0%)	17.5±4.33 c	21.30±1.63 b	1.31±0.54 a

Note: Numbers followed by the same letter indicate no significant differences according to the Duncan's multiple range test at a confidence level of 5%.
* No significant according to ANOVA and the data after transformed by $\sqrt{x+1}$.

In the treatment k_3 to k_5, the effectiveness of WV as an anti-feedant was greater than its larvicidal effect. Compared with controls, reduced food intake was observed in all treated leaves WV consumed by *S. litura*. The highest percent of anti-feedant activity was observed in the treatment k_5, followed by k_4, k_3, k_2, and k_1. Eating tendencies are reflected in this study assessed by the quantity reduction in the consumption of leaves by the larvae. Antifeedant activity of botanicals against insects has been studied in many countries. Arivoli and Tennyson (2012) reported that solvent residues of *Zanthoxylum limonella* leaf components obtained from solvent extracts dissolved in acetone were tested at 1,000 ppm continuously for 24, 48, and 72 h, on the third instar larvae of *S. litura*. The presence of an anti-feedant effect was inferred from the lower food consumption ingested by the larvae on castor leaves containing solvent residues of these botanicals. Larval mortality was also observed when the larvae were fed on treated castor leaves, implying death from either malnutrition or toxicity of this botanical.

This study also revealed a cannibalism phenomenon among *S. litura* larvae, which was seen among hungry larvae that did not find a suitable food source. This demonstrated that WV has an insect repellent effect. As reported by Rahmat *et al*. (2014b), teak WV could suppress the appetite of *Sitophilus zeamais*, thus reducing infestation during corn kernel storage.

Wood vinegar produced from the pyrolysis of various biomass types is similar in terms of its major chemical components. As presented Sunardi and Yuliansyah (2006) There are 24 chemical compounds found in wood vinegar of mangrove wood (*Rhizophora mucronata)*, mostly classified as alcoholic, phenol, ether, ketone, carboxyllate acid, ester, and benzene. Among these compounds are propanoic acid (2,89 %), 2-furancarboxaldehyde (9,86%), 3-furanmethanol (6,09 %), phenol (8,30%), allyl butyrate

(2,63%), phenol, 2-methyl-(2,73%), mequinol (9,91%), phenol,2,6-dimethoxy (19,49%), 1,2,4-trimethoxybenzene (5,55%), and benzene,1,2,3-trimethoxy-5-methyl (3,63%).

Haji (2013) reported the results of the composition analysis of oil palm (*Elaeis guineensis*) bunches WV consisting of 3-hydroxy butanoic acid (1.57%), acetic acid (16%), methyl propanoic (4.45%), propanoic acid (6.62), pirydine (1.62%), furfural alcohol (8.61%), gamma-butyrolactone (3.32%), phenol (3.56), dodecane (0.75%), 4-methyl phenol (20.80%), acid-9,12-hexacdecanoat (21.07%), acid 9,12-octa-decadienoat (8.84%), and acid-1,2-benzendi-carboxylate (2.90%).

Some previous research reports on the effectiveness of single wood vinegar as an insecticide have indicated significant effects when used alone, but when combined with other insecticides a synergistic effect was apparent. Kim *et al*. (2008) reported that wood vinegar itself did not show insecticidal activity on planthoppers (*Nilaparvata lugens* and *Laodelphax striatellus*).When the planthoppers were treated with wood vinegar mixed with carbosulfan, the mortality was greatly increased by the wood vinegar in comparison with a single carbosulfan treatment. This phenomenon is similar to that reported by Hashemi *et al*. (2014) that wood vinegar itself did not show insecticidal activity on mortality of cigarette beetle (*Lasioderma serricorne)*. When the insect was treated with wood vinegar mixed with *Salvia leriifolia* extracts, the mortality induced by methanol extracts was greatly increased by the wood vinegar in comparison with a single methanol extracts treatment. Wood vinegar showed antagonistic effect on aqueous and ethanol extracts. These results suggest that wood vinegar has a synergistic effect on the insecticidal activity of methanol extracts

The efficiency of wood vinegar mixed with each individual of three plants extract such as citronella grass (*Cymbopogon nardus*), neem seed (*Azadirachta indica*), and yam bean seed (*Pachyrhizus erosus*) were tested against the second instar larvae of housefly (*Musca domestica*). The results of the study indicated that treatment of wood vinegar mixed with citronella grass showed the highest larval mortality by topical application method (50.0%) and by feeding method (80.0%) (Pangnakorn and Kanlaya 2014).

CONCLUSIONS

1. The wood waste generated by wooden-sandal craftsman was 16.12%.
2. The carbonization process for 45 min on 1,000 g of wood sandal waste yielded wood vinegar (WV), bio-oil, tar, and biochar in quantities of 442.68 g, 36.5 g, 4.04 g, and 251 g, respectively.
3. The mahogany wood-waste vinegar concentration treatment of 1.5% to 3.0% showed low larvacidal action, which gave LC_{50} value of 12.82%, but it had adequate anti-feedant activity.

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