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Research Article

Wood vinegar: by-product from rural charcoal kiln and its role in plant protection

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Abstract

Students from the RMUTL Pitsanuloke campus developed a charcoal kiln in order to produce charcoal from small fallen branches of nearby trees. During the process of making charcoal, the kiln was designed to produce wood vinegar as a by-product. Use of wood vinegar is becoming popular with some Thai farmers who make use of this cheap source of organic liquid as a plant protection substrate. The experiment examined wood vinegar, produced by the student-designed kiln, as a fungicide, bactericide and as either laying or hatching inhibitor of cowpea weevil. The results suggest that wood vinegar is a promising solution in plant protection, showing good potential for inhibition of pathogenic-fungi and bacterial growth. It was found that cleaning wood vinegar by sedimentation or centrifugation is necessary. Sedimentation of wood vinegar for 3 months resulted in similar characteristics to commercial products. Furthermore, centrifuged wood vinegar could prevent egg laying and penetrated well into the seed of cowpea weevil. These results indicated that people in rural communities could easily build or modify their own charcoal kilns in order to produce wood vinegar for use as an organic method of plant protection for their farms.

Keywords: pathogenic-fungi growth inhibitor, bacterial growth inhibitor, laying inhibitor, hatching inhibitor**,** organic fungicide pesticide, Thailand

Introduction

In rural of Thailand, people usually make charcoal from branches or pieces of wood using an earthen kiln. The branches are often dead wood blown down during storms or sometimes bamboo is used. The charcoal is then used as fuel for cooking or is sold. Conventional kilns produce potentially harmful smoke and pollution into the village. Recently, newer styles of kiln have been introduced to the public however, there is little or no supporting documents and information. A number of these kilns are able to made good quality charcoal and are suitable for adaption to the collection of wood vinegar during was it is essentially a gasification process. Wood vinegar, or pyroligneous acid, is a by-product from the charcoal production process [1]. It is a red-brown liquid generated from the gas and combustion of fresh wood burning under airless conditions. When the gas is cooled, it condenses into liquid, which contains acetic acid, methanol, acetone, wood oils and tars. Acetic acid, which is the largest component of wood vinegar, has exhibited high termiticidal activity [2]. The antifungal efficiency of the wood vinegar is strongly dependent upon their phenolic compound contents and this has been confirmed through the inhibitory growth of the major fungi [3]. Also, the addition of the crude wood vinegar as a medium component into sawdust mulch or compost in the concentration range of 0.1.6% increased yields in fruit-bodies by 21–42% [4]. Nakajima *et. al.* [5] reported that wood vinegar concentration less than one percent would activate plant growth and spraying a dilution of wood vinegar on the ground also reduced pathogenic microorganisms and soil arthropods [5]. Most of the research on wood vinegar has been undertaken in China and Japan, while some work has also focused on using a compound of wood vinegar in feed to improve animal health. This research work undertakes to tested and prove the plant protection properties of wood vinegar, making it a viable and safer alternative to commonly used agrochemicals in order to decrease farming costs. Wood vinegar is thus particularly suitable for farmers to apply in organic farming systems.

Research Methodology

To evaluate the role and effectiveness of wood vinegar in plant protection, research was conducted in three major experiments as follows:

Fungicidal effectiveness test

The effectiveness of wood vinegar as a fungicide was tested on six major plant disease pathogens, namely; *Rhizoctonia solani*, *Sclerotium oryzae*, *Helminthosporium maydis*, *Pythium* sp., *Colletotrichum gloeosporioides* and *Choanephora cucurbitarum*. Completely randomized design with 4 treatments and 4 replications were employed during the experiment. The treatments were a solution of wood vinegar at 0, 2, 3 and 4%, mixed in 100 ml of PDA and poured onto a media plate. The PDA was then left to cool down and thereafter each fungal pathogen colony was transferred by cork borer number three onto the PDA which was mixed with wood vinegar at various strengths. All of the plates were placed into an incubator at room temperature to allow the fungi colonies to freely grow. The diameters of each colony were measured and percentage inhibition of each colony by the wood vinegar solution was calculated using the following formulation:

$$\text{inhibition percentage} = \frac{R1 - R2}{R1} \times 100$$

where; *R1 = colony diameter of control batch*
R2 = colony diameter of treated batch

Bactericidal effectiveness test

The effectiveness of wood vinegar as a bactericidal was tested on two major bacterial pathogens: *Xanthomonas campestris* pv. *Citri* and *Erwinia carotovora* pv. *Carotovora*. Completely randomized design with 4 treatments and 4 replications were employed during the experiment. The treatments were a solution of wood vinegar 0, 2, 3 and 4%. NA was prepared by mixing with each pathogenic bacteria. Wattman’s paper was then soaked in each particular solution of wood vinegar, following which these pieces of paper were placed onto prepared NA plates. All NA plates were incubated at room temperature for 48 hrs. Clear zones were then measured and calculated.

Laying and piercing inhibitor test

Twenty plates with 100 cowpea seeds each were set by 4 treatments with 5 replications. The treatments were spraying wood vinegar solution of 0, 5, 10 and 15% separately. Five pairs of weevils were then put on the seed plates. Laying eggs by female weevils were daily recorded. On the 7th day, the number of pierced seeds was recorded. The data were then statistically analyzed.

Results and Discussion

Fungicidal effectiveness test

The colonies of *Pythium sp*., *Sclerotium oryzae* and *Rhizoctonia solani* were unable to grow on the PDA mixed with wood vinegar 2, 3 and 4%. However, *Choanephora cucurbitarum*, *C. gloeosporioides* and *Helminthosporium maydis* were able to grow on PDA mixed with wood vinegar at 2%. The means of colony diameter were 1.69, 1.83, 3.64 cm respectively. Notably, hypha only grew upwards but not sideways. However, no colonial fungi were able to grow on the PDA mixed with wood vinegar at 3 and 4% (see Table 1).

Table 1. Means of six colonial diameters (cm) of tested pathogenic fungi on PDA mixed with wood vinegar.

Tested pathogenic fungi	Concentrations (%)			
	0	2	3	4
Rhizoctonia solani	9.00	0.00	0.00	0.00
Sclerotium oryzae	9.00	0.00	0.00	0.00
Helminthosporium maydis	9.00	3.64	0.00	0.00
Pythium sp.	9.00	0.00	0.00	0.00
Colletotrichum gloeosporioides	9.00	1.83	0.00	0.00
Choanephora cucurbitarum	9.00	1.69	0.00	0.00

The inhibition percentages of the tested pathogenic fungi by wood vinegar were statistically different. The concentration of wood vinegar at 3 and 4% in PDA completely inhibited all tested pathogenic fungi. The PDA mixed with 2% wood vinegar inhibited *Pythium* sp. and *Rhizoctonia solani* growth as shown in Table 2. However, these concentrations obtained 81.25, 79.72 and 59.58% inhibition respectively, but there was no inhibition on PDA mixed with 0% wood vinegar (see Table 3).

Table 2. Means of percentage inhibition by 4 concentrations of wood vinegar mixed in PDA.

Concentration (%)	Pathogenic fungi		
	R. solani	*S. oryzae*	*Pythium sp.*
0	0.005 b	0.004 b	0.003 b
2	100.00 a	100.00 a	100.00 a
3	100.00 a	100.00 a	100.00 a
4	100.00 a	100.00 a	100.00 a
CV (%)	0.003	0.003	0.003

Note: means with the same alphabet in column were no statistically different by DMRT at 0.05% of confidence

Table 3. Means of percentage inhibition on *Helminthosporium maydis*, *Choanephora cucurbitarum* and *Colletotrichum gloeosporioides* by 4 concentrations of wood vinegar mixed in PDA.

Concentration (%)	Pathogenic fungi		
	H. maydis	*C. gloeosporioides*	*C. cucurbitarum*
0	0.00 c	0.00 c	0.00 c
2	59.58 b	79.72 b	81.25 b
3	100.00 a	100.00 a	100.00 a
4	100.00 a	100.00 a	100.00 a
CV (%)	0.88	1.09	1.17

Note: means with the same alphabet in column were no statistically different by DMRT at 0.05% of confidence

Bactericidal effectiveness test

The results obtained were statistically different among treatments on *Xanthomonas campestris* pv. *Citri* and *Erwinia carotovora* pv. *Carotovora*. The piece of paper placed in wood vinegar solution at 4% resulted in clear zones with diameters 1.40 and 1.18 cm respectively. At the concentrations of 3 and 2%, the clear zones of *X. campestris* pv. *Citri* were 1.35 and 1.01 cm respectively (Fig. 1). Similarly, the clear zones of *E. carativora* pv. *Carotova* were 1.17 and 1.13 cm respectively (Fig. 2). On the other hand, there was no clear zone on the NA without wood vinegar.

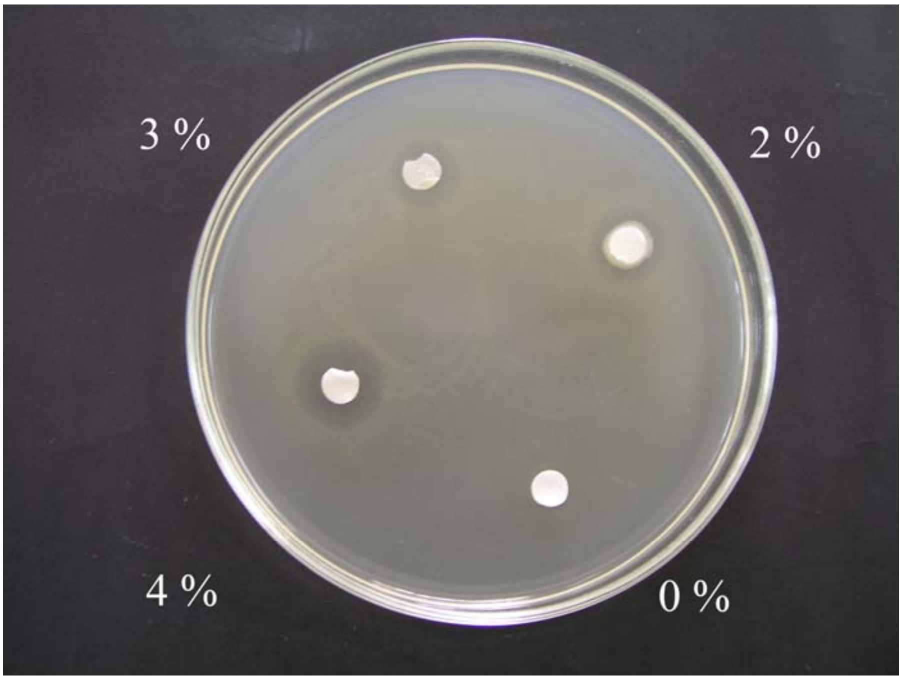

Figure 1. Clear zone on NA culture of *Xanthomonas campestris* pv. *citri* affected by wood vinegar at 0, 2, 3 and 4%.

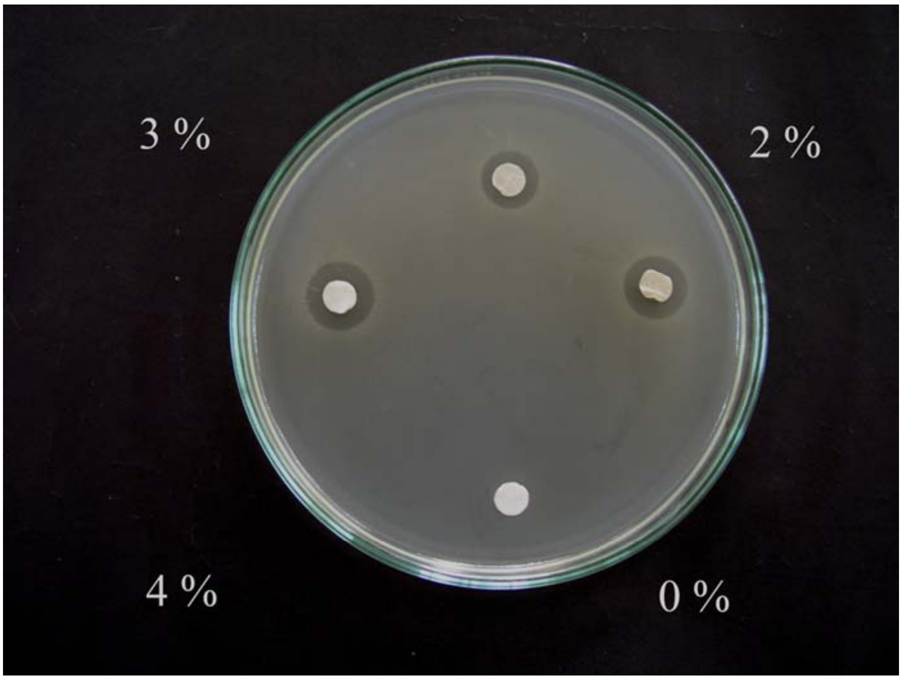

Figure 2. Clear zone on NA culture of *Erwinia carotovora pv. carotivora* affected by wood vinegar at 0, 2, 3 and 4%.

Wood vinegar solution has the ability to inhibit pathogenic fungi and bacteria due to its chemical properties such as acidity and other various chemical components. Acetic acid, formaldehyde and methanol in wood vinegar have been reported to have a lethal effect on fungi, bacteria and virus particles [6]. This research showed that wood vinegar has fungicidal effects. Similarly, Numata *et al*. [7, 8], reported that wood vinegar inhibiedt some pathogenic fungi such as *Pythium sp. Rhizoctonia solani, Penicillium spp., Sclerotium homoeocarpa, Fusarium oxysporum, f. sp. lycopersici, Phytophthora capsici, Pythium aphanidermatum*, by the addition of 10% wood vinegar onto PDA. Inhibition occurred on the fifth day after inoculation.

Laying and hatching inhibitor test

Inhibition on laying and piercing of cowpea weevil (*Collosobruchus maculates* Frabricius) was compared between raw, centrifuged and commercial wood vinegar. The results showed that centrifuged wood vinegar obtained the highest laying inhibition of cowpea weevils ($p<0.05$). By which, means of laying by cowpea weevils was 21.40%, whereas, seed sprayed with raw and commercial wood vinegar showed no statistical difference ($p>0.05$) of laying eggs on seeds at 38.00 and 30.80% respectively (see Table 4).

Table 4. Means of laying and piercing of cowpea weevil (*Collosobruchus maculates* Frabricius) on seed sprayed with different wood vinegars.

Treatments	Laying (%)	Piercing (%)
raw-wood vinegar	38.00 b	12.60 bc
centrifuged-wood vinegar	21.40 c	7.80 c
Commercial-wood vinegar	30.60 b	23.20 b
water	90.40 a	82.40 a

Note: means with the same alphabet in column were no statistically different by DMRT at 0.05% of confidence

In comparing the inhibition effect of wood vinegar on piercing of seed by cowpea weevil, the results were similar to laying inhibition. Those seeds sprayed with centrifuged wood vinegar showed the highest piercing inhibition ($p<0.05$). Only 7.08% of these seeds were pierced (see Table 4), followed by seed sprayed with raw wood vinegar (12.60%). On the other hand, seed sprayed with the commercial wood vinegar and with water had a mean of pierced seeds by weevils of 23.20 and 82.40% respectively (Table 4).

According to the results, centrifuged wood vinegar has the best potential to inhibit laying and piercing on seeds of cowpea by weevils compared to other forms of wood vinegar examined in this work. Centrifuged wood vinegar had the most fragrant smoke among the tested wood vinegars. It has been reported that wood vinegar also shows strong acid activity at pH3 and contains 280 different components, the major ones being acetic and propionic acid [9] and antioxidant substances such as phenolic compounds [10]. Thus wood vinegar is also regarded as containing natural organic acids [11].

In addition, centrifugation will reduce sediments in wood vinegar. Similarly, Wang and Liu [12] reported that wood vinegar affected laying eggs of red mites. Thus, wood vinegar produced by people in rural areas is a promising solution to protect seeds of their agricultural produce during storage instead of using commercial or chemical means. People could produce wood vinegar by themselves and increase their potential by rigorous stirring instead of sedimentation for 3-4 months.

Conclusion

Wood vinegar solutions inhibit growth of some pathogenic fungi on PDA such as *Rhizoctonia solani*, *Sclerotium oryzae*, *Helminthosporium mayis*, *Pythium* sp., *Colletotrichum gloeosporioides* and *Choanephora cucurbitarum.* In addition, bacterial causes of plant pathogens; *Xanthomonas campestris* pv. *Citri* and *Erwinia carotovora* pv. *Carotovora* colonies could be inhibited on NA. The suitable concentration is less than 10%, which also prevents leaf burning by wood vinegar.

Wood vinegar has potential postharvest application on laying and piercing of seed by cowpea weevil. In particular, centrifuged and raw wood vinegar that can readily be produced by rural people shows high potential as an alternative means in sustainable agriculture in terms of plant protection.

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