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Research Article
Antibacterial Effectiveness of Cinnamon Wood (*Cinnamomum burmannii* BL) Liquid Smoke Obtained from Different Pyrolysis Time

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Abstract

Background and Objective: Cinnamon is one of the plantation crops found in West Sumatra. The main product of cinnamon is the dry bark, while the use of the wood is still limited to firewood. This study was designed to discover other alternatives of cinnamon wood as raw material for pyrolysis. This study aimed to determine the effectiveness of the liquid smoke from various levels of pyrolysis time against *Bacillus* sp. and *Staphylococcus aureus*. **Materials and Methods:** The liquid smoke as pyrolysis result was taken every 30 min, during the all pyrolysis period of 240 min. The effectiveness of liquid smoke against bacteria was observed by the antibacterial activity of the Inhibition Zone (IZ), the Minimum Inhibitory Concentration (MIC) and the Minimum Bactericidal Concentration (MBC). **Results:** The results showed that the liquid smoke from the cinnamon wood was a "very strong" anti-bacterial. The best pyrolysis time of cinnamon wood was 240 min as the most effective time. The IZ of cinnamon liquid smoke with pyrolysis time of 240 min against *Bacillus* sp. was 31.7 mm, while the IZ of *S. aureus* was 39.7 mm. The MIC was 1.56% for *Bacillus* sp. and 0.39% for *S. aureus*. MBC was 3.13% for *Bacillus* sp. and 0.78% for *S. aureus*. **Conclusion:** The longer pyrolysis time, the stronger the antibacterial properties. Pyrolysis time for 240 min produced the most effective liquid smoke in inhibiting the growth of *Bacillus* sp. and *S. aureus*. The result of IZ, MIC and MBC concluded that *Bacillus* sp. was more resistant than *S. aureus* to liquid smoke of cinnamon.

Key words: Liquid smoke, dried cinnamon wood, pyrolysis, antibacterial, MIC, MBC

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Liquid smoke is the result of the steam condensation process from direct or indirect combustion, using materials containing lots of lignin, cellulose, hemicellulose and hydrocarbon compounds[1]. Smoke is induced by the thermal degradation or pyrolysis of wood[2]. Liquid smoke can be utilized for its antimicrobial properties against a variety of Gram-positive and Gram-negative bacteria, yeast and molds[3]. Guillen *et al.*[4] found the major proportion of commercial full strength liquid smoke to be composed of water (11-92%), tar (1-17%), acids (2.8-9.5%), carbonyl containing compounds (2.6-4.6%) and phenol derivatives (0.2-2.9%). The content of liquid smoke includes A group of acidic compounds affecting the taste, pH and product shelf-life, A group of carbonyl compounds, reacting with proteins and create colour and Phenols, the main constituents of aroma and has an antioxidant activity[5]. The phenolic content of liquid smoke has antibacterial and anti-fungal properties against several pathogenic bacteria and carcinogenic bacteria[6]. A previous study found that redistilled liquid smoke from oil-palm shells has been proven effective as a fresh fish preservative and as fly repellent in the fish salting-fermenting process due to its antibacterial activity[7]. The addition of coconut shells liquid smoke also extended the shelf life of tandipang (*Dussumieria acuta*) smoked fish[8]. Liquid smoke from walnut can inhibit the activity of *Staphylococcus aureus*, *Escherichia coli*, *Bacterium proteus*, *Bacterium prodigious* and *Aerobacter aerogene*[9].

The content of the lignocellulosic material, the pyrolysis of the wood become liquid smoke is an alternative to increase its economic value. In the pyrolysis process, cellulose and hemicellulose of wood are converted into a group of acidic compounds, while lignin is changed into a group of phenolic compounds[10]. Acid and phenol compounds from liquid smoke have a bacteriostatic effect[11]. The acidic compounds acidify the cytoplasm and damage the membrane permeability of bacteria[12]. While phenolic compound causes the defect of the cell wall by denaturing proteins and dissolving lipids so that the bacterial cell wall disintegrates and disrupts the permeability of cell membranes[13]. Acetic acid has a stronger inhibitory capacity against bacterial growth compared to phenolic compounds. However, when the acidic compound is converted in the form of liquid smoke, a greater inhibitory ability will be produced.

A study about the effectiveness of liquid smoke from cinnamon wood as an antibacterial has not existed yet. This study aimed to determine the effectiveness of the liquid smoke from various levels of pyrolysis time against *Bacillus* sp. and *Staphylococcus aureus*. The IZ, MIC and MBC were several indicators to determine the effectiveness of liquid smoke as an antibacterial and to discover the exact concentration of liquid smoke in inhibiting and eradicating *Bacillus* sp. and *S. aureus*.

MATERIALS AND METHODS

Study area: Pyrolyzer was self-made equipment and assembled in accordance to study requirement. This simple pyrolyzer was able to condense steam of combustion result from wood cinnamon and finally, it was collected to become liquid. The liquid smoke was then analyzed to discover its effectiveness as an antibacterial. *Bacillus* sp. and *S. aureus* were derived from the bacterial collection of Microbiology Laboratory and Agricultural Biotechnology, Faculty of Agricultural Technology andalas University. This study was conducted from January-March, 2020.

The bacteria have been propagated then in the laboratory and treated to liquid smoke based on different time of pyrolysis. The formed inhibition zone of liquid zone treatment determined the ability as antibacterial. Inhibition zone from the antibacterial activity was measured from diameter which was formed in millimeter (mm). The MIC was measured from the turbidity of Nutrient Broth (NB) media and then compared to Standard Solution Turbidity (McFarland Solution). Whereas, MBC was measured from the number of bacteria colony grown in media.

Plant material: *Cinnamon burmanii* BL woods were collected from Lubuk Sikaping, Pasaman Regency, West Sumatra Province. The woods were cut into small size to ease the transportation. Woods were dried for 7 days in Technopark Laboratory, Faculty of Agricultural Technology andalas University.

Extraction of liquid smoke from cinnamon wood: Dried woods were put 2/3 of the pyrolyzer height, heated and passed the boiling point of organic compound in dried woods, so that the organic compounds changed to the gas phase[14]. The gas is dried woods flowed up to the top of the pyrolyzer and went through the connected pipe and finally flowed into condenser for condensation process. Liquid smoke that had changed from solid to liquid exited out through the condenser and ended into a storage tube. Charcoal as the residue of combustion was left in the pyrolyzer. Besides, other materials could be condensed or missing materials and tar. The testing of dried woods was carried out twice.

The pyrolysis time was observed at 30, 60, 90, 120, 150, 180, 210 and 240 min. A record of pyrolysis time began from the first time the liquid smoke dripped. Liquid smoke obtained in 30 min of pyrolysis (AC1) was combined with liquid smoke at 60 min of pyrolysis, coded as an AC2 sample. AC2 was combined with liquid smoke at 90 min of pyrolysis (AC3). The AC3 was combined with liquid smoke at 120 min of pyrolysis (AC4). The AC4 was combined with liquid smoke at 150 min of pyrolysis (AC5). The AC5 was combined with liquid smoke at 180 min of pyrolysis (AC6). The AC6 was combined with liquid smoke at 210 min of pyrolysis (AC7). The AC7 was combined with liquid smoke at 240 min of pyrolysis (AC KM). All liquid smoke was then decanted for a week in a glass container and tightly closed. After the decantation process, the condensate was filtered using filter paper to separate liquid smoke from tar. The liquid smoke was tested for its effectiveness against *Bacillus* and *Staphylococcus* by observing the Inhibition Zone (IZ), the Minimum Inhibitory Concentration (MIC) and the Minimum Bactericidal Concentration (MBC).

Preparation of bacterial culture medium, bacterial suspension and McFarland solution: Two grams of Nutrient Agar (NA) was weighed and dissolved with 100 mL of distilled water in Erlenmeyer, homogenized and boiled. A total of 5 mL of boiled NA was poured into three test tubes and sterilized. Almost 30 min after sterilization, the agar slant was ready to use for bacterial culture. A colony of bacteria was picked with a sterile loop wire and then suspended into a tube containing 2 mL of 0.9% NaCl solution, to obtain similar turbidity as McFarland solution.

Turbidity Standard Solution comprised of H_2SO_4 solution 0.36 N 99.5 mL was combined and completely mixed with 0.5 mL $BaCl_2.2\ H_2O$, 1.175% solution to form a turbid suspension. The density of the suspension of bacterial cells is compared to the McFarland turbidity standard. The culture medium was prepared by pouring 75 mL of NA and adding the bacterial suspension, then left it until solid. Wells were made on medium using a sterile pipette according to the number of samples and used for the antibacterial test.

Zone of inhibition test: Liquid smoke with various pyrolysis time (AC1, AC2, AC3, AC4, AC5, AC6, AC7 and ACKM), distilled water solution as negative control (K^-), Amoxicillin solution as a positive control (K+) was pipetted 0.2 mL on a cotton swab and put into the well. Those were incubated at 37°C for 24 hrs. The observation was performed after 24 hrs of the incubation period. The clear area was an indicator of bacterial sensitivity against antibacterial substance and showed by the width of IZ diameter[15]. The diameter of IZ was measured using a caliper (clear area diameter-well diameter) in millimeters (mm) and categorized based on Davis and Stout's classification[16].

MIC determination: The MIC assay was determined using the dilution method (five tubes as treatment, one tube as positive control, one tube as negative control and one tube as a controlled media)[17]. Each five treatment tubes were filled with 5 mL of NB (Nutrient Broth). Tube No. 1 was added with 5 mL of liquid smoke, then homogenized. The mixture of liquid smoke and NB in tube No. 1 was pipetted 5 mL and transferred to the second tube and homogenized, the dilution was continued five times. Afterwards, 0.1 mL of the mixture from each tube (tubes 1-5) was pipetted, discarded and added 0.1 mL of bacterial suspension (1×10^8 CFU mL^{-1}). The total volume of each tube was 5 mL.

The sixth tube for negative control was filled with 4.9 mL NB added with 0.1 mL bacterial suspension 10^8 CFU mL^{-1} and homogenized. Positive control (Tube No. 7) was filled with 5 mL NB, added with 5 mL amoxicillin 1% and homogenized. As much as 0.1 mL of the stock solution was discarded then added with 0.1 mL of bacterial suspension 108 CFU mL^{-1} and homogenized again. Tube No. 8 was a control medium, containing 1 mL NB. All tube (No. 1-8) was incubated in a water bath shaker at 40°C for 24 hrs and then observed the turbidity in culture media and compared with controls. The first lowest concentration showing clarity of medium was established as the MIC.

MBC determination: The bacterial suspensions resulted in MIC assay, were poured on NA, spread and incubated at 37°C for 24 hrs. The determination of the MBC value was carried out by calculating bacterial colonies growing on NA.

RESULTS AND DISCUSSION

Antibacterial activity: The result of the measurement of the clear zone which was formed from cinnamon liquid smoke against the growth of *Bacillus* sp. and *S. aureus* were shown in Table 1.

The diameter of IZ was larger in *S. aureus* (30.3-39.7 mm) than in *Bacillus* sp. (26.5-31.7 mm). The result showed that liquid smoke has been able to inhibit the activity of *Bacillus* sp. and *S. aureus* with pyrolysis time since 30 min of pyrolysis time. Several different pyrolysis time of cinnamon wood produced liquid smoke with antibacterial activity and included in the "very strong" group. The classification of antibacterial

Table 1: Antibacterial activity of cinnamon liquid smoke against *Bacillus* sp. and *S. aureus*

Treatment	IZ of *Bacillus* sp. (ø, mm)	Increased activity (%)	IZ of *S. aureus* (ø, mm)	Increased activity (%)	Description
AC1	26.5	0	30.3	0	Very strong
AC2	26.6	0.30	31.5	3.96	Very strong
AC3	27.5	3.38	32.9	4.44	Very strong
AC4	27.5	0	33.7	2.43	Very strong
AC5	28.5	3.64	34.3	1.78	Very strong
AC6	28.7	0.70	35.3	2.92	Very strong
AC7	29	1.04	35.4	0.28	Very strong
ACKM	31.7	9.31	39.7	12.15	Very strong
K+	13.6	18	40.2	1.26	Very strong
K^-	0	0	0	0	No activity

AC1: Liquid smoke (ACP) 1-30 min, AC2: 0-60 min, AC3: 0-90 min, AC4: 0-120 min, AC5: 0-150 min, AC6: 0-180 min, AC7: 0-210 min and ACKM: 0-240 min

power is based on IZ criteria, "weak" (ø = ≤5 mm), "moderate" (ø = 5-10 mm), "strong" (ø =10-20 mm), "very strong" (ø = ≥20 mm).

Inhibition Zone (IZ) of antibacterial activity resulted in cinnamon liquid smoke on NA was presented in Fig. 1. Eight paper disks with different concentration of cinnamon liquid smoke (AC1-ACKM) and two disks as control were transferred onto NA with *Bacillus* sp. and *S. aureus*. IZ was formed after paper disks were incubated overnight and antibacterial activity of liquid smoke was assessed by measuring the clear zone on the surface of NA.

All treatment was represented in four petridish in Fig. 1(a-b) showing treatments of eight types of cinnamon liquid smoke with different pyrolysis time (AC1 = 30 min, AC2 = 60 min, AC3 = 90 min, AC4 = 120 min, AC5 = 150 min, AC6 = 180 min, AC7 = 210 min and ACKM = 240 min of pyrolysis time), negative control (K^-) and positive control (K+)) against the growth of *Bacillus* sp. (BP). The largest zone of inhibition against *Bacillus* sp. was liquid smoke with pyrolysis time for 240 min of 31.7 mm (ACKM), followed by AC7 = 29 mm, AC6 = 28.7 mm, AC5 = 28.5 mm, AC4 = 27.5, AC3 = 27.5 mm, AC2 = 26.6 mm and the smallest IZ was AC1 = 26.5 mm.

Figure 1c and d were treatments of 8 types of cinnamon liquid smoke with different pyrolysis time (AC1 = 30 min, AC2 = 60 min, AC3 = 90 min, AC4 = 120 min, AC5 = 150 min, AC6 = 180 min, AC7 = 210 min, ACKM = 240 min of pyrolysis time), negative control (K^-), and positive control (K+)) against the growth of *S. aureus* (SA). The largest zone of inhibition against *Bacillus* sp. was liquid smoke with pyrolysis time for 240 min of 39.7 mm (ACKM), followed by AC7 = 35.4 mm, AC6 = 35.3 mm, AC5 = 34.3 mm, AC4 = 33.7, AC3 = 32.9 mm, AC2 = 31.5 mm and the smallest IZ was AC1 = 30.3 mm.

Cinnamon liquid smoke showed antibacterial activity (IZ) against *Bacillus* sp. and *S. aureus* but the effect of liquid smoke was more apparent to inhibit the growth of *S. aureus*, where IZ of *S. aureus* was larger than IZ of *Bacillus* sp. In this study, the result showed that the longer pyrolysis time, the bigger effect of antibacterial activity against *Bacillus* sp. and *S. aureus*.

Each paper disk contained one concentration of liquid smoke, AC1 = 30 min, AC2 = 60 min, AC3 = 90 min, AC4 = 120 min, AC5 = 150 min, AC6 = 180 min, AC7 = 210 min, ACKM = 240 min of pyrolysis time), as a negative control (K^-) and as positive control (amoxicilline) (K+).

The ability of liquid smoke was evaluated against *Bacillus* sp. (BC) and *S. aureus* (SA). The clear area was measured by caliper from the paper disk to the edge of the clear zone (IZ). The longer the pyrolysis time, the larger zone of inhibition against *Bacillus* sp. (a-b) and *S. aureus* (c-d). Though, the cinnamon liquid smoke was able to inhibit *S. aureus* growth better than the *Bacillus* sp.

Based on the length of pyrolysis time (30, 60, 90, 120, 150, 180, 210 and 240 min), the largest diameter of IZ was formed by liquid smoke concentration at 240 min of pyrolysis time (31.7 and 39.7 mm for *Bacillus* sp. and *S. aureus*, respectively), whereas the smallest diameter of IZ was formed from the concentration of liquid smoke at 30 min of pyrolysis time (26.5 and 30.3 mm for *Bacillus* sp. and *S. aureus*, respectively). The diameter of IZ for each concentration of cinnamon liquid smoke in *S. aureus* (30.0-39.7 mm) was larger than IZ in *Bacillus* sp. (26.5-31.7 mm). The inhibited capacity of cinnamon liquid smoke was more effective to prevent *S. aureus* growth than in *Bacillus* sp. The longer the pyrolysis time of cinnamon liquid smoke, the larger IZ formed, both in *Bacillus* sp. and *S. aureus* in Fig. 2.

Based on the various time of pyrolysis, the ability of liquid smoke as an antibacterial was also different. The longer the pyrolysis time, the larger IZ generated (Fig. 1), both in *Bacillus* sp. and *S. aureus*. This indicated that the pyrolysis time affected the resulting liquid smoke component. The longer pyrolysis time, chemical compounds derived from liquid smoke were produced more effective in inhibiting the growth of *Bacillus* sp. and *S. aureus* (Fig. 2).

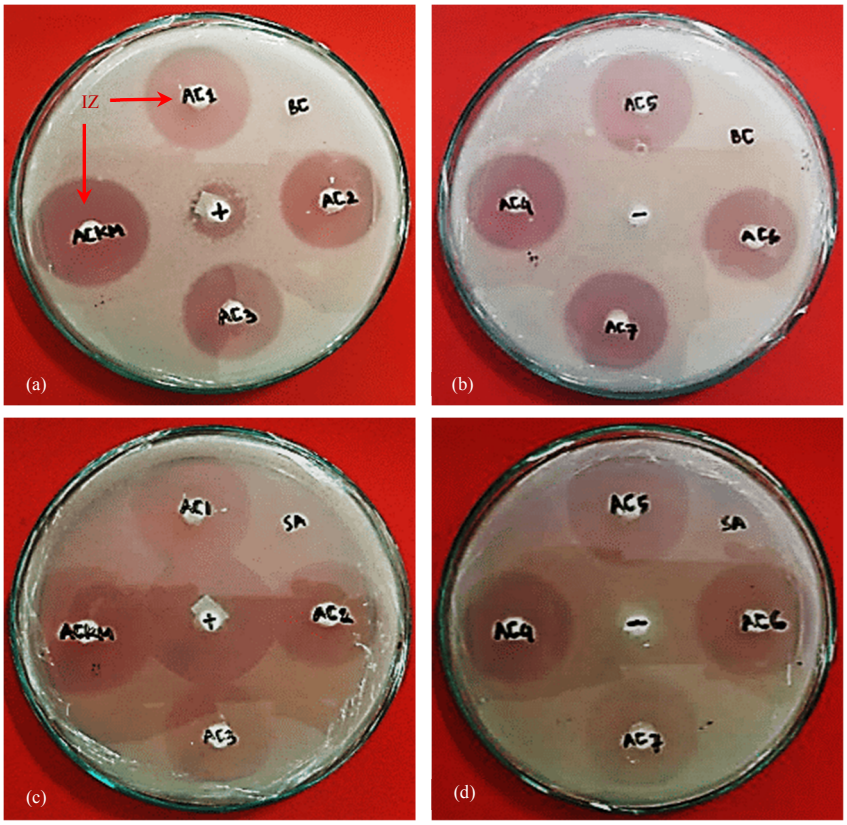

Fig. 1(a-d): Inhibition zone formed by the antimicrobial activity of cinnamon liquid smoke in (a-b) *Bacillus* sp. and (c-d) *S. aureus*

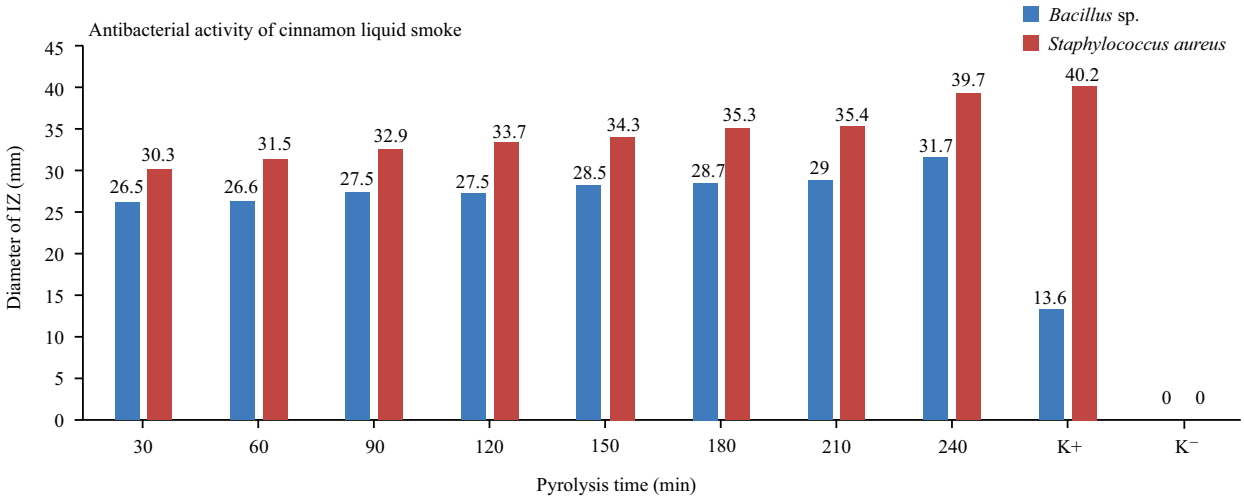

Fig. 2: Comparison of IZ diameter formed by the antimicrobial activity of cinnamon liquid smoke against *Bacillus* sp. and *S. aureus*

Phenol compounds and their derivatives in liquid smoke were influenced by lignin and pyrolysis temperature. Phenol and its derivatives were the results of lignin degradation at 400°C[18]. The most important pyrolysis result of lignocellulosic material (such as wood) was acetic acid and phenol in small amounts. The chemical content of the liquid smoke was antimicrobial and antioxidant[19].

Liquid smoke contained various compounds which were grouped into phenolic, acids and carbonyl compounds. These groups had an important role as antimicrobials, antioxidants, flavouring and colour forming (colouring). The two main compounds in liquid smoke that have a bactericidal or bacteriostatic effect were phenol and acetic acid[20].

Irwandi and Sukmawati[21] suggested that liquid smoke produced from *Piper betle* L. can inhibit the growth of *Salmonella* sp. as Gram-negative and *Staphylococcus aureus* as Gram-positive. Cinnamon has the ability to antimicrobial, anti-fungal, antiviral, antioxidant, anti-tumour capabilities, lower blood pressure, cholesterol and low-fat compounds[22].

The degree of acidity (pH) of liquid smoke was 2.8-3.1, which inhibited the growth of pathogenic bacteria[23]. Liquid smoke has been proved to suppress the growth of rotting bacteria and pathogens such as *Escherichia coli*, *Bacillus subtiliis*, *Pseudomonas* and *Salmonella*[24]. Therefore, liquid smoke can be applied directly to food to protect food quality, extend food shelf life by inhibiting or inactivating spoilage microorganisms and improve food safety by inhibiting or inactivating food-borne pathogens[25]. By comparing to several additives such as borax and formalin as an ingredient of food preservation (such as tofu), the use of liquid smoke was an alternative substitution. Formalin is a chemical property as a disinfectant and it was very dangerous if it was used as a preservative. Because formaline is a carcinogenic compound and cause death[26]. Preserving food with liquid smoke is not only feasible but also safe for consumption, because of no deposit of carcinogenic polycyclic aromatic hydrocarbons in preserved foods[27].

Minimum inhibitory concentration and minimum bactericidal kill concentration: The MIC was the lowest concentration of liquid smoke that showed no turbidity after comparing with NB control (contain no bacterial suspension in NB). MIC was calculated based on the turbid metric method (visual observation). In this study, the turbidity level of growth media was observed after incubating for 24 hrs. The result was negative (-) if no turbidity occurred in NB. Whereas, the result was positive (+) if NB turned turbid. Table 2 described the results of MIC of cinnamon liquid smoke toward bacterial growth.

Analysis of MIC was performed by diluting several concentrations of liquid smoke, ranged from 0.195-50% (Table 2). The MIC value for *Bacillus* sp. was found at a concentration of 1.56%, the growth medium was still clear. While at 0.78% concentration, the medium had changed to turbid. It indicated that a 0.78% concentration of liquid smoke was no longer able to inhibit the growth of *Bacillus* sp.

The MIC for *S. aureus* was found at 0.39%. At this concentration level, NB was still clear, while the smoke liquid in the level concentration of 0.195%, NB changed colour to cloudy, indicated that 0.195% liquid smoke was not able to inhibit the growth of *S. aureus*.

The MBC was the lowest level of antibacterial activity to eradicate bacteria characterized by no growth of microbes on solid medium or colony growth was less than 0.1% of the total number of initial inoculum on solid medium. The analysis of MBC of liquid smoke from cinnamon as antibacterial showed that liquid smoke had bacteriostatic activity. Testing results of plating NB which contained bacterial suspension and liquid smoke on solid NA was indicated by whether or not the colony of bacteria grows. The MBC test was carried out by plating the result of several concentrations of liquid smoke in the MIC dilution test on each 12 sterilized Petri dish containing NA. The MBC results were presented in Table 3.

The growth of *Bacillus* sp. and *S. aureus* colonies at several concentrations of cinnamon liquid smoke showed that at the concentration level of 3.125-50% there was no growth of *Bacillus* sp. colonies on NA. While at concentration level of 0.195-1.56%, *Bacillus* sp. grew on NA (1.1×107 CFU mL^{-1}, 1.56%, 6.2×107 CFU mL^{-1}, 0.78%, 2.1×108 CFU mL^{-1}, 0.39% and >300 CFU mL^{-1} (TBUD), 0.195%). In the negative control, numerous colonies grew than other Petri dish (2.9×108 CFU mL^{-1}). While in the positive control, by adding 1% of amoxicillin tablet, the colony of *Bacillus* sp. still grew (1.1×107 CFU mL^{-1}). MBC of *Bacillus* sp. at a concentration level of 3.125% was able to inhibit *Bacillus* sp. growth in NA.

The MBC of *S. aureus* in 0.78-50% of liquid smoke concentration inhibited the growth of *S. aureus* colonies on NA. Whereas at concentration of 0.195-0.39%, *S. aureus* was still able to grow on NA (3.8×107 CFU mL^{-1}, 0.39%, 2.0×108 CFU mL^{-1}, 0.195%). In the negative control, the number of colonies grew more than others (2.9×108 CFU mL^{-1}) and in the positive control with the addition of 1% amoxicillin tablet, the colony of *S. aureus* still grew (1.1×107 CFU mL^{-1}). The MBC at the concentration level of 0.39% inhibited the growth of *S. aureus* after on NA.

Nosartika *et al.*[28] found that coconut shell liquid smoke (CS-LS) inhibited the growth of *E. coli* better than chlorophenol Camphor menthol (ChM) as commercial root canal sterilization. The Minimum Inhibitory Concentration

Table 2: MIC analysis of cinnamon liquid smoke against *Bacillus* sp. and *S. aureus*

	MIC (%)		Testing result		Description	
No. tubes	*Bacillus* sp.	*S. aureus*	*Bacillus* sp.	*S. aureus*	*Bacillus* sp.	*S. aureus*
1	50	50	-	-	Clear	Clear
2	25	25	-	-	Clear	Clear
3	12.5	12.5	-	-	Clear	Clear
4	6.25	6.25	-	-	Clear	Clear
5	3.125	3.125	-	-	Clear	Clear
6	1.56	1.56	-	-	Clear	Clear
7	0.78	0.78	+	-	Turbid	Clear
8	0.39	0.39	+	-	Turbid	Clear
9	0.195	0.195	+	+	Turbid	Turbid
(-) control	0	0	++	++	Turbid	Turbid
(+) control	1	1	+	+	Rather turbid	Rather turbid
NB control	0	0	-	-	Clear	Clear

Table 3: Results of testing the minimum bactericidal concentration kill (MBC) of wood liquid smoke from cinnamon against *Bacillus* sp.

	Concentration (%)		Number of colony		SPC (CFU mL^{-1})	
No. tubes	*Bacillus* sp.	*S. aureus*	*Bacillus* sp.	*S. aureus*	*Bacillus* sp.	*S. aureus*
1	50	50	0	0	0	0
2	25	25	0	0	0	0
3	12.5	12.5	0	0	0	0
4	6.25	6.25	0	0	0	0
5	3.125	3.125	0	0	0	0
6	1.56	1.56	11	0	1.1×107	0
7	0.78	0.78	62	0	6.2×107	0
8	0.39	0.39	213	38	2,1×108	3.8×107
9	0.195	0.195	TBUD	197	TBUD	2.0×108
(-) control	0	0	288	288	2.9×108	2.9×108
(+) control	1	1	11	11	1.1×107	1.1×107
NB control	0	0	0	0	0	0

(MIC) and the Minimum Bactericidal Concentration (MBC) in a row are at concentrations of 3.125 and 12.5% for *E. coli* bacteria.

The results of MIC and MBC analysis showed that liquid smoke of wood had bacteriostatic activity. The bacteriostatic activity derived from the content of acid, phenol and carbonyl compounds in liquid smoke. This compound content can inhibit bacterial growth. The combination of these three compounds worked synergistically in inhibiting the growth of bacteria[29].

CONCLUSION

Liquid smoke from cinnamon wood was obtained from different pyrolysis time, including in the anti-bacterial group "very strong" in inhibiting the growth of *Bacillus* sp. and *S. aureus*. In this study, the longer the pyrolysis time the stronger liquid smoke inhibition. The best pyrolysis time of cinnamon wood was 240 min as the most effective time. The IZ of cinnamon liquid smoke with pyrolysis time of 240 min against *Bacillus* sp. was 31.7 mm, while the IZ of *S. aureus* was 39.7 mm. MIC was 1.56% for *Bacillus* sp. and 0.39% for *S. aureus*. The MBC was 3.13% for *Bacillus* sp. and 0.78% for *S. aureus*.

SIGNIFICANCE STATEMENT

This study discovered the best pyrolysis time to produce the most effective cinnamon liquid smoke was for 240 min that can be beneficial for inhibiting the growth of *Bacillus* sp. and *S. aureus*. This study will help the researcher to uncover the antimicrobial abilities against other bacteria or other microbes to explore. Thus a theory on the best affectivity of cinnamon liquid smoke as an antibacterial agent to inhibit bacterial growth may be proposed.

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