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Toxicity of Liquid Smoke Cinnamon (*Cinnamomum Burmannii*) Production of Ways for Purification and Different Concentration

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Abstract- This study aims to determine the nature of the toxicity of liquid smoke cinnamon obtained from the purification and concentration of different liquid smoke. This study was carried out experimentally using a factorial experiment in a completely randomized design of 8 (eight) treatment purification with 7 (seven) the concentration of liquid smoke with 3 replicates in order to obtain 168 experimental units. The treatment of liquid smoke purification include purification by distillation temperature of 100 ± 10 ° C; purification by distillation temperature of 140 ± 10 ° C; purification using activated charcoal, purification using activated charcoal and zeolite mixture (50:50), purification by decantation for 1 day, 2 days and decantation decantation 3 days. Treatment includes liquid smoke concentration of 0 ppm, 12.5 ppm, 25 ppm, 50 ppm, 100 ppm, 500 ppm and 1000 ppm. Variables observed consisted of the nature of toxicity in the form of measurement (%) mortality, probit and LC50. The results showed the test results of variance showed that differences in the way of purification provides significant effect on the percentage of mortality also in different concentrations and combinations of liquid smoke purification treatment with liquid smoke concentration significant effect (P <0.05) the percentage of mortality in toxicity. Further purification treatment combined with liquid smoke concentration no significant effect (P> 0.05) to the value of probit toxicity. The mortality percentage in treatment purification liquid smoke wood contained in the purification treatment of liquid smoke decantation for 3 days at 49.524 mm / ppb, probit value of 4.55 ppm, LC50 values of 7.104 ppm and the regression equation y = 2,7891x + 30.187 and values r2 = 0.6498. Furthermore, the greatest percentage of mortality in the treatment of liquid smoke concentration cinnamon shown by the treatment of 1500 ppm of 83.750%, probit value of 6.29 ppm, LC 50 value of 343.02 ppm and the regression equation y = 0.0712x + 25.577 and the value r2 = 0.5464. Based on the nature of the toxicity of combined treatment purification by decantation three days with liquid smoke concentration of 1000 ppm produces the largest percentage of 96.67% mortality, probit value of 6.808 ppm, LC 50 value of 40.33 ppm and the regression equation y = 5,9124x + 57.144 and the value of r2 = 0.6712

Index Terms- purification, concentration, liquid smoke cinnamon, toxicity, Probit.

I. Introduction

Cinnamon (*Cinnamomum burmannii*) is one of the traditional medicinal plants that have been studied usefulness long ago. Cinnamon can be used to cure canker sores, cough medicine, shortness of breath, stomach pain, diarrhea, flatulence, rheumatism, warm the stomach and as an anti-cancer [1]. The active compounds responsible for the anti-cancer activity in cinnamon allegedly was active substance sinamaldehid [2]. On the other side of cinnamon can be made of liquid smoke after the skin was taken. The problems that arise from liquid smoke cinnamon created using pyrolysis still contain toxins, so it is necessary to study the nature of the toxicity.

According to [3], one test of bioactivity that is easy, fast, inexpensive and accurately by using larval shrimp Artemia salina Leach, known as Brine Shrimp Lethality Test (BSLT). Shrimp larvae mortality test is one test method bioactivity in natural materials research compound. Use of shrimp larvae for the benefit of bioactivity studies have been conducted since 1956 and since then it has been a lot done on environmental studies, toxicity, and screening for bioactive compounds from the plant tissue. This test is a preliminary test to observe the pharmacological activity of a compound one of which is anti-cancer. The application for the bioactivity of the system by using the shrimp larvae, among others, to determine pesticide residues, local anesthetic, morpin derived compounds, mycotoxins, carcinogenicity compound and pollutants to the sea as well as an inexpensive alternative method for cytotoxicity assay [4]. The active compounds that have a high bioactivity known based on the value of 50% Lethal Concentration (LC_{50}), which is a value that indicates the concentration of toxic substances that can cause the death of test animals to 50%. Mortality data were obtained and processed by probit analysis formulated by [5] for determining the LC50 values at 95% confidence level. The chemical compound has the potential bioactive if it has LC_{50} values of less than 1,000 pg / ml [3].

BSLT test using larval shrimp Artemia salina done incubate the eggs in seawater assisted by aeration. Artemia salina eggs will hatch into larvae perfectly within 24 hours. A. salina larvae

are well used to test BSLT is aged 48 hours because if more than 48 hours it is feared the death was not due to toxicity of Artemia salina extract but by the limited supply of food [3]. The benefit of shrimp larvae of A. salina for this BSLT test is sensitive to the nature of the test material, the cycle time of life more quickly, easily bred and the price is cheap. A. salina sensitive nature likely caused by circumstances were very thin skin membrane that allows the diffusion of substances from the environment that affect the metabolism in the body. A. salina is found in almost all surface waters in the world who have a salinity range of 10 - 20g / L, it is this which causes easily bred. Newly hatched larvae are called nauplius oval and reddish color with a length of 400 µm with a weight of 15 g. Member body consisting of a pair of small antennae (or antenna anteluena I) and a pair of large antennae (antenna or antenna II). At the front of the two small antennae that are red spots that function as eyes (oselus). At the rear there is a pair of mandibular barbels magnitude (jaw) is small, whereas in the belly (ventral) side of the front there labrum [6].

Shrimp larvae toxicity test toxicity testing is one that is fast, safe, practical and economical for screening, fractionation, and determination of bioactivity of compounds of natural materials. National Cancer Institute of the United State of America (USA NCI) has found a significant relationship between toxicity testing on shrimp nauplius (*Bhrine Shrimp Lethality Test*) with inhibition of human tumor cells in vitro.

Given waste plantation crops such as cinnamon are still widely available in the province of West Sumatra, and yet provide optimum added value, then it needs to be processed into liquid smoke. On the other hand liquid smoke cinnamon grade 3 still met the toxic compounds, hence the need for purification activities. Other factors besides affecting tosksisitas purification is the concentration of liquid smoke. So far there has been no study the toxicity properties of liquid smoke obtained after pemurnian. Berdasarkan that it is necessary to study assessing the nature of liquid smoke toxicity of various purification with different concentrations of liquid smoke. This study aimed to find out the nature of the toxicity of combined treatment purification with different concentrations of liquid smoke.

II. RAW AND METHODS

Materials And Tools

1. Materials Research

a. Raw materials: The raw material liquid smoke cinnamon at a temperature of 400°C pyrolysis, zeolites, activated charcoal, Eggs Artemia salina Leach.

b. Chemicals: The materials used for the analysis of chemical properties of liquid smoke. For analysis of the chemical components of liquid smoke using methanol, reagent Meyer, DMSO, sea water, and distilled water

2.Tools used for research

The tools used in used in this study such as scales, flask, cup petrialis, electric stove, filter paper, oven, outoclaf, incubators, distillation apparatus, analytical balance, oven, porcelain dish, desiccator, filter, thermometer, pH meter , erlenmeyer 125 ml and 500 ml, beaker, filter paper, soxhlet, test tubes, centrifuge tubes, micro burette, pipette, pipette volumetric flask of 250 ml, centrifuge, spectrophotometer, pycnometer. The tools used in the analysis of phenol content is flask, pipette, erlemeyer, buret and sample bottle.

Research methods

This research was conducted at the Laboratory of the University Basic Ekasakti carried out experimentally using a factorial experiment in a completely randomized design of 8 (eight) treatment purification with 7 (seven) the concentration of liquid smoke with 3 replicates in order to obtain 168 experimental units. The treatment of liquid smoke purification include purification by distillation temperature of 100 ± 10 ° C; purification by distillation temperature of 140 ± 10°C; purification using activated charcoal, purification using activated charcoal and zeolite mixture (50:50), purification by decantation for 1 day, 2 days and decantation decantation 3 days. Treatment includes liquid smoke concentration of 0 ppm, 12.5 ppm, 25 ppm, 50 ppm, 100 ppm, 500 ppm and 1000 ppm.

The data were analyzed by analysis of variance on the real level of 5%, significantly different when followed by Tukey's test at 5 percent significance level [7].

Implementation research

Liquid smoke purification is done on raw materials cinnamon with pyrolysis temperature of 400 ± 10°C for standard sign issued by [8] and the toxicity of benzo(e)pyren lower than the second most other raw materials. Activity purification performed on liquid smoke cinnamon on pyrolysis 400°C silenced once 1 week to precipitate Tar, after standing for 1 week followed by administration of the treatment purification by distillation at a temperature of 100 ± 10°C and 140±10°C for 1 hour, filtering (absorption) using activated charcoal, activated charcoal mixture with zeolite (50:50) and zeolite and precipitation for 1,2 and 3 days. The stages of its activities as follows:

a. Distillation

In the process of distillation: a sample of liquid smoke cinnamon result of pyrolysis at temperatures of 400 ° C as much as 100 ml put in a distillation flask where the container where the distillation flask using oil as a good conductor of heat and kept heated using an electric heater. The distillation process is done when the temperature of the heating medium (oil) is already showing the desired temperature appropriate treatment that 100°C and 140°C. Interest distillation to take all factions and is set at a temperature of 100°C and a temperature of 140°C. At each temperature treatment made three replications. Temperatures shown are the temperature of liquid smoke in the distillation flask. The steam is formed and into the coolant pipe behind (condenser) and the distillate is collected in a flask. In this purification process is obtained quality liquid smoke II quality. Results of liquid smoke to analyze the results further purification toxicity [3].

b. Filtering (adsorption) using activated charcoal, mix AA + zeolite and zeolite

Liquid smoke cinnamon result of the pyrolysis temperature of 400°C as much as 100 ml of activated carbon mixed with as much as 3.5% [9] using a funnel further shaken and allowed to stand for 15 minutes after it is filtered using Whatman # 42. The

same activities carried on zeolite materials and a mixture of both ready-made, after settling 15 minutes filtered through Whatman filter paper No. 42. The result of the purification was done subsequently repeated 3 times and analyzed toxicity [3].

c. Precipitation

Liquid smoke prepared cinnamon in a measuring cup of 100 ml each were then deposited / decantation for 1, 2 and 3 days is done with three replications. This treatment refers to the results of research [10]. Furthermore, the analysis of toxicity [3].

d. Uji cytotoxic liquid smoke method Brine Shrimp Lethality Test (BSLT) bioassay system [3], [11]with the phases of work as follows:

1. Selection of eggs Artemia salina Leach

Selection of shrimp eggs made by soaking the eggs in distilled water for one hour. Good egg will sink whereas unfavorable egg will float.

2. Preparation of larvae Artemia Salina Leach

Preparation of shrimp larvae hatch shrimp done 48 hours before the test. Hatching eggs is done by immersing them in sea water sufficiently to illuminate the container that is not occupied by shrimp eggs with incandescent lights.

e. The division of the treatment group

In this study, shrimp larvae were divided into five treatment groups at random, namely:

1. Group K is 10 larvae shrimp fed with a liquid smoke concentrations of 0 ug / ml.
2. The group P1 is 10 larvae shrimp fed liquid smoke with a concentration of 12.5 ug / ml in the media.
3. P2 group is 10 larvae shrimp fed liquid smoke with a concentration of 25 ug / ml in the media.
4. P3 group is 10 larvae shrimp fed with a liquid smoke concentration of 50 ug / ml in the media.
5. P4 group is 10 larvae shrimp fed with a liquid smoke concentration of 100 ug / ml in the media.
6. P5 group is 10 larvae shrimp fed liquid smoke with a concentration of 500 ug / ml in the media.
7. P6 group is 10 larvae shrimp fed liquid smoke with a concentration of 1000 ug / ml in the media.

f. Implementation of toxicity tests

Test execution is done by first equalizing the final volume of liquid smoke results from the combined treatment of purification include purification by distillation temperature of 100 ± 10°C; purification by distillation temperature of 140±10°C; purification using activated charcoal, purification using activated charcoal and zeolite mixture (50:50), purification by decantation for 1 day, 2 days and decantation decantation 3 days with different concentrations of liquid smoke includes 0 ppm, 12.5 ppm, 25 ppm, 50 ppm, 100 ppm, 500 ppm and 1000 ppm is diluted by adding seawater advance into each test tube until liquid smoke above were mixed, then just put shrimp larvae that have been aged 48 hours in a series of test tubes containing liquid smoke which has been prepared respectively of 10 animals so that the volume in each tube to 5 ml. Test tube and then placed under incandescent light illumination for 24 hours, then counted the number of dead shrimp larvae. Standard criteria to assess mortality of shrimp larvae is when the shrimp larvae showed no movement during several seconds of observation.

g. Data collected

The data collected are primary data obtained from the amount of shrimp larvae died 24 hours after treatment in each treatment combination purification with different concentrations of liquid smoke.

h. Hatching Eggs Artemia salina

Artemia soaked in fresh water for 15-30 minutes. Then soaked in 10 liters of seawater. Hatching temperature is 25-30 ° C and pH ± ± 6-7. The eggs will hatch after 18-24 hours and the larvae are called nauplii. Nauplii ready to test BST after these larvae was 48 hours.

i. Uji toxicity BST Extract Method

Liquid smoke results of the combined treatment of raw materials with different pyrolysis temperature taken 50 mg, each dissolved in 5 ml of methanol. Created dilution 1000, 500, 100, 50, 25,12.5 and 0 ug / ml. Testing is done by inserting 10 larvae of Artemia salina was 48 hours into glass jars already containing 1 ml solution of 4 ml of liquid smoke and seawater. After 24 hours, the number of dead larvae were calculated with the aid of a magnifying glass.

Experimental design

The study was conducted using a factorial experimental designs 8 x 7 with three replications system completely randomized design (CRD) in order to obtain 168 experimental units. A factor is a liquid smoke purification include purification by distillation temperature of 100±10°C; purification by distillation temperature of 140 ± 10°C; purification using activated charcoal, purification using activated charcoal and zeolite mixture (50:50), purification by decantation for 1 day, 2 days and decantation decantation 3 days. Factor B level liquid smoke concentration is 0 ppm, 12.5 ppm, 50 ppm, 100 ppm, 500 ppm and 1000 ppm. The parameters measured were the number of dead Artemia 50% of the total larvae test. Then calculated LC_{50} values by entering the numbers probit (50% larval mortality trials).

Data analysis

BSLT the test data were analyzed using Sam [12] based on the calculation of the number of larvae were dead and the living. The death rate or mortality (%) is obtained by comparing the number of dead larvae divided by the total number. LC_{50} value is obtained by determining the value of probit, namely converting the value per cent of deaths by probit table. A substance is said to be active or toxic substances when the LC50 value of less than 1000 ppm to extract and less than 30 ppm of a compound [13]. Plotting data between probit with log concentration value will be obtained regression line:

$y = a + bx$

Information

y = 50 (expressed shrimp larvae that experienced the deaths of 50% after an incubation period of 24 hours).

a = slope

b = intersep

x = menyatakan concentration that causes death to 50% of larvae

III. RESULTS AND DISCUSSION

Toxicity test method Scrimp Brimp Lethality Test (BSLT)

a. Effect of purification of the liquid smoke toxicity properties of cinnamon

Based on analysis of variance scales of interaction between a combination of purification with different concentrations of liquid smoke to (%) mortality of Artemia means purification with different concentrations together affect the percentage of mortality of Artemia. For Probit ANOVA test on the toxicity due to differences purification treatment with different concentrations of liquid smoke did not show any interaction. Average test results of toxicity (%) mortality and Probit liquid smoke given purification treatment with different concentrations of liquid smoke can be seen in Table 1 below.

Table 1. Summary of the average liquid smoke toxicity properties of cinnamon in a manner different purification

Purification Liquid Smoke	Mortalitas (%)	Probit (ppm)	LC_{50} (ppm)	Regresi Equation
1. Destillation 100 ±10°C	24,762 d	3,05	7,104	Y = 2,7891x + 30,187
2. Destillation 140 ±10°C	35.238 c	3,38		r^2 = 0,6498
3. Activated charcoal filtering (AA) : 3,5%	46.667 ab	3,90		
4. Activated charcoal filtering (AA) + Zeolit (Z) comparison 50:50as such us 3,5%	44.286 b	278,57		
5. Filtering Zeolit (Z) : 3,5%	48.095 ab	507,71		
6. The deposition for 1 day	46.667 ab	180,90		
7. The deposition for 2 day	46.667 ab	6,02		
8. The deposition for 3 day	49.524 a	4,55		

* Different superscript letters in columns averaging showed significant difference (P <0.05)

In table 1 shows the average% mortality of Artemia salina in the deposition treatment for 3 days showed the greatest mortality rate (49.524%) was not significantly different from the deposition of 1.2 days, with zeolite filtration, and filtration with activated charcoal and significantly different treatments other. This means that liquid smoke cinnamon by precipitation possess higher mortality compared with other purification. A high percentage of mortality which means the effect of toxins in the smoke liquid such as cinnamon aldehyde compounds, ketone, phenol gives toxic effects, it is shown by the death of Artemia salina.

In the liquid smoke purification by distillation temperature of 100°C shows the percentage of deaths Artemia lower than the temperature of 140°C. It is suspected the higher the temperature of distillation will lead to a growing number of chemical compounds are formed, causing a higher toxic effects. BSLT toxicity tests carried out by determining the LC_{50} value of the activity of the active components of the plant against the larvae of Artemia salina Leach. An extract is said to be toxic by the methods BSLT if the extract can kill 50% of test animals at concentrations of less than 1000 ppm [3].

Toxicity tests conducted on different purification indicate LC_{50} values of 7.104 ppm means to kill as many as 50% Artemia require liquid smoke of 7.104 ppm. This shows that the liquid smoke compounds from different purification has the potential acute toxicity by BSLT method and can be developed as anticancer because LC50 <30 ppm [13]. Based on regression analysis that shows how different purification relationships that are not so closely to the value probit with R^2 values of 0.6498. According to [14] said the potential for acute toxicity possessed liquid smoke is influenced by the content of secondary metabolites which owned the extract. The presence of the flavonoid extract in the cell environment, causing the OH groups in flavonoids bind to the cell membrane integral proteins. This causes terbendungnya active transport of Na + - K +. Active transport stop influx of Na + ions causes uncontrolled into the cells, causing rupture of the cell membrane. Rupture of cell membranes that causes the death of larvae of Artemia salina.

BSLT toxicity test method is acute toxicity test where the toxic effect of a compound is determined in a short time, which ranges up to 24 hours after administration of the test dose [3]. BSLT method chosen because it is one of the methods bioactivity that is easy, fast, cheap and accurate. This method is often used to determine the toxicity of the nature/botanical extracts as well as for screening compounds for their anticancer positive correlation between the methods BSLT with cytotoxic test using cancer cell cultures [15]. BSLT toxicity tests carried out by determining the LC_{50} value of the activity of the active components of liquid smoke to larva Artemia salina Leach. A liquid smoke is said to be toxic by the methods BSLT if liquid smoke can cause death of 50% of test animals at concentrations of less than 1000 ppm [3]. Toxicity testing of liquid smoke is done 3 times (triplo). Larval mortality data obtained are processed by probit analysis formulated by [5] for determining the LC_{50} values at 95% confidence level.

b. Effect of concentration of liquid smoke to the nature of the toxicity

Average test results of toxicity (%) mortality and Probit smoke can be seen in table 2 below. liquid smoke treatment given different concentrations of liquid

Table 2. Activities average liquid smoke toxicity properties of cinnamon with different concentrations of liquid smoke to the bacteria E. coli with that diffusion method.

Liquid Smoke Concentration	Mortalitas (%)	Probit	LC 50	Regresi equation
1. 0 ppm	0,0000 f	1,71	343,02 ppm	Y = 0,0712x + 25,577
2. 12,5 ppm	5,4167 e	156,37		R2 = 0,5464
3. 25 ppm	16,667 d	331,56		
4. 50 ppm	39,583 c	201,98		
5. 100 ppm	75,000 b	248,58		
6. 500 ppm	78,750 b	5,59		
7. 1000 ppm	83,750 a	6,29		

* Different superscript letters in columns averaging showed significant difference (P <0.05)

In Table 2 shows the phenomenon that the higher the concentration of liquid smoke is used there will be an increase in the percentage of deaths Artemia salina. This means that the higher the concentration of liquid smoke is used then the concentration of liquid smoke will become increasingly concentrated, so the percentage of mortality of Artemia salina also be increased. Toxicity tests conducted on liquid smoke from the way different purification indicate LC50 values of 343.02 ppm means to kill Artemia 50% require liquid smoke at 343.02 ppm. This shows that the concentration of liquid smoke in a manner different purification does not have the potential for acute toxicity according to the method BSLT and can not be developed as anticancer ie LC_{50} value <30 ppm [16]. Based on the regression line that means different purification showed a strong relationship with R2 0.5464. The appearance of the image regression line shows the increase in value when the concentration of liquid smoke probit raised except liquid smoke concentration of 500 ppm and 1000 ppm. It is alleged by the higher concentration of liquid smoke, the more there are compounds such as aldehydes, ketones and phenols, causing the value of probit tendency to rise. The compounds are secondary metabolites include saponins and glycosides. Such compounds can act as stomach poisoning or stomach poison. Therefore, when these compounds into the body of larvae, larval digestive system will be disrupted. In addition, these compounds inhibit the taste receptors in the mouth of the larvae. This resulted in the larvae fail to get a taste stimulus that can not recognize the food. As a result, the larvae die of starvation [17], [18].

c. Effect of purification and concentration of liquid smoke to the nature of the toxicity

The observation of the average results of toxicity tests given liquid smoke purification treatment with different concentrations of liquid smoke to (%) mortality of Artemia, probit and LC_{50} value can be seen in Table 3 below.

Table 3. Percentage of Artemia salina mortality (%), the value of Probit, LC50 and the regression equation by giving treatment purification with different concentrations of liquid smoke

Purification	Liquid Smoke Concentration	log kons	(%) mortalitas	Probit	LC_{50} (ppm)	Regresi equation
Destillation 100 + 10°C	0 ppm	0	0±0,00 n	0±0,00a	-	Y = 0 ; r2 = #N/A
	12.5 ppm	1.097	3.333±5,77 mn	3.163±1,73a	90.88	Y=0,5161x+ 3,0939 r^2=0,2593
	25 ppm	1.398	10±0,00 lmn	3.718±0,00a	42.68	Y = 0,873x+ 12,74 r^2 = 0,4801
	50 ppm	1.699	26.667±5,77 ijkl	4.378±0,00a	10.34	Y = 1.7851x + 31.55 r^2 = 0,3901
	100 ppm	2	43.333±5,77 ghi	4.831±5,77a	0.58	Y = 4,9205x + 52.858 r^2 = 0,5648

	500 ppm	2.69	43.333±5,77 ghi	4.831±5,77a	0.71	Y = 5.5165x + 53.925 r^2 = 0,6394
	1000 ppm	3	46.667±5,77 fgh	4.917±5,77a	40.33	Y = 5,9124 + 57,144 r2 = 0.6712
Destillation 140 + 10°C	0 ppm	0	0±0,00 n	0±0,00a	-	Y = 0 ; r2 = #N/A
	12.5 ppm	1.097	3.333±5,77 mn	3.163±1,73a	90.88	Y=0,5161x+ 3,0939 r^2=0,2593
	25 ppm	1.398	16.667±5,77 klmn	4.034±5,77a	42.68	Y = 0,873x+ 12,74 r^2 = 0,4801
	50 ppm	1.699	36.667±5,77 hij	4.660±0,00a	10.34	Y = 1.7851x + 31.55 r^2 = 0,3901
	100 ppm	2	60.000±10 efg	5.253±0,00a	0.58	Y = 4,9205x + 52.858 r^2 = 0,5648
	500 ppm	2.69	63.333±11,55 def	5.340±0.00a	0.71	Y = 5.5165x + 53.925 r^2 = 0,6394
	1000 ppm	3	66.667±5,77 cde	5.432±0.00a	40.33	Y = 5,9124 + 57,144 r2 = 0.6712
activated charcoal (AA)	0 ppm	0	0.000±0,00 n	0±0.00a	-	Y = 0 ; r2 = #N/A
	12.5 ppm	1.097	6.667±5,77 mn	3.501±1.73a	90.88	Y=0,5161x+ 3,0939 r^2=0,2593
	25 ppm	1.398	16.667±5,77 klmn	4.034±0,57a	42.68	Y = 0,873x+ 12,74 r^2 = 0,4801
	50 ppm	1.699	40.000±10 hi	4.747±0,57a	10.34	Y = 1.7851x + 31.55 r^2 = 0,3901
	100 ppm	2	86.667±5,77 ab	6.112±0,57a	0.58	Y = 4,9205x + 52.858 r^2 = 0,5648
	500 ppm	2.69	86.667±5,77 ab	6.112±0,57a	0.71	Y = 5.5165x + 53.925 r^2 = 0,6394
	1000 ppm	3	90.000±0,00 a	6.282±0,00a	40.33	Y = 5,9124 + 57,144 r2 = 0.6712
Zeolit + AA	0 ppm	0	0.000±0,00 n	0±0,00a	-	Y = 0 ; r2 = #N/A
	12.5 ppm	1.097	6.667±5,77 mn	3.501±2,14a	90.88	Y=0,5161x+ 3,0939 r^2=0,2593
	25 ppm	1.398	16.667±5,77 klmn	4.034±2,40a	42.68	Y = 0,873x+ 12,74 r^2 = 0,4801
	50 ppm	1.699	46.667±5,77 fgh	4.917±0,57a	10.34	Y = 1.7851x + 31.55 r^2 = 0,3901
	100 ppm	2	70.000±10 bcde	5.524±0,00a	0.58	Y = 4,9205x +

						52.858
	500 ppm	2.69	80.000±10 abcd	5.842±0,57a	0.71	$r^2 = 0,5648$ Y = 5.5165x + 53.925 $r^2 = 0,6394$
	1000 ppm	3	90.000±10 a	6.282±1,52a	40.33	Y = 5,9124 + 57,144 r2 = 0.6712
Zeolit (Z)	0 ppm	0	0.000±0,00 n	0±0,00a	-	Y = 0 ; r2 = #N/A
	12.5 ppm	1.097	3.333±5,77 mn	3.163±1,73a	90.88	Y=0,5161x+ 3,0939 r^2=0,2593
	25 ppm	1.398	20.000±0,00 jklm	4.158±0.00a	42.68	Y = 0,873x+ 12,74 $r^2 = 0,4801$
	50 ppm	1.699	43.333±5,77 ghi	4.831±2,74a	10.34	Y = 1.7851x + 31.55 $r^2 = 0,3901$
	100 ppm	2	86.667±5,77 ab	6.112±3,37a	0.58	Y = 4,9205x + 52.858 $r^2 = 0,5648$
	500 ppm	2.69	90.000±0,00 a	6.282±0,00a	0.71	Y = 5.5165x + 53.925 $r^2 = 0,6394$
	1000 ppm	3	93.333±5,77 a	6.948±1,15a	40.33	Y = 5,9124 + 57,144 r2 = 0.6712
The deposition for 1 day	0 ppm	0	0.000±0,00 n	0±0,00a	-	Y = 0 ; r2 = #N/A
	12.5 ppm	1.097	6.667±5,77 mn	3.501±1,73a	90.88	Y=0,5161x+ 3,0939 r^2=0,2593
	25 ppm	1.398	16.667±5,77 klmn	4.034±2,14a	42.68	Y = 0,873x+ 12,74 $r^2 = 0,4801$
	50 ppm	1.699	33.333±5,77 hijk	4.568±0,00a	10.34	Y = 1.7851x + 31.55 $r^2 = 0,3901$
	100 ppm	2	86.667±5,77 ab	6.112±0,57a	0.58	Y = 4,9205x + 52.858 $r^2 = 0,5648$
	500 ppm	2.69	90.000±0,00	6.282±0,00a	0.71	Y = 5.5165x + 53.925 $r^2 = 0,6394$
	1000 ppm	3	93.333±5,77 a	6.948±1,15a	40.33	Y = 5,9124 + 57,144 r2 = 0.6712
The deposition for 2 day	0 ppm	0	0.000±0,00 n	0±0,00a	-	Y = 0 ; r2 = #N/A
	12.5 ppm	1.097	3.333±5,77 mn	3.163±1,73a	90.88	Y=0,5161x+ 3,0939 r^2=0,2593
	25 ppm	1.398	16.667±5,77 klmn	4.034±0,58a	42.68	Y = 0,873x+ 12,74
	50 ppm	1.699	43.333±5,77 ghi	4.831±0,52a	10.34	

						$r^2 = 0,4801$ Y = 1.7851x + 31.55
	100 ppm	2	83.333±5,77 abc	5.966±0,25a	0.58	$r^2 = 0,3901$ Y = 4,9205x + 52.858
	500 ppm	2.69	86.667±5,77 ab	6.112±0,25a	0.71	$r^2 = 0.5648$ Y = 5.5165x + 53.925
	1000 ppm	3	93.333±5,77 a	6.948±1,40a	40.33	$r^2 = 0,6394$ Y = 5,9124 + 57,144
The deposition for 3 day	0 ppm	0	0.000±0,00 n	0±0,00a	-	r2 = 0.6712 Y=0,5161x+ 3,0939
	12.5 ppm	1.097	10.000±10,00 lmn	3.718±2,28a	90.88	r^2=0,2593 Y = 0,873x+ 12,74
	25 ppm	1.398	20.000±0,00 jklm	4.158±0,00a	42.68	$r^2 = 0,4801$ Y = 1.7851x + 31.55
	50 ppm	1.699	46.667±5,77 fgh	4.917±0,15a	10.34	$r^2 = 0,3901$ Y = 4,9205x + 52.858
	100 ppm	2	83.333±5,77 abc	5.966±0,25a	0.58	$r^2 = 0,5648$ Y = 5.5165x + 53.925
	500 ppm	2.69	90.000±0,00 a	6.282±0,00a	0.71	$r^2 = 0,6394$ Y = 5,9124 + 57,144
	1000 ppm	3	96.667±5,77 a	6.808±1,41a	40.33	r2 = 0.6712

Description: * Different superscript letters in columns averaging showed significant difference (P <0.05)

Based on Table 3 above shows that the higher the concentration of liquid smoke used in each purification the higher the number of toxicity. This is presumably due to the higher concentration of liquid smoke is used then the concentration was so high that the higher the level of toxicity. Based on the way of purification that purification by distillation at 100°C showed the lowest toxicity levels at various concentration levels of liquid smoke is used. It is presumed by purification by distillation would vaporize many toxic substances contained in the liquid smoke so that the nature of toxicity becomes lower.

Toxicity tests conducted on liquid smoke cinnamon in a manner different purification and concentration of different shows LC_{50} values at a concentration of liquid smoke 0 of 0 ppm, 12.5 ppm concentration of 90.88 ppm, 25 ppm of 42.68 ppm , concentration of 50 ppm 10.34 ppm, 100 ppm concentration was 0.58 ppm, 500 ppm concentration was 0.71 ppm and 1000 ppm of 40.33 ppm. According [19], the level of toxicity of a pollutant in the fish can be divided into the following criteria: (a) <1 mg / L classified Very high, 1-10 mg / L is classified, 10-100 mg / L relatively mild and> 100 mg / L relatively mild. Based on the above criteria, the LC50 values at liquid smoke cinnamon purification with different concentrations of liquid smoke has a very high level of toxicity to moderate. Potential acute toxicity possessed liquid smoke is influenced by the content of secondary metabolites which owned the liquid smoke. According to [14], the flavonoid extract in the cell environment, causing the OH groups in flavonoids bind to the cell membrane integral proteins. This causes unstoppable active transport of Na + - K +. Active transport stop influx of Na + ions causes uncontrolled into the cells, causing rupture of the cell membrane. Rupture of cell membranes that causes the death of larvae of Artemia salina.

In addition to flavonoids, there are some secondary metabolites are present in liquid smoke. The compounds are secondary metabolites include saponins and glycosides. Such compounds can act as stomach poisoning or stomach poison. Therefore, when these compounds into the body of the larvae, the larvae digestive system will be disrupted. In addition, these compounds inhibit the taste receptors in the mouth of the larvae. This resulted in the larvae fail to get a taste stimulus that can not recognize the food. As a result, the larvae die of starvation [17], [18].

IV. CONCLUSION

1. The percentage mortality on treatment purification liquid smoke wood contained in the purification treatment of liquid smoke decantation for 3 days at 49.524 mm / ppb, probit value of

4.55 ppm, LC_{50} values of 7.104 ppm and the regression equation $y = 2,7891x + 30.187$ and the value of $r^2 = 0.6498$.

2. The percentage of mortality in the treatment of liquid smoke concentration cinnamon shown by the treatment of 1500 ppm of 83.750%, probit value of 6.29 ppm, LC_{50} value of 343.02 ppm and the regression equation $y = 0,0712x + 25.577$ and the value $r^2 = 0.5464$.

3. The nature of the toxicity of combined treatment purification by decantation three days with liquid smoke concentration of 1000 ppm produces the largest percentage of 96.67% mortality, probit value of 6.808 ppm, LC 50 value of 40.33 ppm and the regression equation $y = 5,9124x + 57.144$ and the value of $r^2 = 0.6712$

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