

# Effectiveness of Soursop Seed Extract (*Annona muricata* L) as a Natural Repellent on *Aedes Aegypti*



S. Alhamda, E. Barlian, A. Razak, M. Sari, N. Herawati

**Abstract:** *Background: Dengue Hemorrhagic Fever (DHF) is a disease caused by the dengue virus. City Regencies in Indonesia contracted DHF in 2014 amounting to 84.74%, 2015 amounted to 86.77%, and in 2016 amounted to 90.08%. Soursop plants can act as a repellent (insect repellent). The study aimed was to determine the effectiveness of soursop seed extract (*Annona muricata* L) as a natural repellent against *Aedes aegypti* mosquitoes. Method: The study was experimental with the independent variable concentration of soursop seed extract (*Annona muricata* L) and the dependent variable was *Aedes aegypti* mosquito that perched on the hand which had been sown with soursop seed extract (*Annona muricata* L). The sample of this study was 500 *Aedes aegypti* mosquitoes. This study used 5 concentrations and 5 repetitions for 6 hours of observation. The study was conducted on August 6-10 2018 starting at 08.30-13.30. Result: This study indicate that the concentration that concentrations of 20%, 40%, 60%, 80% and 100% have a mean of 75.2%, 90.8%, 93.5%, 95% and 97.7%, respectively. Based on the results of the ANOVA test, the results of the V-watch observation were obtained 61.60%, 85.07%, 89.89%, 92.88%, and 95.42% respectively. Conclusion: From the results obtained, the concentration of effective soursop seed extract is a concentration of 80% and 100% with a protection power of > 90%. The most effective concentration is 100%. It is recommended to the public to use soursop seed extract as a natural repellent to reduce the use of chemicals containing DEET (N, N-diethylmetatoluamide).*

**Keywords:** *aedes aegypti; DHF; soursop seeds; natural; repellent*

## I. INTRODUCTION

Dengue Hemorrhagic Fever (DHF) is a disease caused by the dengue virus belonging to the Arthropod-Borne Virus, the genus Flavivirus, and the family Flaviviridae. DHF is transmitted through mosquito bites from the genus *Aedes*, especially *Aedes aegypti* (Indonesian Ministry of Health, 2015, p.187). *Aedes aegypti* is the primary carrier (primary vector) and with *Aedes albopictus* females creates a cycle of dengue spread<sup>1</sup>.

Revised Manuscript Received on October 30, 2019.

\* Correspondence Author

**S. Alhamda\***, Doctorate Program in Environmental Science, Padang State University, West Sumatra Indonesia email: syukra909604@gmail.com

**E. Barlian**, Professional in Environmental Science, Padang State University, West Sumatra Indonesia email: eribarlian@yahoo.com

**A. Razak**, Associate Professor in Environmental Science, Padang State University, West Sumatra Indonesia email: ar710322@gmail.com

**M. Sari**, Faculty of Public Health, University of Fort De Kock Bukittinggi, West Sumatra Indonesia email: milasari11326@gmail.com

**N. Herawati**, Polytechnic of Health Padang, West Sumatra Indonesia email: nova\_syukra@yahoo.co.id

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According to WHO data (2014), Dengue hemorrhagic fever was first reported in Southeast Asia in 1954, namely in the Philippines, then spread to various countries. Before 1970, only 9 countries experienced dengue outbreaks, but now DHF has become an endemic disease in more than 100 countries, including Africa, America, the Eastern Mediterranean, Southeast Asia and the Western Pacific which have the highest rates of dengue cases. The number of cases in America, Southeast Asia and the Western Pacific has surpassed 1.2 million cases in 2008 and more than 2.3 million cases in 2010. In 2013 there were reported as many as 2.35 million cases in America, of which 37,687 cases were severe dengue. The development of dengue cases at the global level is increasing, as reported by the World Health Organization (WHO), which is from 980 cases in almost 100 countries in 1954-1959 to 1,016,612 cases in almost 60 countries in 2000-2009.<sup>2</sup>

In Indonesia, based on disease control data issued by the Ministry of Health in 2016, it is known that the number of City Districts that contracted DHF in 2014 was 84.74%, in 2015 it was 86.77%, and in 2016 amounted to 90.08%. This figure shows an increase in cases each year.<sup>3</sup>

In the Province of West Sumatra, the Regency/City that has contracted DHF has increased every year. It is known that the number of regencies/cities in West Sumatra Province is 19 regencies/cities. In 2014 and 2015 there were 18 districts / cities with dengue cases, in 19 regions there were 19 districts / cities. The data shows that every Regency/City in West Sumatra Province has dengue cases.<sup>3</sup>

The number of DHF sufferers per Regency/City in West Sumatra Province in 2015 was 2,282 cases with 12 deaths (IR = 45.75% per 100,000 people and CFR = 1%). In 2016 the number of cases was 3,985 cases with 18 deaths (IR = 75.75% per 100,000 people and CFR = 0.4%).<sup>3</sup>

In Agam Regency, dengue cases in 2015 were 280 people (IR = 58.7 per 100,000 people and CFR = 0), in 2016 there were 356 cases (IR = 62.1 per 100,000 people and CFR = 0). Compared to the target of the 2016 Agam District Strategic Plan, the dengue morbidity rate was 33 per 100,000 people, thus the morbidity rate in Agam Regency in 2016 was still above the target, in 2016 there was an increase in cases compared to 2015.<sup>4</sup> The mosquito repellents currently recommended are those containing N, N-diethyl-metoluamide (DEET) as active ingredients. DEET can repel mosquitoes, mites/ticks and other orthophodies when applied to skin and clothing. At present almost all repellents in the form of lotions in the market contain DEET. DEET has a very good repellent power but in its use can cause hypersensitivity and irritation<sup>5</sup>.

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DEET concentrations up to 50% are recommended for adults and children over 2 months and lower concentrations will not last long in the body so it is necessary for replication.<sup>6</sup>

The active substance DEET in anti-mosquito products is corrosive so that it can cause skin irritation, endanger the skin that is injured and the mucous membrane of the body. The higher the level of DEET in a product, the higher the level of corrosion in the skin.

Soursop plants can act as insecticides, larvacides, repellents (insect repellents) and antifeedant (food inhibitors) by working as contact poisons and poisons. Essential oils on soursop function as repellents, compounds of plant essential oils chemical compounds proved to affect locomotor activity. The odor contained in essential oils seeps into the pores of the skin and the arena of body heat and the environment, essential oils will evaporate into the air. This odor will be detected by chemical receptors contained in mosquito antennae and transmitted to nerve impulses, responded to the brain so that the mosquitoes will express themselves to avoid. Soursop leaves contain essential oils which are equal to 0.01% (Kossouh, 2007, p.307-309). Soursop seeds contain essential oils 42-45%.<sup>7</sup>

According to Ardraviz (2012) that soursop seeds, skin, stem and roots contain secondary metabolites, namely alkaloids. Yasril (2011) said that the bioactive content contained in soursop seeds is an alkaloid compound consisting of acetogenin and annonaine. Maryani (1995) in Tohir (2010) suggested that soursop seeds contain bioactive acetogenin, a compound that is insecticidal and anti-feedant<sup>8</sup>. Previous research uses soursop leaf extract as a repellent for *Aedes aegypti* mosquitoes conducted by Nurlaeli (2016), from the results of the study it is known that soursop leaf extract can be used as a repellent with a concentration of 95% can protect 75, 01% with 6 hours of observation. Another research on repellent was also carried out by Rizma (2017), from the research it was found that soursop seed extract with a concentration of 35% could protect 94.8% with a length of observation of 3 hours. Repellent is considered effective if the observations are carried out for 6 hours and the protection results are above 90%.<sup>9</sup> The increase in dengue cases every year and the limited research on the utilization of soursop seed extract (*Annona muricata* L) as a repellent against *Aedes aegypti* mosquito which is a vector of dengue disease is the reason the researchers conducted the study entitled "Effectiveness of Soursop Seed Extract (*Annona muricata* L) as Natural Repellent Against *Aedes aegypti* Mosquitoes".

## II. SUBJECT & METHOD

This research was conducted at the Central Laboratory of Science at the Faculty of Public Health University of Fort De Kock. The time of the study was carried out in July-August 2018. This research is experimental where *Aedes aegypti* mosquitoes receive direct treatment. *Aedes aegypti* mosquito was inserted into the observation cage with a size of 50 cm x 30 cm x 30 cm and then put into the hand that had been smeared with soursop seed extract (*Annona muricata* L) with various concentrations. The design of this study was the post-test only control group design. This design was chosen because it was not pretested against the sample before treatment.

The object of this research is adult female *Aedes aegypti* mosquitoes, with the criteria: Inclusion criteria; Female

*Aedes aegypti* mosquitoes, *Aedes aegypti* mosquitoes aged 2-5 days, live mosquitoes. Exclusion Criteria; Mosquitoes that die before treatment. Many treatments, the number of samples of *Aedes aegypti* mosquitoes per observation cage is 20 animals. The number of repetitions can use the formula as follows:

$$(t-1)(r-1) \geq 15$$

Then the least number of repetitions of treatment is done 5 times. So the total number of samples is:

The number of mosquitoes per box x number of repetitions x number of treatments

$$20 \times 5 \times 5 = 500 \text{ tails}$$

## III. RESULT & DISCUSSION

Soursop seeds are cleaned, dried by aerated in the open air which is protected from sunlight for 7 days until it is completely dry, there is no water content, for 7 days the factors of temperature and humidity are considered so it is not easily overgrown with fungi. After that in the oven to make sure it is completely dry for 2-3 hours. The dried soursop seeds are then ground/mashed. Weigh the fine soursop seeds as much as 500 grams then soak them in ethanol 96% solvent with a ratio of 1: 3 (b / v) Inserted into the microwave for 20 minutes at medium temperature. Then left for 1 x 24 hours. After 1 x 24 hours then filtered with filter paper, the results of filtering in the oven for 5 days at 40oC, so that ethanol evaporates to produce thick extracts ready for use. Save the extract made into the refrigerator. Making Solution Treatment :

By using the dilution formula:  $M_1 \times V_1 = M_2 \times V_2$

**Table 1:**  
Calculation of concentration of Soursop Seed Extract

Variation of concentrate	Dilution
Group 1 : 20 %	$M_1 \times V_1 = M_2 \times V_2$ $100 \% \times V_1 = 20\% \times 25 \text{ ml}$ $100\% V_1 = 500 \text{ ml}$ $V_1 = 5 \text{ ml}$
Group 2 : 40 %	$M_1 \times V_1 = M_2 \times V_2$ $100 \% \times V_1 = 40\% \times 25 \text{ ml}$ $100\% V_1 = 500 \text{ ml}$ $V_1 = 10 \text{ ml}$

Group 3 : 60 %  $M_1 \times V_1 = M_2 \times V_2$   
 $100 \% \times V_1 = 60\% \times 25 \text{ ml}$   
 $100\% V_1 = 500 \text{ ml\%}$   
 $V_1 = 15 \text{ ml}$

Group 4 : 80 %  $M_1 \times V_1 = M_2 \times V_2$   
 $100 \% \times V_1 = 80\% \times 25 \text{ ml}$   
 $100\% V_1 = 500 \text{ ml\%}$   
 $V_1 = 20 \text{ ml}$

Group 5 : 100 %  $M_1 \times V_1 = M_2 \times V_2$   
 $100 \% \times V_1 = 100\% \times 25 \text{ ml}$   
 $100\% V_1 = 500 \text{ ml\%}$   
 $V_1 = 25 \text{ ml}$

Breeding mosquitoes, *Aedes aegypti* eggs are soaked in plastic trays that have been given aquates for hatching eggs. Then after being larvae transferred into an open container filled with clean water. Larvae are fed with fish feed mixture. The mosquito larvae that have become pupae are then transferred to mosquito breeding cages of 30cm x 30cm x 30cm. After that, there were 500 adult mosquitoes which were scattered to the observation cage, 20 each. How It Works The Reppellent Effectiveness Test of Soursop Seed Extract (*Annona muricata* L) on *Aedes aegypti* mosquitoes. Prepare female *Aedes aegypti* mosquitoes with the age of 2-5 days as many as 20 birds per cage and reported. Prepare soursop seed extract with a concentration of 20%, 40%, 60%, 80%, 100%. The human volunteer left hand is smeared with soursop seed extract at a dose of 3 ml evenly from the tip of the finger to the wrist. Then the left hand is inserted into a cage that has been filled with mosquitoes through the hole. Count lots of mosquitoes that perch and immediately do it so that the mosquitoes don't suck blood. This is so that there are no mosquitoes that are full during testing which will interfere with the attraction of mosquitoes. Observe for 5 minutes. The right hand of the human volunteer is inserted into the same cage after removing the left hand. Human volunteer right hand is not treated, because it is used as a control. Count lots of mosquitoes that perch and immediately do it so that the mosquitoes don't suck blood. This is so that there are no mosquitoes that are full during testing which will interfere with the attraction of mosquitoes. Observe for 5 minutes. After testing of the left and right hands are done, awaited one hour later for the next repetition in the same way until the 6th hour. The use of a one-hour interval with a duration of five minutes is considered to be representative for testing protection power. In one day the test was only carried out for one concentration of soursop seed extract. One day testing was carried out by five human volunteers using a different test enclosure<sup>9</sup>.

Benefits of soursop seed extract (*Annona muricata* L) are determined based on the protection power calculated by the formula:

$$\text{Protection power (PP)} = \frac{(K-R)}{K} \times 100\%$$

K: the number of mosquitoes perched on the control hand  
 R: the number of mosquitoes perched on the treatment hands

If the result of the calculation of protection power is > 90%, then the extract is assumed to be effective (Pesticide Commission of the Ministry of Agriculture, 1995). The results obtained from the protection power formula above were then analyzed statistically using two-way Variance (ANOVA) analysis to find out whether there were significant differences in protection power between treatment groups and testing times. Calculation of the number of mosquitoes perched for 5 minutes per treatment. The 0 o'clock starts at 8:30 a.m. WIB, the second hour starts from 10:30 a.m. WIB, the third hour starts at 11:30 a.m., the IV hour starts at 12:30 a.m., the V hour starts at 13:30 WIB. The following is the result of calculating the average number of *Aedes aegypti* mosquitoes that perch in the hands of volunteers. The right hand is in control, while the left hand is treated with soursop seed extract (*Annona muricata* L).

**Table 2. The Amount of *Aedes aegypti* Mosquito that Perched and the Protection Power of Soursop Seed Extract (*Annona muricata* L) with a Concentration of 20% in August 6, 2018**

R	0 Hour		1st Hour		2nd Hour		3th Hour		4th Hour		5 th Hour	
	K	P	K	P	K	P	K	P	K	P	K	P
I	40	7	39	7	40	8	40	9	39	12	40	14
II	39	6	40	7	39	8	40	9	40	13	40	15
III	40	6	41	7	39	9	39	8	38	14	39	15
IV	41	10	40	8	40	8	39	9	40	14	40	16
V	40	6	40	6	39	7	40	10	39	11	39	16
Amount	200	35	200	35	197	40	198	45	196	64	198	76
Average	40	7	40	7	39	8	40	9	39	13	40	15
PP (%)	82.5		82.5		79.7		77.3		67.3		61.6	

R: repetitions

Based on the table above, the average number of mosquitoes perched on the left-hand (treatment) was the highest 15 at the V hour while the lowest was 7 at the 0 hour and the second hour. The protection of soursop seed extract has the highest concentration of 20% 82.5% at 0 hours, while the lowest is 61.6% at the 5th hour.

**Table 3 The Amount of *Aedes aegypti* Mosquito that Perched and the Protection Power of Soursop Seed Extract (*Annona muricata* L) with a Concentration of 40% In August 7, 2018**

Based table the number	R	0 Hour		1st Hour		2nd Hour		3th Hour		4th Hour		5th Hour		on the above, average of
		K	P	K	P	K	P	K	P	K	P			
I	40	2	40	2	39	3	40	3	40	5	40	6		
II	41	2	40	2	40	2	40	4	39	5	40	5		
III	40	2	41	3	41	3	39	4	40	4	40	6		
IV	41	4	40	2	40	4	40	3	40	5	40	7		
V	40	2	40	3	40	4	40	4	39	5	41	6		
Amount	202	12	201	12	200	15	199	18	198	24	201	30		
Average	40	2	40	2	40	3	40	4	40	5	40	6		
PP (%)	94.1		94.0		92.5		91.0		87.9		85.1			

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Mosquitoes perched on the left-hand (treatment) was the highest 6 heads at the 5th hour while the lowest was 2 at the 0 hours and the second hour. The protection of soursop seed extract has the highest concentration of 40% 94.1% at 0 hours, while the lowest is 85.1% at the 5th hour.

**Table 4**

**The Amount of *Aedes aegypti* Mosquito that Perched and the Protection Power of Soursop Seed Extract (*Annona muricata* L) with a Concentration of 60%**

**In August 8, 2018**

	08.30		09.30		10.30		11.30		12.30		13.30	
	K	P	K	P	K	P	K	P	K	P	K	P
I	40	1	40	2	40	1	40	3	39	4	39	4
II	40	1	39	2	39	2	40	3	40	5	40	5
III	40	1	40	1	40	1	40	2	38	4	39	4
IV	40	2	40	2	39	3	40	3	40	3	40	3
V	40	2	41	1	39	2	40	3	39	3	40	4
Amount	200	7	200	8	197	9	200	14	196	19	198	20
Average	40	1	40	2	39	2	40	3	39	4	40	4
PP (%)	96.5		96.0		95.4		93		90.3		89.9	

Based on the table above, the average number of mosquitoes perched on the left-hand (treatment) was the highest 4 tails at the 4th hour and the fifth hour while the lowest was 1 tail at the 0 hours. The protection of soursop seed extract was the highest concentration of 60.5% at 0 o'clock, while the lowest is 89.9% on 5th hour.

**Table 5**

**The Amount of *Aedes aegypti* Mosquito that Perched and the Protection Power of Soursop Seed Extract (*Annona muricata* L) with 80% Concentration in August 9, 2018**

R	08.30		09.30		10.30		11.30		12.30		13.30	
	K	P	K	P	K	P	K	P	K	P	K	P
I	39	1	40	1	40	2	40	2	39	2	40	3
II	40	1	40	2	39	1	40	3	40	3	40	3
III	40	0	41	1	39	2	39	2	38	2	39	3
IV	41	2	40	1	40	2	39	2	39	3	40	2
V	40	1	40	2	39	1	40	3	39	3	38	3
Amount	200	5	201	7	197	8	198	12	195	13	197	14
Average	40	1	40	1	39	2	40	2	39	3	39	3
PP (%)	97.5		96.5		95.9		93.9		93.3		92.9	

Based on the table above, the average number of mosquitoes perched on the left-hand (treatment) was the highest 3 heads at the 4th hour and the fifth hour while the lowest one was at the 0 o'clock and the second hour. The protection of soursop seed extract has the highest concentration of 80% 97.5% at 0 hours, while the lowest is 92.9% at the 5th hour.

**Table 6**

**The Amount of *Aedes aegypti* Mosquito that Perched and the Protection Power of Soursop Seed Extract (*Annona muricata* L) with 100% Concentration in August 10, 2018**

R	08.30		09.30		10.30		11.30		12.30		13.30	
	K	P	K	P	K	P	K	P	K	P	K	P
I	40	0	39	0	40	1	40	1	39	2	40	1
II	39	0	40	0	40	1	40	1	40	1	39	2
III	40	0	41	1	39	0	39	2	38	1	39	2
IV	40	0	40	0	40	0	39	1	39	2	40	3
V	40	0	40	0	39	1	40	2	39	1	38	1
Amount	199	0	200	1	198	3	198	7	195	7	196	9
Average	40	0	40	0	40	1	40	1	39	1	39	2
DP (%)	100.0		99.5		98.5		96.5		96.4		95.4	

Based on the table 6 above, the average number of mosquitoes perched on the left-hand (treatment) was the highest 2 tails at the V hour while the lowest was no mosquito perched at 0 o'clock in the second hour. The protection of soursop seed extract has the highest concentration of 100% 100% at 0 hours, while the lowest is 95.4% at the 5th hour.

Anova-test for Soursop Seed Extract (*Annona muricata* L) to find out whether there were significant differences in protection power between treatment groups and testing times.

**Table 7**

**Effectiveness of Soursop Seed Extract (*Annona muricata* L) as Natural Repellent on *Aedes Aegypti* Mosquitoes at 0 o'clock**

Concentrate	Mean	SD	95% CI	P value
20%	82.54	4.01	77.56 – 87.52	0.000
40%	94.07	2.14	91.14 – 96.73	
60%	96.50	1.36	94.79 – 98.20	
80%	97.51	1.72	95.36 – 99.65	
100%	100	0.00	100.0 – 100.0	

Based on the table 7 above, the average protection of soursop seed extract with a concentration of 20% at 0 o'clock is 82.54% with a standard deviation of 4.01, a concentration of 40% at 0 o'clock is 94.07% with a standard deviation of 2.14, a concentration of 60% at hour to 0 is 96.50% with a standard deviation of 1.36, a concentration of 80% at 0 o'clock is 97.51% with a standard deviation of 1.72 and a concentration of 100% at 0 o'clock is 100% with a standard deviation of 0.00.

The statistical test results obtained p-value = 0,000, p-value ≤ alpha (0.0000 ≤ 0.05). It can be concluded that there are differences in the protection power of the five concentrations used. The concentration that has a significant difference is the concentration of 20% with 40%, concentration of 20% with 60%, concentration of 20% with 80%, concentration of 20% with 100% and concentration of 40% with 100%.

**Table 8**

**Effectiveness of Soursop Seed Extract (*Annona muricata* L) as Natural Repellent on *Aedes Aegypti* Mosquitoes at the 1st hour**

Concentrat e	Mean	SD	95% CI	P value
20%	82.49	1.79	80.26 – 84.72	0.000
40%	94.03	1.32	92.39 – 95.67	
60%	95.98	1.41	94.23 – 97.73	
80%	96.51	1.38	94.79 – 98.22	
100%	99.51	1.09	98.15 – 100	

Based on the table above, the average protection of soursop seed extract with a concentration of 20% at the first hour is 82.49% with a standard deviation of 1.79, the concentration of 40% at 1st hour is 94.03% with a standard deviation of 1.32, a concentration of 60% at 1st hour to is 95.98% with a standard deviation of 1.41, the concentration of 80% at the 1st hour is 96.51% with a standard deviation of 1.33 and a concentration of 100% at the 1st hour is 99.51% with a standard deviation of 1.09.

The statistical test results obtained p-value = 0,000, p-value ≤ alpha (0.0000 ≤ 0.05). It can be concluded that there are differences in the protection power of the five concentrations used.

The concentration that has a significant difference is the concentration of 20% with 40%, concentration of 20% with 60%, concentration of 20% with 80%, concentration of 20% with 100%, concentration of 40% with 100%, concentration of 60% with 100%, concentration 80 % with 100%.

**Table 9**

**Effectiveness of Soursop Seed Extract (*Annona muricata* L) as Natural Repellent on *Aedes Aegypti* Mosquitoes in the second hour**

Concentrate	Mean	SD	95% CI	P value
20%	79.69	1.83	77.41 – 81.97	0.000
40%	92.49	1.77	90.29 – 94.69	
60%	95.41	2.17	92.70 – 98.11	
80%	95.94	1.35	94.26 – 97.63	
100%	98.48	1.38	96.77 – 100	

Based on the table 9 above, the average protection of soursop seed extract with a concentration of 20% in the 2nd hour is 79.69% with a standard deviation of 1.83, the concentration of 40% at the 2nd hour is 92.49% with a standard deviation of 1.77, a concentration of 60% at 2nd hour is 95.41% with a standard deviation of 2.17, the concentration of 80% at 2nd hour is 95.94% with a standard deviation of 1.35 and a concentration of 100% at 2nd hour is 98.48% with a standard deviation of 1.38.

The statistical test results obtained p-value = 0,000, p-value ≤ alpha (0.0000 ≤ 0.05). It can be concluded that there are differences in the protection power of the five concentrations used. The concentration that has a significant difference is the concentration of 20% with 40%, concentration of 20% with 60%, concentration of 20% with 80%, concentration of 20% with 100% and concentration of 40% with 100%.

**Table 10**

**Effectiveness of Soursop Seed Extract (*Annona muricata* L) as Natural Repellent on *Aedes Aegypti* Mosquitoes at 3rd hour**

Concentarte	Mean	SD	95% CI	P value
20%	77.28	1.60	75.29 – 79.27	0.000
40%	90.94	1.41	89.18 – 92.71	
60%	93.00	1.11	91.61 – 94.38	
80%	93.84	1.32	92.30 – 95.59	
100%	96.46	1.39	94.73 – 98.87	

Based on the table 10 above, the average protection of soursop seed extract with a concentration of 20% at the 3rd hour is 77.28% with a standard deviation of 1.60 concentration of 40% at the 3rd hour is 90.94% with a standard deviation of 1.41, a concentration of 60% at 3rd hour to III is 93.00% with a standard deviation of 1.11, a concentration of 80% at 3rd hour is 93.84% with a standard deviation of 1.32 and a concentration of 100% at 3rd hour is 96.46% with a standard deviation of 1.39.

The statistical test results obtained p-value = 0,000, p-value ≤ alpha (0.0000 ≤ 0.05). It can be concluded that there are differences in the protection power of the five concentrations used. The concentration that has a significant difference is the concentration of 20% with 40%, concentration of 20% with 60%, concentration of 20% with 80%, concentration of 20%

with 100%, concentration of 40% with 80%, concentration of 40% with 100%, and concentration 60% with 100%.

**Table 11**

**Effectiveness of Soursop Seed Extract (*Annona muricata* L) as Natural Repellent on *Aedes Aegypti* Mosquitoes at 4th hours**

Konsentrasi	Mean	SD	95% CI	P value
20%	67.33	3.40	63.10 – 71.56	0.000
40%	87.87	1.20	86.38 – 89.36	
60%	90.30	2.10	87.69 – 92.91	
80%	93.34	1.33	91.68 – 95.00	
100%	96.40	1.40	94.66 – 98.15	

Based on the table 11 above, the average protection power of soursop seed extract with a concentration of 20% at 4th hour is 67.33% with a standard deviation of 3.40 concentration of 40% at 4th hour is 87.87% with a standard deviation of 1.20, a concentration of 60% at 4th hour is 90.30% with a standard deviation of 2.10, the concentration of 80% at 4th hour is 93.34% with a standard deviation of 1.33 and the concentration of 100% at 4th hour is 96.40% with a standard deviation of 1.40. Statistical test results obtained p-value = 0.000, p-value ≤ alpha (0.0000 ≤ 0.05) it can be concluded that there are differences in the protective power of the five concentrations used. Concentrations that have significant differences are concentrations of 20% by 40%, concentrations of 20% by 60%, concentrations of 20% by 80%, concentrations of 20% by 100%, concentrations of 40% by 80%, concentrations of 40% by 100%, and concentrations 60% with 100%.

**Table 12**

**Effectiveness of Soursop Seed Extract (*Annona muricata* L) as a Natural Repellent against *Aedes Aegypti* Mosquitoes at 5th hour**

Concentrate	Mean	SD	95% CI	P value
20%	61.60	2.33	58.70 – 64.50	0.000
40%	85.07	1.77	82.86 – 87.27	
60%	89.89	1.77	87.69 – 92.09	
80%	92.88	1.19	91.39 – 94.36	
100%	95.42	2.07	92.84 – 98.00	

Based on the table 12 above, the average protection of soursop seed extract with a concentration of 20% at 5th hour is 61.60% with a standard deviation of 2.33 concentrations of 40% at 5th hour is 85.07% with a standard deviation of 1.77, a concentration of 60% at V is 89.89% with a standard deviation of 1.77, the concentration of 80% at 5th hour is 92.88% with a standard deviation of 1.19 and a concentration of 100% at 5th hour is 95.42% with a standard deviation of 2.07.

The statistical test results obtained p-value = 0,000, p-value ≤ alpha (0.0000 ≤ 0.05). It can be concluded that there are differences in the protection power of the five concentrations used. The concentration that has a significant difference is the concentration of 20% with 40%, concentration of 20% with 60%, concentration of 20% with 80%, concentration of 20% with 100%, concentration of 40% with 60%, concentration 40% with 80%, concentration 40 % with 100%, and concentration of 60% with 100%.

Repellent test of Soursop Seed extract (*Annona muricata* L) is carried out by inserting the hands of the volunteers into the test enclosure for 5 minutes alternately between the right-hand and the left-hand.



# Effectiveness of Soursop Seed Extract (*Annona muricata* L) as a Natural Repellent on *Aedes Aegypti*

Right-hand as control (without smeared Soursop seed extract (*Annona muricata* L) and left-hand as treatment (smeared with Soursop seed extract (*Annona muricata* L).

Based on the table of the number of *Aedes aegypti* mosquitoes that perch on the hand with various concentrations of soursop seed extract and the length of time of observation, there are differences in the average number of mosquitoes that perch. More right-hand mosquitoes are perched compared to the left-hand. In the left hand of the V clock there are more mosquitoes perched than at 0 o'clock.

Concentration of 20% at 0 o'clock has the highest protection power of 82.5% and lowest 61.6% at hour V. Concentration of 40% of 0 hours has the highest protection power 94.1% and lowest 85.1% at hour V. Concentration of 60% at 0 o'clock has the highest protection power of 96.5% and lowest of 89.9% at hour V. Concentration of 80% at hour 0 has the highest protection power of 97.5% and lowest of 92.9% at hour V. Concentration of 100% at hour 0 has the highest protection power of 100% lowest 95.4% on hour V.

Repellent is a compound that reacts locally or at a certain distance, which has the ability to prevent orthopods (including mosquitoes) to fly, land, or bite on the surface of human skin. The repellent mechanism of action is that mosquitoes have the ability to find prey by smelling carbon dioxide, lactic acid and other odors that come from warm and moist skin. The smell assessment was captured by chemoreceptors on the antennae of female mosquitoes. Repellent blocks lactic acid receptors so that it can damage flying ability as a result mosquitoes lose contact with the host.

Extract is an activity to withdraw soluble chemical so that it is separated from the insoluble material with liquid solvents contained in natural ingredients. The purpose of extraction is to attract and separate compounds contained in materials from plants, animals and marine biota using organic solvents. Maseration is one simple extraction method. Maseration is done by soaking the simplic powder in the fluid of the dancer. The solvent liquid entering the cell creates a difference in concentration between the solution inside and outside the cell. Low concentration solutions are inside while high concentrations are pushed out of the cell. The dancer liquid used can be water, ethanol, water-ethanol or other solvents.

Research conducted by Kardinan (2004) showed that ethanol extract from soursop seeds (*Annona muricata* L) had the highest levels of acetogenin compared to other solvents such as ethyl acetate<sup>7</sup>.

The main active compounds of soursop seeds are annonacin and squamocin which are classified as acetogenin compounds. Acetogenin compounds have quite effective toxicity against insects from the Diptera order which are cytotoxic and neurotoxic. Acetogenin compounds can inhibit the action of the NADH enzyme on myticria which causes the death of larvae, as well as contact toxins and stomach poisons in insects<sup>10</sup>.

Acetogenin compounds in essential oils of soursop seed extract function as repellents, chemical compounds proved to influence locomotor activity. The odor contained in essential oils seeps into the pores of the skin and the arena of body heat and the environment, essential oils will evaporate into the air. This odor will be detected by chemical receptors contained in mosquito antennae and transmitted to nerve impulses, responded to the brain so that mosquitoes will express themselves to avoid. Essential oil compounds have

properties that can evaporate at room temperature and evaporation is greater with the increase in temperature.

The study by Nurlaeli (2016) on the Effectiveness of Soursop Leaf Extract (*Annona muricata* L) as a Repellent of *Aedes aegypti* Mosquitoes, stated that the number of mosquitoes perched on test and control hands varied greatly depending on the level of concentration used as a repellent and the liquid and volatile<sup>11</sup>. Another study by Rizma (2017) about the Concentration Differences of Soursop Seed Extract (*Annona muricata* L) as a Repellent Against the Rejection Power of *Aedes aegypti* Mosquitoes with a concentration of 5%, 20% and 35% with 3 hours observation, stated that the longer the exposure time the more lots of mosquitoes that land<sup>12</sup>.

According to the researchers' assumption, the number of *Aedes aegypti* mosquitoes that perch depends on the concentration of the soursop seed extract, the higher the concentration the more acetogenin compounds are contained and the fewer the number of mosquitoes that perch. While the longer the exposure time, the less volatile oil content, this is due to the nature of essential oils can evaporate if there is an increase in temperature.

## IV. CONCLUSION

Based on the data the results of the study can be concluded as follows:

1. The highest number of mosquitoes perched on the skin after soursop seed extract (*Annona muricata* L) with a concentration of 20% in the fifth hour was 15, while the lowest concentration was 100% at the 0th hour when no mosquitoes alighted.
2. Concentration of Soursop Seed Extract (*Annona muricata* L) which is effective as a natural repellent for 6 hours of monitoring, namely concentrations of 80% and 100% with a protective power of 92.88% and 95.42% respectively.

## ACKNOWLEDGMENT

Thanks to Padang State University, Lecturer & all of the people support until this research finish and can publish. Special thanks to my team and my family.

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## AUTHORS PROFILE



**S. Alhamda**, Syukra Alhamda was born in Bukittinggi in 1970. He had the bachelor degree in Public Health from University of Indonesia Jakarta in 1998. His master degree in Public Health from Gadjah Mada University Jogjakarta Central Java 2004, and right now ongoing PhD program in environmental science at Padang State University.



**E. Barlian**, Eri Barlian was born in Padang in 1961. He had the bachelor degree in sport education of IKIP Padang in 1986. His master degree in environmental science at University of Indonesia (1991) and his PhD on education from Jakarta State University in 1999. Since 1999 he is a lecturer at graduate school of Padang State University and currently he is head of Environmental science doctoral program. He has various experience in assisting sport science project as well as the environmental research. His research interest in natural resource management and environmental impact. He has several national papers in national journal and papers in international journal.



**A. Razak**, Abdul Razak was born in 1971. He had the bachelor degree in Biology Mathematics and Natural Science from Andalas University Padang. His master degree in Biology Mathematics and Natural Science from Bogor Institute of Agriculture and his PhD in environmental science from Bogor Institute of Agriculture. His research interest in entomology, biophysics, personal protective equipment. He has several papers in international journal.



**M. Sari**, Mila Sari was born in Pilubang, in 1989. She had the bachelor degree in environmental health at Polytechnic of Health Padang in 2012. Her master degree in Environmental Science from Padang State University in 2016. Her research interest in Public Health. She has national paper in national journal.



**N. Herawati**, Nova Herawati was born in 1976. She had the bachelor degree in dental nursing from Gadjah Mada University 2010. Her master degree in Public Health from University of Indonesia Jakarta 2015. Her research interest in Public Health. She has national paper in national journal.