



## Utilization of Stem Bark and Leaves of *Kluwih* (*Artocarpus Altilis Park*) as an Anti-Mosquito Repellent: A Case Study of Total Mosquito Mortality (*Anopheles Sp*)

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### ABSTRACT

The purpose of this study was to demonstrate an alternative solution as an insecticide from Indonesian natural ingredients. We investigated the potential use of *Artocarpus altilis park* (known as *kluwih* plants in Indonesia) for an anti-mosquito repellent because it contains flavonoids and saponins. The use of this type of insecticide is harmless, inexpensive, and largely available in Indonesia. In the experiment, we extracted bark and leaves of *kluwih* to obtain insecticidal material to repel *Anopheles sp.* To ensure the precise extraction and insecticidal effect, the experiments were conducted four times with three repetitions. For each experiment, we used 8.4 mL with the concentrations of 20, 30, and 40% of flavonoid. The results showed that the higher the concentrations of bark and leaf extracts of *Artocarpus altilis park* the higher the mortality rate of *Anopheles sp.* is.

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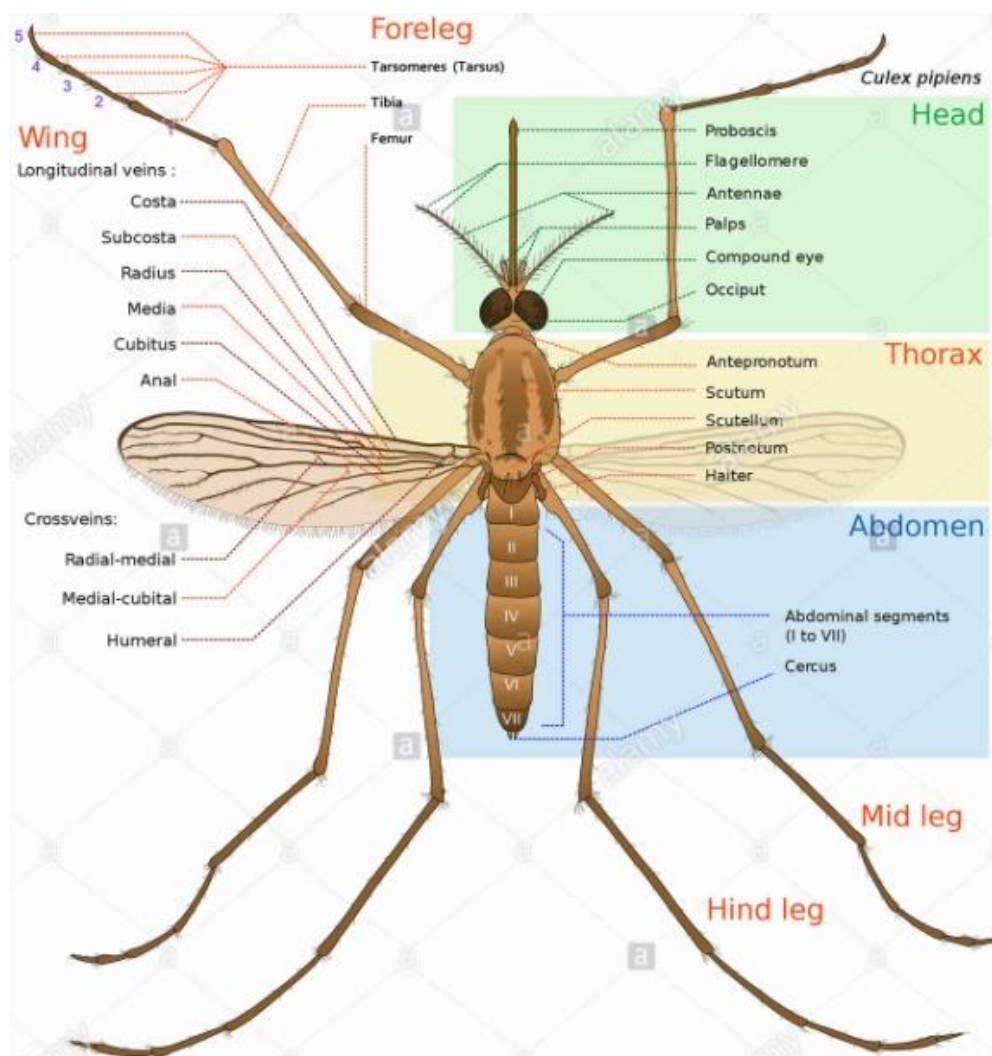
Mosquito mortality

## 1. INTRODUCTION

Diseases spread by *Anopheles sp* of mosquitoes, as the main problems for malaria and allergies, have increased over time in tropical countries, such as Indonesia, Malaysia, and Thailand. Nowadays, mosquitoes of this type are also found in sub-tropical countries and they come in summer (Husna *et al.*, 2020). The spread of the disease occurs due to various factors, such as Indonesia's geographical location, which is suitable for breeding mosquitoes (Irawati *et al.*, 2017). *Anopheles sp* mosquitoes that attack humans are females with proboscis. They

bite humans to get blood which is used for egg maturation. Egg formation requires protein obtained from the blood of humans and other mammals (Muema *et al.*, 2017).

Mosquitoes (see **Figure 1**) are considered to be a pest and have been the object of research for humans, to develop mosquito repellent products. Anti-mosquito repellents are used to prevent mosquito bites on human skin. In modern times, as today many humans create anti-mosquito products, however, these products contain chemicals that can harm human health (Murugan *et al.*, 2012).



**Figure 1.** Morphology of *Anopheles sp* of mosquitoes (Kawada, 2012)

Most mosquito repellent products sold in the market generally contain chemicals such as transflutrin, motofletrin and D-alletrin. These chemicals can cause disruption to the human body if they are used regularly (Murugan *et al.*, 2012). The effects of anti-mosquito-based chemicals interfere with the health of the human body (Raja *et al.*, 2015). The use of plants as medicine or prevention of a disease has been known since ancient times (Gnankiné & Bassolé, 2017). There are currently many types of herbal medicines and preventive agents that have been developed using both traditional and modern methods. Being a tropical country, Indonesia is rich in biodiversity which is very potential (Sriyakul *et al.*, 2019) for ingredients in the treatment of diseases or prevention. One of these ingredients that is still traditionally used is taken from kluwih plants (Kurniawan & Nurcahyani, 2019).

The *kluwih* is one of the plants commonly found in Indonesia. *Kluwih* fruit is similar to breadfruit but it has rough skin with seeds. Flowers and leafy leaves of *Kluwih* contain saponins, polyphenols, and tannins, while its stem skin contains flavonoids (Palupi *et al.*, 2019a). *Kluwih* flowers can be used as toothache medicine, while its leaves are for skin medicine. Its fruit can be consumed as food in Indonesia (Palupi *et al.*, 2019b).

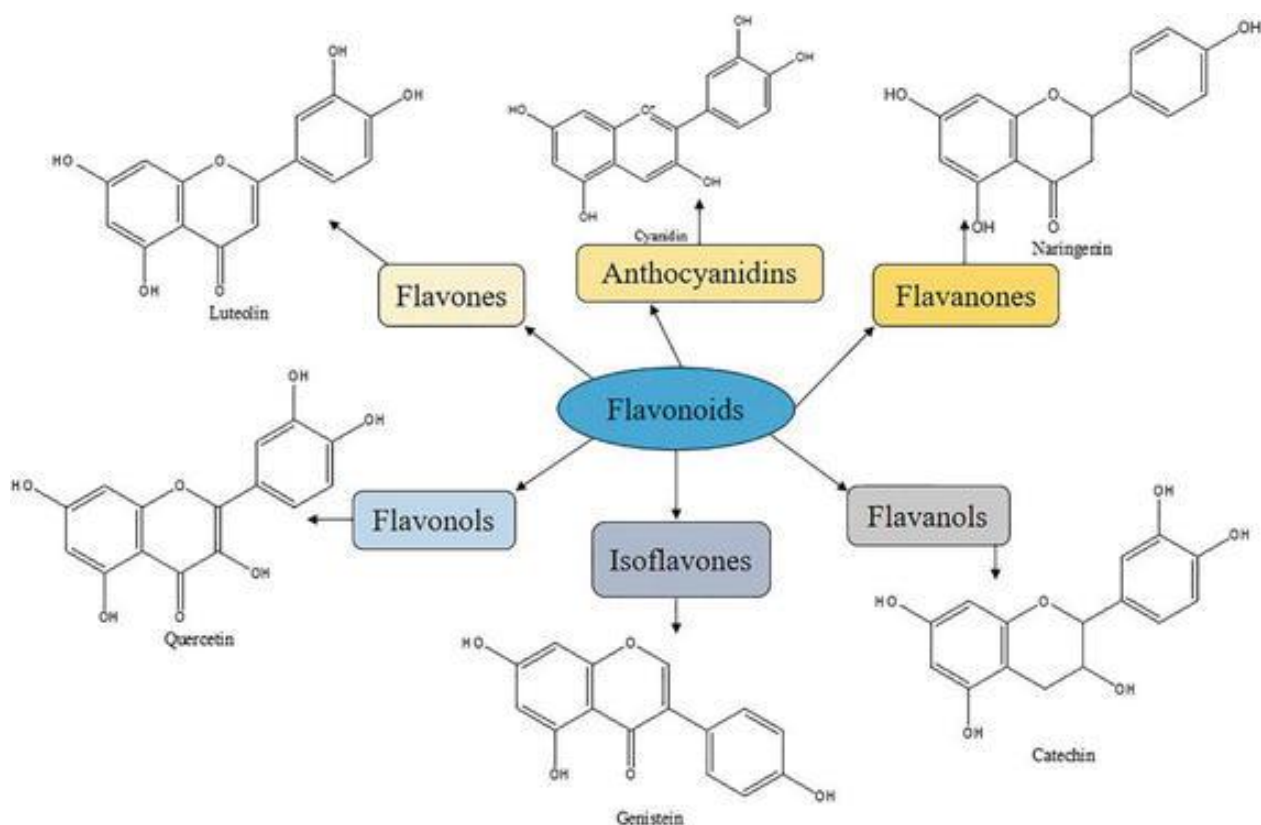
*Kluwih* flowers can be used not only for a toothache medicine, but also used for a traditional mosquito repellent by burning it. Due to its content of toxins, saponins and tannins, male *kluwih* flowers can repel or even kill insects so that they are still widely used as a mosquito repellent by rural communities (Narulita *et al.*, 2019).

From the flowers, bark, and leaves of *Kluwih* plants, researchers can obtain flavanoids which have the potential for producing a natural anti-mosquito repellent.

Active insecticides with flavonoids as active material can be used as an insect repellent, where the flavonoid compounds contained are in contact with insects (Kumar & Pandey, 2013). When insects are in contact with flavonoids, it is possible to poison their nerves (Irzaman *et al.*, 2016), stomachs (Irzaman *et al.*, 2014), through contact poisons and respiratory poisons (Parker *et al.*, 2017).

The respiratory system is an organ system that is used to breathe oxygen (O<sub>2</sub>) and release carbon dioxide (CO<sub>2</sub>) to produce energy from metabolic results (Palupi *et al.*, 2019c). The respiratory system in animals such as insects breathe using an air tube is called the trachea (Panche *et al.*, 2016). Air comes in and out of the trachea vessels through small holes on an exoskeleton called spiracles (Debboun & Strickman, 2013).

Flavonoids are one of the largest phenol compounds found in nature (Darmasetiawan *et al.*, 2002). These compounds are red, purple, and blue found in plant bodies (see **Figure 2**) (Umam & Sommanawat, 2019). A flavonoid has a carbon base framework consisting of 15 carbon atoms, where two benzene rings (C<sub>6</sub>) are bound to a propane chain (C<sub>3</sub>) to form a C<sub>6</sub>-C<sub>3</sub>-C<sub>6</sub> arrangement (Tapas *et al.*, 2008). This arrangement can produce three types of structures, namely 1,3-diarylpropan or flavonoid; 1,2-diarylpropan or isoflavonoid; and 1,1-diarylpropan or neoflavonoid (Corradini *et al.*, 2011).



**Figure 2.** Structure of the flavonoids (Tapas *et al.*, 2008)

Flavonoids are one of the strongest respiratory inhibitors. Flavonoids inhibit the growth of the three main hormones of insects, namely brain hormones (brain hormones), edicone hormones (Rahmat *et al.*, 2018a), and growth hormones (juvenile hormone) (Kurniawan *et al.*, 2018). The development of these hormones can prevent larvae from developing into adult mosquitoes. In addition, flavonoids have a way of working as a toxin that attacks the respiratory nervous system (Sangperm & Jermisittiparsert, 2019). These compounds enter through spiracles that are on the surface of the body (Rahmat *et al.*, 2018b), causing a nervous wilt and damage to spiracles so that insects cannot breathe and eventually die (Williams & Pinto, 2012).

Based on the above explanation, we were interested in conducting a study on the use of *kluwih* stems and leaf skin (*Artocarpus altilis park*) as an electric anti-mosquito repellent against the mortality of

*Anopheles sp* of mosquitoes. Information on *kluwih* flowers that are used as a mosquito repellent can only be based on the habits of people who have used *kluwih* as a mosquito repellent since a long ago (Kurniawan *et al.*, 2019). This information is supported by the research conducted by Dra. Oey KamNio (1989) who stated that the saponin content that exists (especially in male flowers) is this leafy plant that is useful as an insect repellent (especially mosquitoes) (Maretta *et al.*, 2019). The use of male *kluwih* flower as an anti-mosquito fuel in ancient times turned out to be relevant to the scientific theory based on the content of substances in it (Pedell, 2006).

This study aims to utilize the bark and leaves of *kluwih* as an electric mosquito repellent that is environmentally friendly and safe for humans (Mitprasat *et al.*, 2020). In this study *kluwih* leaves were chosen because the waste from the liquid was found more in the bark and leaves of

*kluwih* than in the flowers and other faster growing parts of the plant (Martin & Rahmat, 2017). This research was conducted to determine the effectiveness and benefits of *kluwih* bark and leaves as an electric mosquito repellent. In addition, this study aims to make a contribution to generate interests in using natural anti-mosquito repellents.

## 2. EXPERIMENTAL PROCEDURES

*Anopheles sp* of mosquito repellent was taken from a mosquito breeding/ breeding site. The breeding place is located in the area of Desa Way Muli, District of Rajabasa, Regency of South Lampung, Indonesia. To take the insects, a pipette or mosquito larvae filter was used and they were collected in a bucket. The test insect samples used were *Anopheles sp*. Larvae which were approaching the phase of becoming a pupa. The body sizes were quite large (4-6 mm) so that they were easily observed by the naked eye (Puspita et al., 2019), the thorns of the chest begin to clear, the color of the head is dark and the organs are complete.

The sample population in this study was *Anopheles sp*. The number of samples was 375 with each cage containing 25 tails. The design of this study used a Completely Randomized Design (CRD) method. We prepared 375 female *Anopheles sp* females which were divided into 5 treatment groups and used 3 repetitions.

The treatment was aquadest with a concentration of 0% (as a negative control), sidametrin solution (as a positive control), bark and leaves of *Kluwih* extract with concentrations of 20, 30, and 40% (as a treatment). The parameters measured were the utilization of bark and leaves of *Kluwih* extract on the mortality of *Anopheles sp* of mosquitoes.

We prepared 15 cages with a size of 50 cm x 50 cm x 30 cm and filled 25 *Anopheles sp* of adult mosquitoes in each cage. The mortality observation of adult mosquitoes

*Anopheles sp* treated with bark and leaves of *Kluwih* extract (*Artocarpus altilis park*) treatment was analyzed after evaporation at the 1st hour, 2nd hour, 12th and 24th hour.

This research was conducted with repetitions at three times for each treatment. The tools used in this study were a mosquito storage 15 pieces of size 50 x 50 x 50 cm, filter paper, cameras, plastic buckets, blenders, inventor incubators, filters, bottles, handles, glass jars, evaporation tools, measuring cup, mosquito filter, and distilled water. The materials used were bark and leaves of *Kluwih* extract (*Artocarpus altilis park*) 1 kg, 90% ethanol 1 liter, adult mosquito sidametrin solution *Anopheles sp*.

### 2.1 The Process of bark and leaf extract of *Kluwih* with maseration method

- a. We washed the bark and leaves of the *kluwih* in a bucket until they were clean, then we put them on top of the plate and dry.
- b. The bark and leaves of *kluwih* were mashed with a blender to form powder.
- c. Then the bark powder and *kluwih* leaves were put in each glass jar by adding 90% ethanol to 1 liter. Stirred and left  $\pm$  24 h closed.
- d. After 24 hours, the juice was filtered and taken for evaporation, for 24 h.
- e. After that it was cooled to produce 30 ml of *kluwih* bark extract and leaves.

### 2.2 Sterilization and Adhesions of bark and leaf extract of *Kluwih* on Cork

- a. Cork insect repellent containing d-aletrin 45 mg/mat was installed and heated by an electric insect repellent until the color of the cork turned white. This process took around 24 h.
- b. Corks were soaked in 70% alcohol for 2x24 h. After that, sterilization using 70% alcohol was completed. The corks



were soaked with distilled water for 12 hour.

- c. The corks were dried using an incubator for 30 min to ensure that no alcohol / distilled water is left in the sterile cork. Indicator of neutral cork by looking at the remaining dyes and not smell from d-allethrin.
- d. The corks were then soaked in extract solution according to the predetermined concentration and awaited until the extract is absorbed into the cork for approximately 3 min. The corks which contained the extract were ready for use.

### 2.3 Making Concentration for Variations in Stem Extracts and Kluwih Leaves into Solutions

In this study, the desired solution concentration was 20, 30, and 40%. Five types of treatments were used in three repetitions, and aquadest with negative controls, and positive controls was used with a sidamethrin solution. Whereas for the treatment group extract, three doses of solutions of bark extract and *kluwih* leaf (*Artocarpus altilis park*) were used: 20, 30, and 40%. Each dose was dissolved in distilled water until it reached a volume of 3 mL.

The effectiveness of bark and leaves of *Kluwih* extract (*Artocarpus altilis park*) was analyzed using a statistical analysis of variance and to determine which treatment had the best effect followed by BNT test at the level of 5%.

**Table 1.** Data on the effect of bark and leaves of *kluwih* extract (*Artocarpus altilis park*) as vegetable insecticides on *Anopheles sp* during 1 h

Concentration Insecticide (%)	Total of mosquitoes dead			Total of mosquitoes dead	The average of mosquitoes dead	Average (%)
	Repetition					
	1	2	3			
Negative control	0	0	0	0	0	0
20	2	3	2	7	2.33	9.33
30	2	4	4	10	3.33	13.33
40	3	4	6	13	4.33	17.33
Positive control	11	17	13	41	13.66	54.66
Total	18	28	25	71	25	94.66

### 3. RESULTS AND DISCUSSION

The results of the research have proven that bark and leaves of *Kluwih* extract (*Artocarpus altilis park*) with various concentrations (i.e. 20, 30, and 40%) can kill *Anopheles sp* of mosquitoes. In this study, 25 *Anopheles sp* of mosquitoes. put in a cage with two controls and three repetitions, the controls used were positive controls and negative controls.

In the positive control of *Anopheles sp* treated with 3 mL of sidamethrin solution, while the negative control of *Anopheles sp* treated with distilled water as much as 3 mL and at the concentration treatment (20, 30, and 40%) of *Anopheles sp* treated with bark and leaves of *Kluwih* extract (*Artocarpus altilis park*) according to the concentration used (20, 30, and 40%), each dose was dissolved in distilled water to reach a volume of 3 mL.

In each cage containing 25 *Anopheles sp* of mosquitoes. treated with bark and leaves of *Kluwih* extract (*Artocarpus altilis park*) with different doses and times showed different effects on the death of *Anopheles sp* of mosquitoes. The observation test was carried out every hour for 24 hours (see **Table 1**).

#### 3.1 Observations at the 1st hour

In **Table 1**, the effect of bark and leaves of *Kluwih* extract (*Artocarpus altilis park*) as vegetable insecticides on *Anopheles sp* during 1 h.

The results of the effectiveness test of bark and leaves of *Kluwih* extract (*Artocarpus altilis park*) showed various mortality differences from each concentration at different times. In the first h of the first test (see **Table 1**), the negative control treatment (3 mL) killed 0 mosquito, a concentration of 20% (3 mL) killed 2 mosquitoes, a concentration of 30% (3 mL) killed 2 mosquitoes, concentrated 40% (3 mL) killed 3 mosquitoes, and a positive control treatment (3 mL) killed 11 mosquitoes.

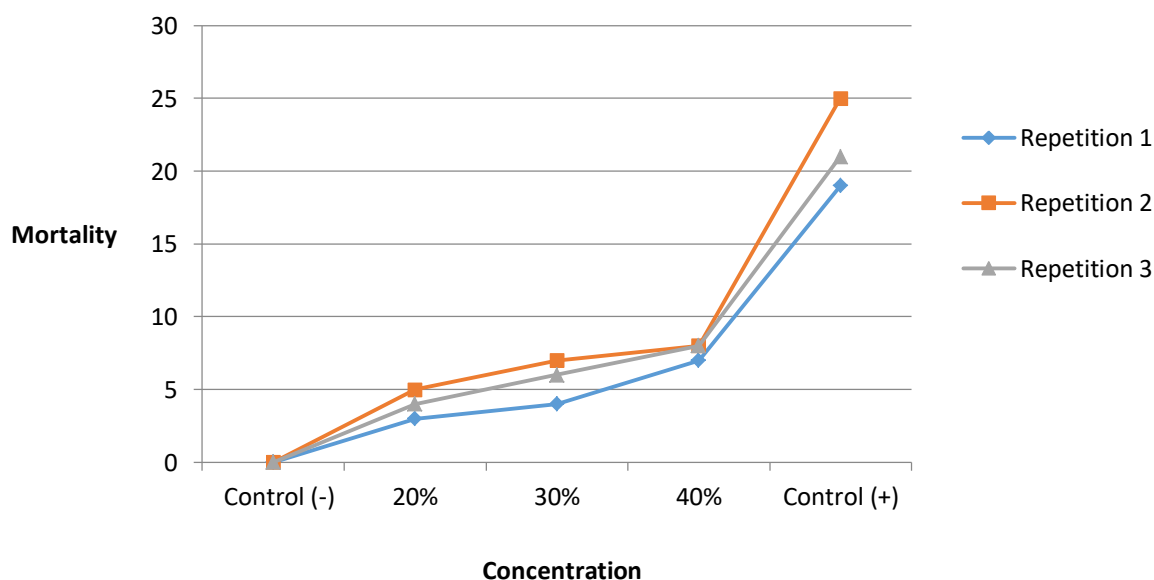
In the second test, the negative control treatment (3 mL) killed 0 mosquito, a concentration of 20% (3 mL) killed 3 mosquitoes, a concentration of 30% (3 mL) killed 4 mosquitoes, a concentration of 40% (3 mL) killed 4 mosquitoes, and positive control treatment (3 mL) killed 17 mosquitoes.

In the third test, the negative control treatment (3 mL) of 0 mosquito, a concentration of 20% (3 mL) killed 2 mosquitoes, a concentration of 30% (3 mL)

killed 4 mosquitoes, the concentration of 40% (3 mL) killed 6 mosquitoes and positive control treatment (3 mL) killed 13 mosquitoes.

The effectiveness of bark and leaves of *Kluwih* extract (*Artocarpus altilis park*) in killing *Anopheles sp* of mosquitoes. In the 1st h shown in **Figure 3**. The figure shows the difference in the number of *Anopheles sp* of mosquitoes dead in various concentrations of bark and leaves of *Kluwih* extract (*Artocarpus altilis park*) in the 1st h.

Data were analyzed at the first h, then tested by the F test. The test is shown in **Table 2**. The F test results show the effect of bark and leaves of *Kluwih* extract (*Artocarpus altilis park*) effective in increasing the mortality of *Anopheles sp* of mosquitoes, so it can be concluded that the administration of leaf extract and *kluwih* bark are effective in increasing mosquito mortality and there were concentrations of bark and leaves of *Kluwih* extract (*Artocarpus altilis park*) at a



**Figure 3.** Effect of bark and leaves of *Kluwih* extract on the death of *Anopheles sp* in the 1st hour

concentration of 40% and the most influential concentrations of positive controls compared to the effects of negative controls and by the treatment of bark and leaves of *Kluwih* extract.

This experiment has a diversity coefficient (DK = 7%). The effect of the substance was then examined using with the BNT test. The test results are shown in **Table 3**.

The results of this BNT test showed that at the test level of 5% the highest effect of (*Artocarpus altilis park*) extract at the concentration of 40% was significantly different from the negative control treatment effect and significantly different from the treatment of bark and leaves of *Kluwih* extract at other concentrations.

### 3.2 Observations for 2 h

Observations at the 2nd hour produced different experimental data on the mortality of mosquitoes at various concentrations. The results of the experimental data are presented in **Table 4**.

Based on **Table 4**, the use of bark and leaves of *Kluwih* extract (*Artocarpus altilis park*) shows different mortality differences from each concentration at different times. In the 2nd hour, on the first test, the negative control treatment (3 mL) killed 0 mosquito, a concentration of 20% (3 mL) killed 3 mosquitoes, a concentration of 30% (3 mL) killed 4 mosquitoes, a concentration of 40% (3 mL) killed 7 mosquitoes, and positive control treatment (3 mL) killed 19 mosquitoes.

**Table 2.** The result of the F test influence of the bark and leaves of *Kluwih* extract (*Artocarpus altilis park*) as vegetable insecticides on *Anopheles sp* during 1 h

Source of diversity	Degree of freedom	Number of squares	Middle squares	F Count	F Table 5%
Insecticide	4	330	825	24,2**	3,48%
Error	10	34	3,4	-	-
Total	14	364	-		

Information \*\* = Very real

**Table 3.** The results of the BNT test of the effect of bark and leaves of *Kluwih* extract on the death of *Anopheles sp* during 1 h

Insecticide Levels	Average	BNT <sub>0,05</sub> (3,34)	
Negative control	0	a	a
20 %	2,33	ab	a
30 %	3,33	abc	a
40 %	4,33	abcd	a
Positive control	13,66	abcde	e



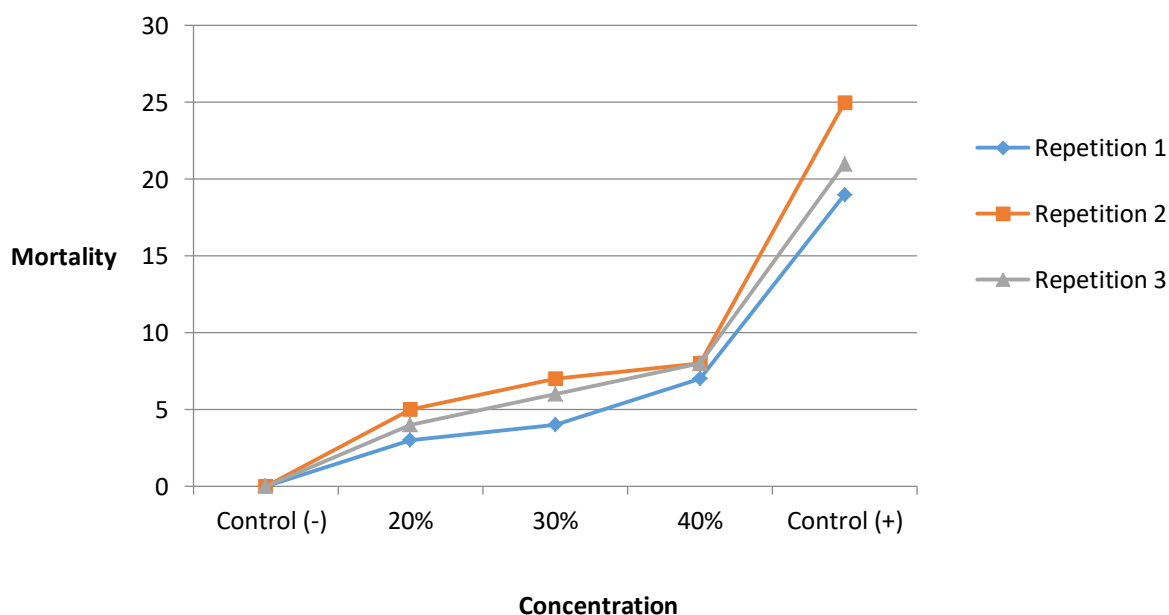
In the second test, the negative control treatment (3 mL) killed 0 mosquito, a concentration of 20% (3 mL) killed 5 mosquitoes, a concentration of 30% (3 mL) killed 7 mosquitoes, a concentration of 40% (3 mL) killed 8 mosquitoes, and positive control treatment (3 mL) killed 25 mosquitoes.

On the third test, the negative control treatment (3 mL) of 0 mosquito, a

concentration of 20% (3 mL) killed 4 mosquitoes, a concentration of 30% (3 mL) killed 6 mosquitoes, a concentration of 40% (3 mL) killed 8 mosquitoes and positive control treatment (3 mL) killed 21 mosquitoes. The table shows the effectiveness of bark extract and kluwih leaves in killing *Anopheles sp* of mosquitoes. At the 2nd hour, the result is shown in **Figure 4**.

**Table 4.** Data on the effect of bark and leaves of kluwih extract (*Artocarpus altilis park*) as vegetable insecticides on *Anopheles sp* during 2 h

Concentration Insecticide	Total of mosquitoes dead			Total of mosquitoes dead	The average of mosquitoes dead	Average %
	Repetition					
	1	2	3			
Negative control	0	0	0	0	0	0 %
20 %	3	5	4	12	4	16 %
30 %	4	7	6	17	5,66	22,66 %
40 %	7	8	8	23	7,66	30,66 %
Positive control	19	25	21	65	21,66	86,66 %
Total	33	45	39	117	39	156 %



**Figure 4.** Effect of bark and leaves of kluwih extract on the death of *Anopheles sp* in the 2nd hour

**Figure 4** shows the difference in the number of *Anopheles sp* of mosquitoes dead at various concentrations of bark and leaves of *kluwih* extract (*Artocarpus altilis park*). in the 2nd h. The data from the above analysis were tested by the F test. The test is shown in **Table 5**.

The results of the F test above show the effect of bark and leaves of *kluwih* extract is effective in increasing the mortality of *Anopheles sp* of mosquitoes, so it can be concluded that the use of bark and leaves of *kluwih* extract is effective in increasing mosquito mortality and there are concentrations of bark and leaves of *kluwih* extract which have a significant effect with the concentration of 40% as the most influential compared to the influence of negative controls and by the treatment of bark and leaves of *kluwih* extract.

This experiment has a diversity of coefficient (DK = 4.13%). The effect was then measured by the BNT test. The test results are shown in **Table 6**.

The results of this BNT test showed that at the test level of 5% the effect of bark and leaves of *kluwih* extract (*Artocarpus altilis park*) at a concentration of 40% was significantly different from the positive control effect and significantly different in the treatment of bark and *kluwih* leaf extract (*Artocarpus altilis park*) at other concentrations.

### 3.3 Observation for 12 h

Observations at the 12th hour provide different experimental data on the mortality of mosquitoes at various concentrations. The experimental data are presented in **Table 7**.

**Table 5.** The result of the F test of the influence the bark and leaves of *kluwih* extract (*Artocarpus altilis park*) as vegetable insecticides on *Anopheles sp* during 2 h

Source of diversity	Degree of freedom	Number of squares	Middle squares	F Count	F Table 5%
Insecticide	4	816,4	204,1	7,85**	3,48 %
Error	10	26	2,6	-	-
Total	14	842,4	-		

Information \*\* = Very real

**Table 6.** The results of the BNT test of the effect of bark and leaves of *kluwih* extract on the death of *Anopheles sp* during 2 h

Insecticide Levels	Average	BNT <sub>0,05</sub> (2,93)	
Negative control	0	a	a
20 %	4	ab	b
30 %	5,66	abc	b
40 %	7,66	abcd	b
Positive control	21,66	abede	e

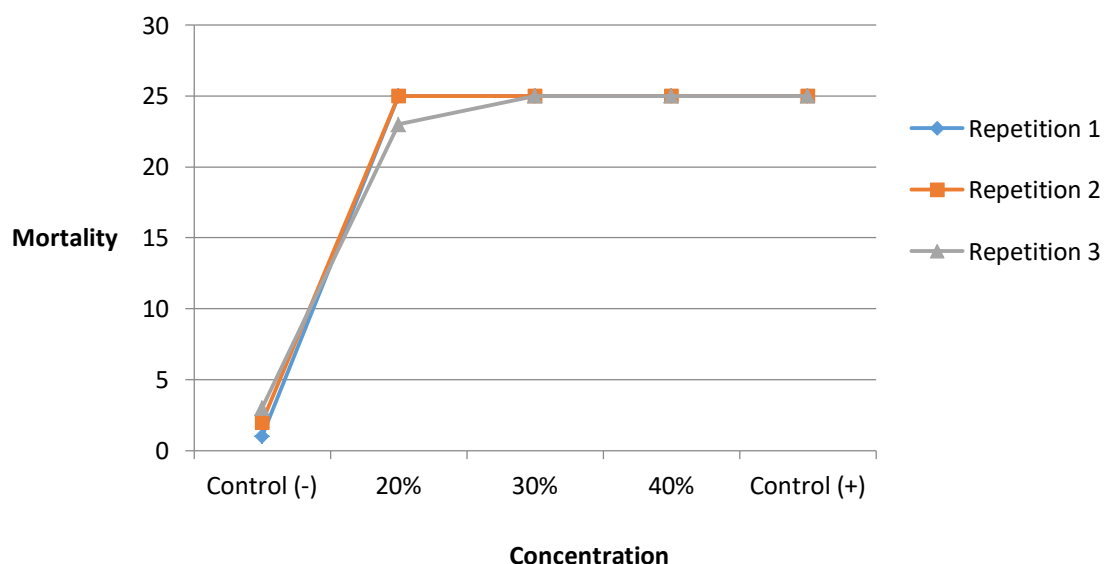
Based on the **Table 7**, the administration of bark and leaves of kluwih extract showed varying mortality rates from each concentration with different times. Within 12 h of the first test, the negative control treatment (3 mL) killed 1 mosquito, a concentration of 20% (3 mL) killed 25 mosquitoes, a concentration of 30% (3 mL) killed 25 mosquitoes, a concentration of 40% (3 mL) killed 25

mosquitoes, and a positive control treatment (3 mL) killed 25 mosquitoes.

In the second test, a negative control treatment (3 mL) killed 2 mosquitoes, a concentration of 20% (3 mL) killed 25 mosquitoes, a concentration of 30% (3 mL) killed 25 mosquitoes, a concentration of 40% (3 mL) killed 25 mosquitoes, and positive control treatment (3 mL) killed 25 mosquitoes.

**Table 7.** Data on the effect of bark and leaves of kluwih extract (*Artocarpus altilis park*) as vegetable insecticides on *Anopheles sp* during 12 h

Concentration Insecticide	Total of mosquitoes dead			Total of mosquitoes dead	The average of mosquitoes dead	Average %
	Repetition					
	1	2	3			
Negative control	1	2	2	5	1,66	6.66%
20 %	25	25	23	73	24,33	97.33%
30 %	25	25	25	75	25	100%
40 %	25	25	25	75	25	100%
Positive control	25	25	25	75	25	100%
<b>Total</b>	101	102	100	303	101	403.99%



**Figure 5.** Effect of bark and leaves of kluwih extract on the death of *Anopheles sp* in the 12th hour

In the third test, the negative control treatment (3 mL) of 2 mosquitoes, a concentration of 20% (3 mL) killed 23 mosquitoes, a concentration of 30% (3 mL) killed 25 mosquitoes, the concentration of 40% (3 mL) killed 25 mosquitoes and positive control treatment (3 mL) killed 25 mosquitoes. The effectiveness of bark and leaves of kluwih extract (*Artocarpus altilis park*) in killing *Anopheles sp* of mosquitoes at 12th h is shown in **Figure 5**.

**Figure 5** shows the difference in the number of *Anopheles sp* of mosquitoes dead at various concentrations of bark and leaves of kluwih extract (*Artocarpus altilis park*) in the 12th h.

At this 12th h, the concentration is still 40% which is the most effective concentration in killing mosquitoes compared to negative controls and other concentrations. The data from the above analysis are then tested by the F test. The test is shown in **Table 8**.

The results of the F test above show that the effect of bark and leaves of kluwih extract (*Artocarpus altilis park*) is very significant in increasing the mortality of *Anopheles sp*. and there is one treatment of insecticide concentration which has a very large effect compared to the effect of negative controls and other treatments.

This experiment has a diversity of coefficient (DK = 0.577%), and therefore further testing was carried out with the BNT test. The test results are shown in **Table 9**.

The results of this BNT test showed that at the test level of 5% the effect of bark and leaves of kluwih extract (*Artocarpus altilis park*) at the concentration of 40% was significantly different from the positive control effect and significantly different in the treatment of bark and kluwih leaf extract (*Artocarpus altilis park*) at other concentrations.

**Table 8.** The result of the F test influence of the bark and leaves of kluwih extract (*Artocarpus altilis park*) as vegetable insecticides on *Anopheles sp* during 12 h

Source of diversity	Degree of freedom	Number of squares	Middle squares	F Count	F Table 5%
Insecticide	4	1289	322,25	94,77**	3,48 %
Error	10	3,4	0,34	-	-
Total	14	1292,4	-		

Information \*\* = Very real

**Table 9.** The results of the BNT test of the effect of bark and leaves of kluwih extract on the death of *Anopheles sp* during 12 h

Insecticide Levels	Average	BNT <sub>0,05</sub> (3,34)	
Negative control	1,66	a	a
20 %	24,33	ab	b
30 %	25	abc	c
40 %	25	abcd	c
Positive control	25	abcde	c

### 3.4 Observation for 24 h

Observations at the 24th hour provide different experimental data on the mortality of mosquitoes at various concentrations. Data from the experimental results are presented in **Table 10**. Based on the data, the administration of bark and leaves of kluwih extract (*Artocarpus altilis park*) shows different mortality differences from each concentration at different times. At 24th h. on the first test, the negative control treatment (3 mL) killed 2 mosquitoes, a concentration of 20% (3 mL) killed 25 mosquitoes, a concentration of 30% (3 mL) killed 25 mosquitoes, a concentration of 40% (3 mL) killed 25 mosquitoes, and positive control treatment (3 mL) killed 25 mosquitoes.

In the second test, a negative control treatment (3 mL) killed 5 mosquitoes, a concentration of 20% (3 mL) killed 25 mosquitoes, a concentration of 30% (3 mL) killed 25 mosquitoes, a concentration of 40% (3 mL) killed 25 mosquitoes, and

positive control treatment (3 mL) killed 25 mosquitoes.

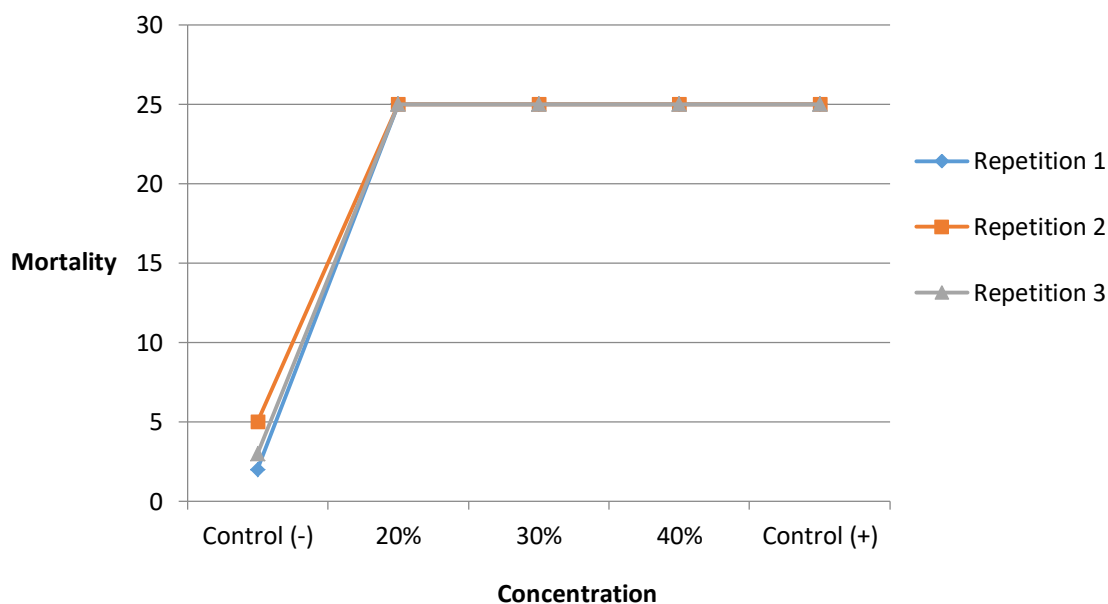
In the third test, the negative control treatment (3 mL) of 3 mosquitoes, a concentration of 20% (3 mL) killed 25 mosquitoes, a concentration of 30% (3 mL) killed 25 mosquitoes, the concentration of 40% (3 mL) killed 25 mosquitoes and positive control treatment (3 mL) killed 25 mosquitoes. The effectiveness of bark and leaves of kluwih extract in killing *Anopheles sp* of mosquitoes at 24th hour is shown in **Figure 6**.

**Figure 6** shows a difference in the number of *Anopheles sp* of mosquitoes dead at various concentrations of bark and leaves of kluwih extract (*Artocarpus altilis park*) in the 24th hour.

At 24 h, the concentration of 40% is still the most effective concentration in killing mosquitoes compared to negative controls and other concentrations. The data from the above analysis are then tested by the F test. The test is shown in **Table 11**.

**Table 10.** Data on the effect of bark and leaves of kluwih extract (*Artocarpus altilis park*) as vegetable insecticides on *Anopheles sp* during 24 h

Concentration Insecticide	Total of mosquitoes dead			Total of mosquitoes dead	The average of mosquitoes dead	Average %
	Repetition					
	1	2	3			
Negative control	2	5	3	10	3,33	13,33%
20 %	25	25	25	75	25	100%
30 %	25	25	25	75	25	100%
40 %	25	25	25	75	25	100%
Positive control	25	25	25	75	25	100%
<b>Total</b>	102	105	103	310	103,33	413,33%



**Figure 6.** Effect of bark and leaves of kluwih extract on the death of *Anopheles sp* in the 24th hour

**Table 11.** The result of the F test of the influence the bark and leaves of kluwih extract (*Artocarpus altilis park*) as vegetable insecticides on *Anopheles sp* during 24 h

Source of diversity	Degree of freedom	Number of squares	Middle squares	F Count	F Table 5%
Insecticide	4	1127	81,75	56,37**	3,48 %
Error	10	5	0,5	-	-
Total	14	1132	-		

Information \*\* = Very real

The results of the F test above indicate that the effect of bark and leaves of kluwih extract (*Artocarpus altilis park*) is very significant in increasing the mortality of *Anopheles sp*. the concentration of insecticides whose effects are very prominent compared to the effects of negative controls and other treatments.

This experiment has a diversity of coefficient (KK = 3.06%), and therefore further testing was carried out with the

BNT test. The test results are shown in **Table 12.**

The results of this BNT test showed that at the 5% test level the effect of bark and leaves of kluwih extract at a concentration of 40% was significantly different from the effect of a positive control treatment and significantly different in the treatment of bark and leaves of *kluwih* extract at other concentrations.



**Table 12.** The results of the BNT test of the effect of bark and leaves of kluwih extract on the death of *Anopheles sp* during 24 h

Insecticide Levels	Average	BNT <sub>0,05</sub> (1,27)	
Negative control	3,33	a	a
20 %	25	ab	b
30 %	25	abc	b
40 %	25	abcd	b
Positive control	25	abcde	b

Based on experimental data, the effectiveness test of bark and leaves of kluwih extract (*Artocarpus altilis park*) by electric method in concentrations of 20, 30 and 40% gave a significant effect on mortality of *Anopheles sp*. At the time of observation of 1 hour to 5 hours the average mortality of mosquitoes was below 95% with the highest mortality of mosquitoes at a concentration of 40% and the lowest at a concentration of 20% in each hour. At the time of observation of 6 h the average mortality of mosquitoes above 95% with the highest mortality of mosquitoes was at a concentration of 40% with an average of 97% and the lowest at a concentration of 20% and 30% with an average below 95%. At the time of observation of 12 h the results of the average mortality of mosquitoes were obtained which all concentrations were above 95% with the highest mortality of mosquitoes at the concentrations of 30% and 40% with a mean of 100% and the lowest at 20% with an average of 97% At the 24th hour the average mortality of all mosquitoes concentrated above 95% with the death of mosquitoes at concentrations of 20, 30, and 40% with a mean of 100%.

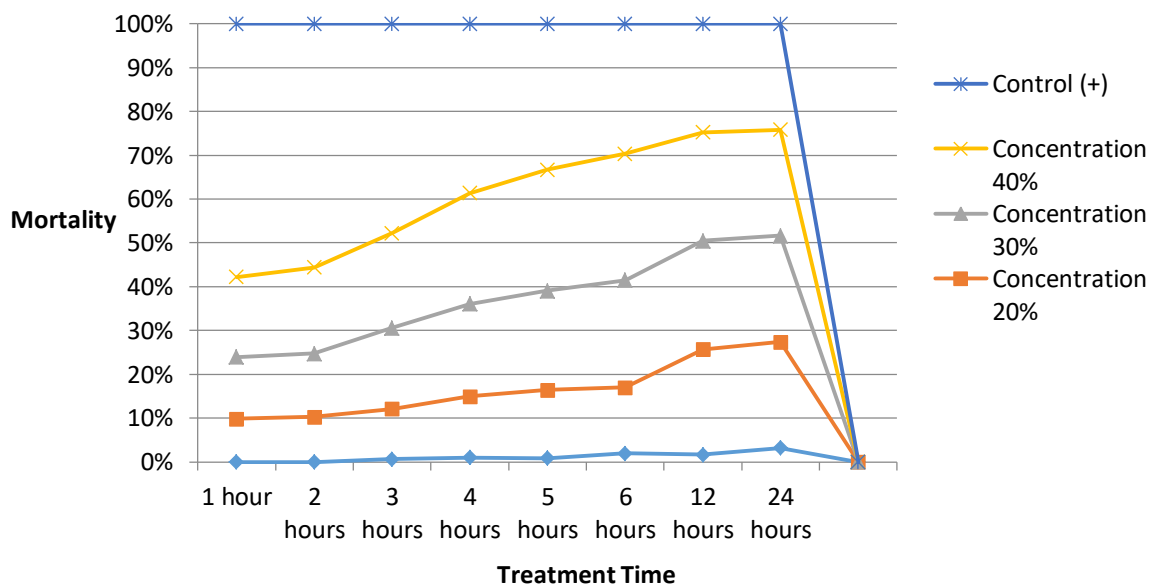
The average results of death of *Anopheles sp* of mosquitoes used bark and leaves of kluwih extract At the 1st to the 24th hour there were differences in each concentration at different times as shown in **Figure 7**.

The hypothesis in this study is that the bark and leaves of kluwih extract

(*Artocarpus altilis park*) as vegetable-based insecticides affect the mortality of *Anopheles sp* of mosquitoes. The results showed that the higher the concentration of bark and leaves of kluwih extract (*Artocarpus altilis park*) were used, the higher the mortality rate of *Anopheles sp*. This can be seen from the number of mosquito deaths in the administration of bark and leaves of kluwih extract (*Artocarpus altilis park*) with a concentration of 40%, reaching an average of 95% at the 6th, 12th, and 24th hour. The best test for insecticide effectiveness is if it is able to kill mosquitoes with a vulnerability of 95-100%.

This bark and leaves of *kluwih* extract (*Artocarpus altilis park*) aims to control the *Anopheles sp* of mosquito and suppress the spread of malaria. *Anopheles sp* of mosquito this is generally the most sensitive to contact poison insecticides in breathing, because it has a spiracle on the posterior abdomen.

The effect of various concentrations of bark and leaves of kluwih extract (*Artocarpus altilis park*) in each treatment showed a different number of deaths for each concentration. This difference was caused by the concentration of bark and leaves of *kluwih* extract (*Artocarpus altilis park*) having different flavanoid contents so the killing power is different depending on the amount of concentration of bark and leaves of kluwih extract (*Artocarpus altilis park*).



**Figure 7.** Effect of bark and leaves of kluwih extract (*Artocarpus altilis park*) on the death of *Anopheles sp* of mosquitoes during treatment

The more flavanoid content of bark and *kluwih* leaves, the mortality of *Anopheles sp.* the greater it is. Thus the treatment with a concentration of 40% is able to kill *Anopheles sp* of mosquitoes highest because it contains more flavanoid compounds.

The negative control treatment in this study found that there were mosquitoes that died at 3 h with a mean of 0.33% at 4 and 5 h, the average number of deaths was 2.66% and at 6 and 12 h the average increased to 6.66% and at the 24-hour mortality rate increased to 13.33%, this was because we moved the mosquitoes from their breeding sites to jars and then moved them again to the cage using an aspirator so that the mosquitoes became weak and eventually died.

#### 4. CONCLUSION

The conclusions are:

1. Bark and leaves of *kluwih* (*Artocarpus altilis park*) on mortality of *Anopheles sp* can be used as a botanical

insecticide because it shows the mortality rate of *Anopheles sp* which is quite high, although it cannot match the synthetic insecticide, but it should be noted that bark and leaves of *kluwih* extract (*Artocarpus altilis park*) are safer for health, animals and the environment compared to the use of synthetic insecticides.

2. The best testing of the effectiveness of botanical insecticides, if it is able to kill test insects by 95-100%. Botanical insecticides are also said to be good and selective when applied in the field, besides being effective in killing mosquitoes, these botanical insecticides are selective towards beneficial insects such as spiders, ants and other insects so that natural predators are not extinct. Whereas for concentrations that are not effective in killing mosquito larvae so that they are not applied in the community with the aim of avoiding resistance.

## 5. AUTHORS' NOTE

The authors declare that there is no conflict of interest regarding the publication of this article. The authors confirm that the data and the paper are free of plagiarism.

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