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Artocarpus altilis Extracts as Liquid Electric Mosquito Repellent Against Aedes aegypti

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Abstract

Aedes aegypti is the main vector of dengue virus carriers. One alternative to deal with these problem is by using insecticides derived from the leaves of breadfruit (Artocarous altilis) with the method of liquid electric. The research was purely experimental, with a posttest-only control group design with six variations of extract concentration of 9%, 13%, 20%, 30%, 44%, and 66% with 3 repetitions. The result of Kruskal Wallis test showed that there was a difference of the average number of Aedes aegypti mosquito mortality on various concentration of breadfruit leaf extract, that indicated by significance or probability value of 0.008 <0.05. The deadliest concentration of breadfruit leaf was 66% concentration with mosquito mortality of 83.3% from all mosquito test. The probit analysis obtained LC_{50} at concentration of 17.850% and LC_{90} at concentration of 98.348%.

Abstrak

Aedes aegypti merupakan nyamuk yang menjadi vektor utama pembawa virus dengue. Salah satu alternatif lain untuk menangani permasalahan tersebut yaitu dengan menggunakan insektisida yang berasal dari tumbuhan atau nabati, yaitu daun sukun (Artocarous altilisi) dengan metode elektrik cair. Jenis penelitian ini adalah eksperimen murni, dengan rancangan posttest only control group design dengan enam variasi konsentrasi ekstrak sebesar 9%, 13%, 20%, 30%, 44%, dan 66% dengan masing-masing 3 kali pengulangan. Hasil uji Kruskal Wallis menunjukkan bahwa ada perbedaan rata-rata jumlah kematian nyamuk Aedes aegypti pada berbagai konsentrasi ekstrak daun sukun, ditunjukkan dengan nilai signifikansi atau probabilitus adalah 0,008 < 0,05. Konsentrasi ekstrak daun sukun yang paling banyak mematikan nyamuk yaitu konsentrasi 66% dengan kematian nyamuk 83,3% dari keseluruhan nyamuk. Uji analisis probit didapatkan LC_{so}pada konsentrasi 17.850% dan LC_{so}pada konsentrasi 98.348%.

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INTRODUCTION

The mosquito that became the main vector of Dengue Hemorrhagic Fever (DHF) is from the genus Aedes, especially Aedes aegypti or Aedes albopictus. Aedes aegypti mosquitoes spread dengue virus. This is in line with the increasing mobility and population density. Therefore, DHF is still a major public health problem in Indonesia (DG of Disease Control & Environmental Health, 2011).

Semarang City Health Office (2016) noted that DHF is the second leading cause of death, which reached 16%. This is because Semarang city is a dengue endemic area. The death rate (CFR) in Semarang City in 2016 had increased from 1.2 in 2015 to 5.12 in 2016. In the last 10 years, IR of DHF in Semarang City has always been much higher than IR of DHF in Central Java and IR of DHF in National.

Currently, the vaccine for dengue is still in development stage, further research is needed in the laboratory, so for the community, there is no medicine and vaccine to overcome the dengue virus infection, therefore, breaking the transmission chain that is by controlling the DHF vector which is considered the most important at this time. Several methods of vector control are performed to break the chain of transmission of DHF diseases, namely, chemically by using synthetic insecticide that is used to control mosquitoes and larvacides used to control larvae, biologically by using natural enemies such as predators, bacteria, the predators are used to control mosquito larvae, environmental management such as managing or eliminating mosquito breeding habitat known for 3M plus or PSN movement and integrated vector control (DG of Disease Control & Environmental Health, 2011). Based on research conducted by Santi (2015) there are several factors that have a relationship with the presence of Aedes aegypti larvae in Semarang city, one of which is the bad implementation of PSN, it is also consistent with studies conducted by Alma (2014). In addition, Septiano et al (2014) also stated that there is a connection between physical, chemical, and biological practices of PSN with the presence of Aedes aegypti larvae. Poor home sanitation also has a connection with the presence of Aedes aegypti mosquitoes (Sukowinarsih et al, 2010). Among the various types of vector control efforts, the effort by using synthetic insecticide is still become the main choice, because of the factor namely ease of use, easily obtained, and immediate results can be seen by the community.

Various kinds of household insecticides have been widely sold in the market. The community has been using it in everyday necessities to repel mosquitoes. The results of survey showed that the type of household insecticide used by the community to control the vector of DHF is packaged in various formulations, namely liquid, coil, aerosol, vaporizer, lotion, and burn paper. In addition to formulations, the active ingredients and concentrations used are also various. Almost all household insecticides use active ingredients from synthetic pyrethroid groups that include d-allethrin, d-phenothrin, cyphenothrin, permethrin, imiprothrin, transfluthrin, prallethrin, metofluthrin, cypermethrin, cyfluthrin and deltamethrin, which in continuous use can cause resistance in insects and may have a cumulative effect on the human respiratory organs(Joharina et al, 2011; Marcombe et al, 2012; Syafie et al, 2012).

Continuous use of household insecticides will cause resistance to vectors. The results show that most of the *Aedes aegypti* DHF vectors in some areas of Central Java and the Special Territory of Yogyakarta have been resistant to 0.8% Malathion that applied by fogging, 0.1% bendiocarb that applied to household insecticides with aerosol formulations, 0.05% lambda-cyhalothrin, 0.75% permethrin, 0.05% deltamethrin, and 0.5% etofenprox that mostly applied to other household insecticide types (DG of Disease Control & Environmental Health, 2011).

Based on these problems, it is necessary to find other alternatives in efforts to control the DHF vector which is by using insecticides derived from plants or vegetable insecticides. Plant-based insecticide is natural substances derived from plants that have a secondary metabolite group containing thousands of bioactive compounds such as alkaloids, terpenoids, phenolics, and other secondary substances. The important thing to note, there is also the emergence of resistance from various species of mosquitoes which become the vector of a disease caused by the frequent use of chemical insecticides (Marcombe et al., 2012; Shafie et al., 2012). Another reason to use a plant-based insecticide in dengue vector control is that the vegetable insecticides generally show a higher level of safety because the molecules are easily broken down into harmless compounds against the environment.

Indonesia has a very high source of biological diversity, including plant species that have active ingredients to be developed as vegetable insecticides, compounds contained in plants and suspected to function as insecticides, among them cyanide, saponin, tannin, flavonoids, alkaloids, essential oils and steroids. One of the plants that has the potential as a bio-insecticide is breadfruit plants, because breadfruit plants contain insecticidal compounds such as alkaloids, flavonoids, phenols, saponins, and tannins that have an impact on insects. In addition, breadfruit plant is a plant that is easy to get because it spread in Indonesia. Based on the results of phytochemical test, the methanol extract of dry breadfruit leaves (*Artocarpus altilis*) that contain al-

kaloids, flavonoids, tannins, phenols, and saponins (Maharani, et al). The breadfruit leaf extract has a tannin content of 593.596 mg TAE/g extract and flavonoid levels of 1503.763 QE extract. Selection of breadfruit leaves as an anti mosquito-making material because the leaves of breadfruit easily found in the community and has not been widely used by the community.

The formulation of insect repellent that will be used in this research is liquid electric formulation because the liquid electric formulation is more practically used, it can be refilled, leaves no trash like vaporizer electric mosquito coil, leaves no ashes like mosquito coil, and does not cause smelly smoke.

Based on these various reasons, the research aimed to find out the power of killing of breadfruit leaf extract (*Artocarpus altilis*) as anti-mosquito against *Aedes aegypti* mosquito mortality in liquid electric formulation.

METHOD

This research is a true experimental research, with post-test design with control group (post-test only control group design), which is a measurement consisting of 2 groups of samples that is control group and treatment group. The sample control group was given an aquadest (negative control) and a transfluthrin-based mosquito repellent (positive control). The treatment group, the samples were given a breadfruit leaf extract with a concentration of 9% (treatment 1), 13% concentration (treatment 2), 20% concentration (treatment 3), 30% concentration (treatment 4), 44% concentration (treatment 5), and 66% concentration (treatment 6).

The samples in this study were 20 Aedes aegypti mosquitoes aged 2-5 days. In this study for each group was repeated 3 times. The treatment group was divided into 6 groups with various treatment concentrations. The total sample required for the study was 500. The amount was obtained from 20 mosquitoes x 8 (2 control groups and 6 treatment groups) x 3 repetitions. In addition, there were needed 20 mosquitoes to determine the presence or absence of contamination in the glass chamber.

The tools needed in making breadfruit leaf extracts were knives, scales, aerated trays, blenders, filter cloth and rotary evaporators. The material for making breadfruit leaf extract as the main ingredient and ethanol as a solvent. Tools required during the test were thermohygrometer, stopwatch, aspirator, glass chamber, liquid electric mosquito repellent container, stationery, and recording sheet. Materials needed during the test were *Aedes aegypti* mosquitoes, sugar water, liquid electric from transputrin, aquadest, paper cup, rubber, and cotton.

Breadfruit leaf extraction was performed by

maceration method with ethanol solvent. Steps in extraction are as follows: clean the leaves of breadfruit with water and separate the leaves from the stalks, dry the leaves of breadfruit by way of aerated, weigh the leaves in dry state as much as 1 kg, cut the small leaves, and blend until smooth, after it has been blended, and it has become macerated powder by using 96% ethanol solvent into a tightly closed inert container at room temperature for 2 days, then filter it, and the pulp is macerated twice by using ethanol solvent each for 1 day, the maserate is merged then it is fed into the rotary evaporator at 60°c until the ethanol solvent evaporates and the desired extract is obtained (Mukhriani, 2014).

The test was carried out through the following steps: Record the temperature and humidity of the room, insert 20 mosquitoes to the glass chamber to determine the sterility, heat the electric liquid vapor mosquito repellent in the draft glass chamber for 4 hours and then movie into the test glass chamber for 3 minutes, transfer the electric vapor liquid from the glass chamber into the draft glass chamber (the mosquito repellent is still heated), release 20 mosquitoes into the test glass chamber, observe for 20 minutes and count or record the unconscious/ dead mosquitoes within each 30 second period and minutes of 1, 2, 3, 4, 5, 6, 7, 8, 10, 15, and 20. Move all the mosquitoes into paper cup by using aspirator, then store or hold for 24 hours, and calculate and record the amount of unconscious/dead mosquitoes. The percentage of mosquito mortality was calculated by the number of dead mosquitoes + the number of mosquitoes divided by the number of mosquitoes in each test group x 100%.

The data were analyzed statistically by probit test, normality data test by using Saphiro-wilk, homogenity test of variation with Levene test, Kruskal-Wallis test, and continued with Post-Hoc analysis with Mann-Whitney test.

RESULTS AND DISCUSSION

The average percentage of mortality (%) of *Aedes aegypti* mosquitoes on various concertation of breadfruit leaf extract can be seen in Figure 1.

Based on the graph in Figure 1, there was an increase in the concentration of breadfruit leaf extract followed by the increase of mosquito mortality until 66% concentration which can cause the mosquito mortality of 83,3%.

Based on the calculation of Lethal Concentration probit on the insecticide of breadfruit leaf extract ($Artocarpus\ altilis$) with liquid electric method obtained result that LC₅₀ of 17.850 % and LC₉₀ of 98.348 %.

Interpretation of Kruskal-Wallis test results with p = 0.008 which means that there was a diffe-

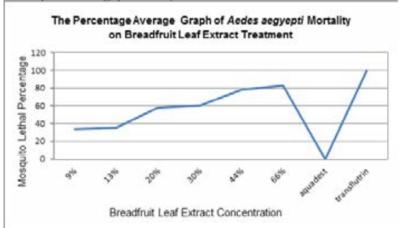


Figure 1. The Graph of Aedes aegyepti Mortality on Breadfruit Leaf Extract Treatment.

rence in average number of mosquito mortality due to significance or p < 0.05.

The result of post hoc test showed p-value <0.05 which means there was a difference of mosquito mortality rate at every concentration. The concentration values did not differ significantly namely 9% concentration with concentrations of 13%, 20%, 30%, and 44%. Concentration of 13% with concentrations of 20%, 30%, and 44%. Concentration of 20% with concentrations of 30% and 44%. Concentration of 30% with concentrations of 44% and 66%. 44% concentration with 66% concentration.

Observations on research conducted at Entomology Laboratory P2B2 Banjarnegara showed the mortality of Aedes aegypti mosquitoes. The mortality of mosquitoes due to contact with liquid electric mosquito killer which has an insecticide from breadfruit leaf extract, which saponin substance that has bitter taste, and sharp and can cause stomach irritation when it is eaten, besides that saponin also able to bind sterols, sterol is precursor hormone, so that if the amount of free sterol decreases, it will interfere the process of skin replacement, and absorbed which will cause blood hemolysis. It causes respiratory enzymes to be inhibited, and lead to death (Widawati et al, 2013). Alkaloids are active components that work in the nerves, but it also can cause indigestion because alkaloids can act as poison (Syamsul, 2014). Flavonoid is one of the content found in the leaves of breadfruit (*Artocarpus altilis*) which serves as respiratory toxins or respiratory inhibitors (Nikmah, 2016).

From the results of observations during the 24 hours in the study, found that the average% of mosquito mortality at 9% concentration of 33.3%, 13% concentration of 35%, 20% concentration of 58.3%, at 30% concentration of 60%, at 44% concentration of 78.3%, at 66% concentration of 83.3%, in negative control of 0%, and positive control of 100%. The greater the concentration of breadfruit leaf extract, the more mosquitoes die. This is possible because the greater the concentration of breadfruit leaf extract, the greater the active substance contained in it. Based on the calculation of Lethal Concentration probit on the insecticide of breadfruit leaf extract (Artocarpus altilis) with liquid electric method obtained result that LC50 of 17.850 % and LC₉₀ of 98.348 %. There was a difference of the number of active substances in each concentration of extract used, thus causing a different amount of active substance on each mosquito when the research was conducted.

From the observation, Aedes aegypti mosquitoes which have been treated with breadfruit leaf extract by liquid electric method would experience a change of behavior, where the movement that previously active would be slow and difficult to move. Aedes aegypti mosquitoes were said to knockdown

Table 1. Results of Probit Test

Probability	95% Confidence L	95% Confidence Limits for concentration				
	Estimate	Lower Bound	Upper Bound			
Probit 0.5	17.850	11.211	24.512			
0.9	98.348	57.302	400.477			

Table 2. Results of Kruskal-WallisTest

Kruskal Wallis	Mortality Number	-
Chi-square	19.182	
Df	7	
Asymp. Sig.	0.008	

when falling, floating in supine state, with the movement progressively slower. Aedes aegypti mosquitoes were said to die if there is no movement after the treatment. To verify whether the observed mosquito has died or not after being left 24 hours and after being exposed for 20 minutes, the researchers put the mosquito body on paper and blew it occasionally. When they touched mosquitoes and mosquitoes still moved, it can be said that the mosquito just fainted.

Aedes aegypti mosquito mortality was present in all treatment groups, except for negative controls. This proves that the mortality in the treatment group was caused by breadfruit leaf extract in liquid electric form, not because of environmental factors (temperature, humidity, etc.). The mortality of Aedes aegypti mosquitoes was caused by poisoning when the liquid electric apparatus was heated by using a heater. At the time of liquid electric from the extract of breadfruit leaves heated the liquid electric mosquito killer would release the secondary metabolite content of flavonoids.

CONCLUSION

Breadfruit leaf extract has the potential to kill Aedes aegypti mosquitoes. The deadliest concentration of breadfruit leaves extract was 66% concentration with mosquito mortality of 83.3% from whole mosquito test. Based on the calculation of Lethal Concentration probit on the insecticide of breadfruit leaf extract (Artocarpus altilis) with liquid electric method obtained result that LC $_{50}$ of 17.850% and LC $_{90}$ of 98.348%. Suggestion for further research is to prepare mosquitoes whose numbers exceed the required samples.

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