Environmental and Toxicology Management (2023), **3.2**, 17–23

RESEARCH ARTICLE

Comparison of variaton in extract concentration onion leaf (*Allium fistulosum L.*) to mortality larvae *Aedes aegypti* mosquito instar III

Siska Wulandari[1], Yauwan Tobing Lukiyono[1*], Ary Andini[1], Ersalina Nidianti[1]

[1]DIV-Health Analyst, Faculty of Health, Universitas Nahdlatul Ulama Surabaya, Indonesia

[*]**Corresponding author: tobing@unusa.ac.id**

Article history:
Received : 2023/05/17
Revised : 2023/06/09
Accepted : 2023/08/15
Available Online : 2023/08/31
DOI : 10.33086/etm.v3i2.6389

Abstract

Dengue Hemorrhagic Fever (DHF) is a disease caused by the Dengue virus which is transmitted through the *Aedes aegypti* mosquito. DHF is caused by the Dengue virus whose main vector is the *Aedes aegypti* mosquito and the secondary vector is the *Aedes albopictus* mosquito. Eradicating *Aedes aegypti* and *Aedes albopictus* is one way that can be done to prevent the spread of the Dengue virus. Control of the dengue vector can be carried out on larvae and adult mosquitoes with the aim of breaking the chain of transmission. One way that can be done is by using a natural larvicide which utilizes the *Allium fistulosum* plant which contains secondary metabolite compounds including phenolics, flavonoids and tannins which can kill larvae. This study aims to determine the comparison of variations in the concentration of *Allium fistulosum* leaf extract on the mortality of *Aedes aegypti* instar III. The type of research used in this research was experimental with a completely randomized design (RAL). The potential level of the extract is reviewed from the Lethal Concentration (LC50) and Lethal Time (LT50) values. The data obtained were analyzed using a probit regression test or probit analysis to determine the potential of *Allium fistulosum* extract as an alternative larvicide for third instar *Aedes aegypti* larvae and stated as Lethal Concentration (LC50) and Lethal Time (LT50) with a confidence level of 95.0%. The results of the research show that *Allium fistulosum* leaf extract has the potential as an alternative larvicide for the death of third instar *Aedes aegypti* larvae as proven by a p-value of 0.000 (< 0.05) with a value of LC50-6 hours is 52.850%, LC50-8 hours is 42.674%, LC50-24 hours is 30.900%, LT50-20% is 37.674 hours, LT50-30% is 24.423 hours and LT50-40% is 14.954 hours.

Keywords: Dengue Hemorrhagic Fever (DHF), Larvacide, *Aedes aegypti*, *Allium fistulosum*

INTRODUCTION

Dengue Hemorrhagic Fever (DHF) is a disease Which caused by the Dengue virus which is transmitted through the *Aedes aegypti* mosquito. Dengue fever caused by the Dengue virus whose main vector is the *Aedes aegypti* mosquito and vector secondary that is mosquito *Aedes albopictus*.

DHF is an acute disease that has clinical symptoms, namely bleeding which can cause shock and can lead to death (Prasetyani et al., 2015).

Until now, the *Aedes aegypti* mosquito is still a problem of health in Indonesia. Based on data which obtained from the Ministry of Health of the Republic of Indonesia, Kemenkes, 2020, disease spread figures DHF is still high in Indonesia. In 2019, the number of cases recorded as many as 112,954 cases and in 2020 the number of cases in January until July reach 71,633 case, increase more from 50% (63.4%) compared to previous year. This number is expected to continue increase until end year 2020 (Dewi et al., 2022).

Eradication on stage larvae considered more effective used compared to vector hich already develop breed become mosquito mature. Larvicide is famous in Indonesia for controlling larvae *Aedes aegypti* is a 1% abate which if used in the long term encourages moret *Aedes aegypti* population grows, the more resistance it will give rise to chemical pollution in environment. As a result of the negative impacts caused by chemical larvicides, another method that can be used is using alternative larvicides from plants. To overcome that problem, alternative compounds are needed that are environmentally friendly and safe for non-organisms targets include humans which could be developed as a larvicide against *Aedes aegypti* (Ravaomanarivo et al., 2014).

In Indonesia, there are several types of plants that contain compounds secondary metabolites that can be used for natural larvicides such as onions leaves that are easy to obtain and have the potential as a larvicide that is relatively safe and easily decomposed. Spring onion is a type plant which known as plant which already long known and cultivatedin Indonesia. In spring onions there is the compound isoquercitrin and quercitrin is a subclass of flavonoids, tannins, thiosulphinate (allicin), and phenolics acid which can have effect larvicide (Dinata et al., 2017).

Based on the background of the problem, research was carried out which aim for know comparison of concentration variations in *Allium fistulosum L.* extract on the mortality of *Aedes aegypti* instar III, so that it can be used as an effort to control the *Aedes aegypti* larvae vector that causes dengue fever use natural larvicide.

MATERIALS AND METHODS

Materials and Tools

The materials used in this study were *Aedes aegypti* instar III larvae, fresh dried spring onions (A*llium fistulosum L.*), 96% ethanol, distilled water, 1% Abate and labels. The tools used in this study were a blender, filter paper, a 1.5 meter container, a funnel, a beaker glass, a 10 ml measuring pipette, a measuring cup, a test cup, a test cup cover, an evaporator, and aluminum foil.

Methods

The type of study is experimental design with post test-only and control group design with giving variations treatment in the concentration of *Allium fistulosum L.* extract on the mortality of *Aedes aegypti* larvae. Control group in this research consists from control positive use Abate 1% and control negative use aquadest. On this research, the experimental group was given *Allium fistulosum L.* extract to *Aedes aegypti* larvae instar III with a number of variation concentrations that 20%, 30% and 40%. Data collection techniques are divided into pre-analytic, analytical, and post-analytic stages. The pre-analytical stage is making *Allium fistulosum L.* extract by dried leaf then smoothed use blender then filtered add ethanol 96% with comparison 1:2 (g:mL) filtrate which generated Then deposited during 24 hours. After settle down, filtrate part on deposited using a rotary vacuum evaporator until obtained extract leaf which thick.

The analytical stage is making different concentration of *Allium fistulosum L.* extract that 20%, 30% and 40%. Then give treatment or test effectiveness to *Aedes aegypti* larvae instar III with method prepare 25 fruit glass plastic. Each 5 fruit glass plastic given label control positive, control negative, concentration 20%, concentration 30%, and concentration 40%. Plastic glass with positive control

label filled with 100 mL of Abate 1%. Plastic glass with label control negative filled with 100 mL aquadest. Plastic glass with label concentration 20% filled with 100 mL extract 20%. A plastic cup labeled with a concentration of 30% is filled with 100 mL extract 30%. Plastic cup with 40% concentration label filled with 100 mL 40% extract. Then, each plastic cup is filled with 25 tail *Aedes aegypti* larvae instar III. post-analytic stage is that the researcher makes observations and counts the number of larvae that died during 6, 8, and 24 hours.

Data Analysis Procedure

This study use bivariate analysis which is analysis a single statistic is used to determine the relationship between two variable. In this research, bivariate analysis was used to find out the concentration that can kill 50% of third instar *Aedes aegypti* larvae (LC50) and the time required to kill 50% of *Aedes aegypti* larvae instar III (LT50) using *Allium fistulosum L.* extract. Data obtained from LC50 and LT50 observations larvae *Aedes aegypti*. Instar III then data analysis was carried out using program application Statistics Products and Service Solutions (SPSS).

RESULTS AND DISCUSSION

Results

Research has been carried out with the results of data obtained from treatment of *Allium fistulosum L.* extract with concentrations of 20%, 30%, 40%, using 2 control groups, namely negative control using Aquadest and positive control using Abate. The number of larval deaths was calculated for 24 hours, observations and data recording were carried out for 6 hours, 8 hours and 24 hours. This study aims to determine the effect of variations in the concentration of *Allium fistulosum L.* extract on the death of third instar *Aedes aegypti* mosquito larvae.

Table 1. Research result

			Number of Death							
Treatment	Time	Amount of *Aedes aegypti*	Replication					Total Dead Larvae	Larval Mortality	Average
			1	2	3	4	5			
Negative control	6 hours	125	0	0	0	0	0	0	0%	0
Aquadest	8 hours	125	0	0	0	0	0	0	0%	0
	24 hours	125	0	0	0	0	0	0	0%	0
Posotive control	6 hours	125	12	12	13	13	11	61	48.8%	12.2
Abate 1%	8 hours	125	17	17	18	17	18	87	69.6%	17.4
	24 hours	125	23	24	24	24	24	119	95.2%	23.8
Concentration 20%	6 hours	125	2	3	3	2	3	13	10.4%	2.6
	8 hours	125	4	5	4	5	5	23	18.4%	4.6
	24 hours	125	8	9	9	9	9	44	35.2%	8.8
Concentration 30%	6 hours	125	4	5	5	4	5	23	18.4%	4.6
	8 hours	125	7	7	7	8	7	36	28.8%	7.2
	24 hours	125	12	12	12	12	13	61	48.8%	12.2
Concentration 40%	6 hours	125	7	7	9	9	7	39	31.2%	7.8
	8 hours	125	11	12	11	12	12	58	46.4%	11.6
	24 hours	125	16	16	16	15	15	78	62.4%	15.6

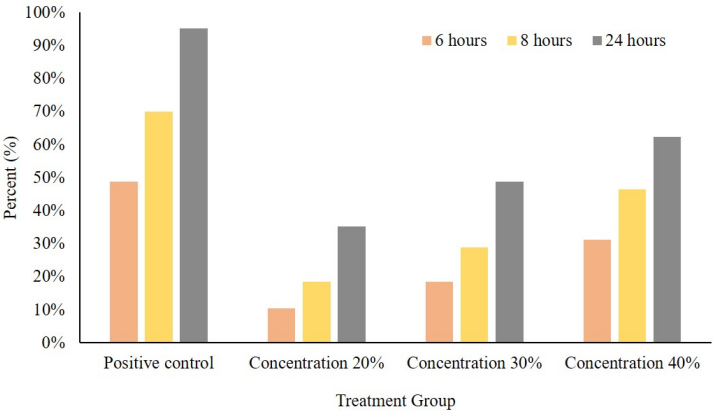

Figure 1. Comparison of the different concentration of *Allium fistulosum L.* extract with positive control

Mortality data due to the effect of giving *Allium fistulosum L.* extract to negative control, positive control, concentrations of 20%, 30% and 40% on 25 third instar *Aedes aegypti* larvae with observations for 6 hours, 8 hours and 24 hours. In each treatment group, repetition was carried out 5 times. So the results obtained from 6 hour observations were 0% in control (-), 48.8% in control (+), 10.4% in 20% concentration, 18.4 in 30% concentration, and 31.2% in concentration 40%. At 8 hour observation, namely 0% in control (-), 69.6% in control (+), 18.4% at 20% concentration, 28.8% at 30% concentration, and 46.4% at 40% concentration. At 24 hour observation it was 0% in control (-), 95.2% in control (+), 35.2% at 20% concentration, 48.8% at 30% concentration, and 62.4% at 40% concentration.

Table 2. LC50 Results of *Allium fistulosum L.* extract

Treatment	Concentration	%Death	LC50	Information
T6 hours	Positive control	48.8%	52.850	Toxic
	Concentration 20%	10.4%		
	Concentration 30%	18.4%		
	Concentration 40%	31.2%		
8 hours	Positive control	69.6%	42.678	Toxic
	Concentration 20%	18.4%		
	Concentration 30%	28.8%		
	Concentration 40%	46.4%		
24 hours	Positive control	95.2%	30.900	Very toxic
	Concentration 20%	35.2%		
	Concentration 30%	48.8%		
	Concentration 40%	62.4%		

Based on the results at 6 hours the LC50 result was 52.850 in the toxic category, while at 8 hours the LC50 result was 42.678 in the toxic category and in the 24 hours treatment the LC50 result was 30.900 in the very toxic category.

Table 3. LT50 probit analysis result

Treatment	Time	%Death	LT50	Information
Negative control	6 hours	0%		
	8 hours	0%		
	24 hours	0%		
Positive control	6 hours	48.8%	4.467	Very effective
	8 hours	69.6%		
	24 hours	95.2%		
Concentration 20%	6 hours	10.4%	37.674	Ineffective
	8 hours	18.4%		
	24 hours	35.2%		
Concentration 30%	6 hours	18.4%	24.423	Effective
	8 hours	28.8%		
	24 hours	48.8%		
Concentration 40%	6 hours	31.2%	14.954	Very effective
	8 hours	46.4%		
	24 hours	62.4%		

Based on the results at a 20% concentration the LT50 result is 37.674 hours in the ineffective category, while at a 30% concentration the LT50 result is 24.432 hours in the effective category and at a 40% concentration the LT50 result is 14.954 hours in the very effective category.

Table 4. Normality test result

Treatment	%Death	P-Value	Information
Negative control	0%		
Positive control	95.2%	0,000	Not Significant
Concentration 20%	35.2%	0,000	Not Significant
Concentration 30%	48.8%	0,000	Not Significant
Concentration 40%	62.4%	0.006	Significant

Based on the results of the normality test, the results obtained are P-Value > 0.05, which means it is significant, so it can be assumed that the data is normally distributed, data results that have a P-Value < 0.05, which means it is not significant, can be assumed that the data is not distributed properly.

Table 5. Homogeneity test result

Treatment	%Death	P-Value	Information
Negative control	0%		
Positive control	71.2%	0.281	Homogeneous
Concentration 20%	21.3%	0.300	Homogeneous
Concentration 30%	32%	0.463	Homogeneous
Concentration 40%	46.6%	0,000	Not Homogeneous

Based on the results of Levene's test, it was obtained that P-Value > 0.05 which can be assumed that the data is homogeneous, while the results that obtained a P-Value < 0.05 can be assumed that the data is not homogeneous.

Table 6. *Kruskal-Wallis* test result

Variable (group)	P-Value	Information
Negative control Positive control Concentration 20% Concentration 30% Concentration 40%	0.000	Significant

Based on the results of the Kruskal-Wallis statistical test, it is known that the comparison of variations in the concentration of *Allium fistulosum L.* extract shows a P-value < 0.05, which means it is significant.

Discussion

This study aims to determine the ability of *Allium fistulosum L.* extract at concentrations of 20%, 30% and 40% to kill *Aedes aegypti* larvae at 6 hours, 8 hours and 24 hours. The ability to apply ethanol extract in the form of a solution exposed to *Aedes aegypti* larvae in a container. Exposure to solutions can cause damage to the larval cell walls, potentially causing death of *Aedes aegypti* mosquito larvae.

Scallions are plants originating from Southeast Asia which can be planted in various regions with tropical and sub-tropical climates. This plant is known as loncang or muncang and is commonly used by people as an ingredient for cooking. Leeks have been known for their potential as a source of natural antioxidants. Leeks contain various oxidant contents and components such as phenolic compounds, flavonoids (Sukri et al., 2020).

According to research by Siregar et al., 2015, it shows that spring onions have the potential as a source of natural antioxidants containing metabolic compounds in the alkaloids, tannins, phenolics, steroids and flavonoids, where these compounds can be used as natural larvicides. Like alkaloids themselves, they have toxic properties which function as inhibitors of larval growth and then cause a chemical reaction in the body's metabolism, by not developing larvae it can cause a failure in metamorphosis and can degrade a cell membrane and damage cells so that it can disrupt the working nervous system of the larvae by inhibits the work of enzymes, resulting in a decrease in muscle coordination which results in death of the larvae.

CONCLUSIONS

The results of research regarding to the comparison of variations in the concentration of leek extract (*Allium fistulosum L.*) as a natural larvicide on the mortality of third instar *Aedes aegypti* mosquito larvae, it can be concluded that: leek extract (Allium fistulosum L.) as a larvicide significantly affects the mortality of Aedes aegypti mosquito larvae. Aedes aegypti instar III as evidenced by the Sig value. 0.000 (< 0.05). The concentration of *Allium fistulosum L.* extract influences the death of *Aedes aegypti* larvae with a LC50 value of 50 -6 hours at a concentration of 52.850%, LC50 50 -8 hours at a concentration of 42.678%, and an LC50 of 50 -24 hours at a concentration of 30.900%. The time of administration of *Allium fistulosum L.* extract affected the death of *Aedes aegypti* larvae. The LT 50 - 20% value was 37.674 hours, LT 50-30% was 24.423 hours, and LT 50-40% was 14.954 hours. The advice given by researchers is that further research needs to be carried out regarding comparative tests of variations in the concentration of *Allium fistulosum L.* extract as a natural larvicide against the death of *Aedes aegypti* mosquito larvae. In further research, plant parts in the form of stems/roots from *Allium fistulosum L.* can be used.

Author Contribution

Siska Wulandari: Conceptualization, writing draft, writing review dan editing; Yauwan Tobing Lukiyono, Ary Andini, Ersalina Nidianti: Data curation, formal analysis.

Conflict of Interest

There is no conflict of interest in this study.

Acknowledgment

Thank you to all parties involved in completing this research. We would also like to thank the Surabaya Polytechnic Entomology Laboratory for providing laboratory facilities for carrying out this research.

Data Availability

We thank all respondents involved in this research project.

REFERENCES

Dewi, N. K. D. R., Satriani, N. L. A., & Pranata, G. K. A. W. (2022). Hubungan pengetahuan dan sikap terhadap perilaku pencegahan demam berdarah dengue pada masyarakat di kabupaten buleleng. *Jurnal Riset Kesehatan Nasional*, *6*(1), 67–73.

Dinata, C., Rusmartini, T., & Saefulloh, A. (2017). Perbandingan efektivitas larvasida ekstrak etanol bawang putih (allium sativum linn) dan ekstrak etanol bawang daun (allium fistulosum linn) terhadap larva aedes aegypti sebagai vektor demam berdarah dengue. *Prosiding Pendidikan Dokter*, 561–568.

Kemenkes. (2020). Hingga juli, kasus dbd di indonesia capai 71 ribu. *Kementrian Kesehatan Republik Indonesia*.

Prasetyani, R. D., et al. (2015). *Hubungan dukungan keluarga, interaksi sosial dan fungsi kognitif dengan depresi pada lanjut usia di kecamatan rajabasa bandar lampung* [Doctoral dissertation, Fakultas Kedokteran].

Ravaomanarivo, L. H. R., Razafindraleva, H. A., Raharimalala, F. N., Rasoahantaveloniaina, B., Ravelonandro, P. H., & Mavingui, P. (2014). Efficacy of seed extracts of annona squamosa and annona muricata (annonaceae) for the control of aedes albopictus and culex quinquefasciatus (culicidae). *Asian Pacific Journal of Tropical Biomedicine*, *4*(10), 798–806.

Siregar, T. M., Eveline, E., & Jaya, F. A. (2015). Kajian aktivitas dan stabilitas antioksidan ekstrak kasar bawang daun (allium fistulosum l.) *Prosiding Sains Nasional dan Teknologi*, *6*(1).

Sukri, M. S. A., Santoso, A. P. R., Oktavia, A. D. I., Iskandar, F. T., & Andini, A. (2020). Pengaruh pemberian air perasan bawang daun (allium fistulosum l.) terhadap daya bunuh larva nyamuk culex sp. *PROSIDING NATIONAL CONFERENCE FOR UMMAH*, *1*(1).