EFFECTIVENESS OF KECOMBRANG FLOWER (ETLINGERA ELATIOR) AS A LARVACIDE FOR Aedes aegypti INSTAR III MOSQUITOES

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ABSTRACT

Dengue hemorrhagic fever (DHF) is a disease caused by dengue virus infection where the vector is the female Aedes aegypti mosquito. DHF causes high mortality and morbidity because a vaccine for DHF has not yet been found. DHF therapy is still supportive and symptomatic, causing this disease to remain a health problem for the community. The purpose of this study was to determine the effectiveness of kecombrang flower (Etlingera elatior) as Aedes aegypti instar III larvicide. This research was conducted at the Institute of Tropical Disease (ITD) Universitas Airlangga in July 2023. The research design used was true experimental design with post-test only group control design. Divided into 7 groups, namely negative control, 0.1%, 0.6%, 1.1%, 1.6%, 2.1% and 2.6%. The number of samples used in the study was 560 larvae. Each group contained 20 larvae with 50ml of solution containing kecombrang flower extract. Repetition was done 4 times at each concentration. The test used was Kruskal-Wallis (p<0.05), Post-Hoc Mann Whitney U test (p<0.05). The results showed the highest average larval mortality at concentrations of 2.1% and 2.6% at 100% and the lowest average at a concentration of 0.1% at 28.75%. In the Kruskal-Wallis test, each treatment group showed significant differences (p<0.05). The Mann Whitney U post hoc test showed significant differences between the two treatment groups.

Keywords: Aedes aegypti, Kecombrang flower (Etlingera elatior), Larvicide

ABSTRAK

Demam berdarah dengue (DBD) merupakan penyakit yang disebabkan oleh infeksi virus dengue dimana vektornya adalah nyamuk Aedes aegypti betina. DBD menyebabkan angka kematian dan kesakitan yang tinggi karena vaksin DBD belum ditemukan. Terapi DBD yang masih bersifat suportif dan simptomatik menyebabkan penyakit ini masih menjadi masalah kesehatan masyarakat. Tujuan penelitian ini adalah untuk mengetahui efektivitas bunga kecombrang (Etlingera elatior) sebagai larvasida nyamuk Aedes aegypti instar III. Penelitian ini dilakukan di Institute of Tropical Disease (ITD) Universitas Airlangga pada bulan Juli 2023. Desain penelitian yang digunakan adalah true eksperimen design dengan post-test only group control design. Digunakan 7 kelompok yaitu kontrol negatif, 0,1%, 0,6%, 1,1%, 1,6%, 2,1% dan 2,6%. Jumlah sampel yang digunakan dalam penelitian adalah 560 larva. Setiap kelompok berisi 20 larva dengan 50 ml larutan yang mengandung ekstrak bunga kecombrang. Pengulangan dilakukan sebanyak 4 kali pada masing-masing konsentrasi. Uji yang digunakan adalah uji Kruskal-Wallis (p<0,05), uji Post-Hoc Mann Whitney U (p<0,05). Hasil penelitian menunjukkan rata-rata kematuan larva tertinggi pada konsentrasi 2,1% dan 2,6% sebesar 100% dan rata-rata terendah pada konsentrasi 0,1% sebesar 28,75%. Pada uji Kruskal-Wallis setiap kelompok perlakuan menunjukkan perbedaan yang signifikan (p<0,05). Uji post hoc Mann Whitney U menunjukkan perbedaan yang signifikan antara kedua kelompok perlakuan.

Keywords: Aedes aegypti, bunga kecombrang, Etlingera elatior, larvasida
INTRODUCTIONS

Dengue Hemorrhagic Fever (DHF) causes high mortality and morbidity due to the absence of a DHF vaccine and the primarily supportive and symptomatic nature of its treatment. This disease is transmitted by the Dengue virus through the bites of Aedes aegypti and Aedes albopictus mosquitoes.

In 2019, Southeast Asia recorded 658,301 cases of DHF, with Indonesia being one of the five countries in the region with the highest DHF cases. In January to September 2022, there were 87,501 DHF cases and 816 deaths in Indonesia. East Java reported 8,894 cases and 110 deaths, with Surabaya ranking third, following West Java and Bali, in the number of DHF cases in 2018.

Mosquito population control is crucial to combat DHF cases. Temephos 1% is a commonly used chemical larvicide. However, its long-term use can lead to mosquito resistance, and it can also contaminate drinking water.

The use of natural substances as an alternative is being considered due to their environmentally friendly nature. Local plants, such as Etlingera elatior, also known as “Kecombrang flowers,” can serve as larvicides. Kecombrang flowers contain useful larvicidal components, such as alkaloids, flavonoids, tannins, and saponins.

Alkaloid in kecombrang flowers are toxic to insect nerves, while flavonoid inhibits the insect’s respiratory system. Tannin disrupts the metabolism of the insect’s digestive system by inhibiting protease enzymes. Saponins lead to an inflammatory process in the insect’s gastrointestinal mucosa, causing fluid extravasation and significant loss of body fluid.

Based on above explanation, the researchers are interested in studying the effectiveness of kecombrang flowers (Etlingera elatior) as a larvicide for Aedes aegypti instar III. The concentrations used in this study are 0.1%, 1.1%, 1.6%, 2.1% and 2.6%. The extraction method used in this study is maceration with 96% ethanol as the solvent.

The goal of this research is to determine the effectiveness of kecombrang flowers (Etlingera elatior) extracts as a larvicide for Aedes aegypti instar III. This study aims to measure the effectiveness of kecombrang leaves on the mortality of third instar Aedes aegypti.

MATERIAL AND METHOD

Materials

The equipment used in this research includes a micropipette, blender, rotatory evaporator, cloth, vacuum, spatula rod, digital scale, sieve, Erlenmeyer flask, measuring glass, and reaction tube. The materials used consist of kecombrang flowers (Etlingera elatior), distilled water (aquadest), tween 20, 96% ethanol and Aedes aegypti instar III.

Methods

This study is a true-experimental research with a post-test only group control design. The research was conducted at the Entomology Laboratory of the Institute of Tropical Disease (ITD), Universitas Airlangga, Surabaya. The study consists of seven groups, including one negative control (aquadest 50ml) and six treatment groups with concentration 0.1%, 0.6%, 1.1%, 1.6%, 2.1% and 2.6%.

Larva samples were obtained from rearing at Entomology Labpratory ITD, Universitas Airlangga. The larvicide test used third instar Aedes aegypti larvae that were not fed for 24 hours. The samples used were Aedes aegypti larvae that had reached instar III. Each treatment group had 20 larvae with four repetitions as calculated using the Freeder formula.

Data collection was carried out after 24 hours of larval exposure to the extract. The extract is considered effective if the statistical test result yield p<0.05. Data analysis was conducted in stages, starting with a normality test using the Saphiro-Wilk test to determine whether the data was normally distributed. Subsequently, a homogeneity test was performed using the Lavenne test to assess data homogeneity. If the result of both normality and homogeneity test had p>0.05, a One-Way ANOVA test would be conducted. However, in this study, the data did not follow a normal distribution and were not homogeneous, so a non-parametric Kruskal-Wallis test was used. If the Kruskal-Wallis test yield p<0.05, a post hoc Mann Whitney-U test would be conducted to identify groups with significant differences.

Preparations of Kecombrang Flower (Etlingera elatior) Extract

This study used 5.5 kg of kecombrang flowers were obtained from Bangli, Bali. After that, the kecombrang flower will be checked and adjusted to the morphological characteristics of the kecombrang flower. The flowers were washed with running water until clean and the drained. The flower petals were cut into smaller pieces and dried in a shaded area. Once dried, the flowers were ground into a fine powder using a blender and sieved to obtain kecombrang simpelisia powder.

The extraction process involves macerating the simpelisia powder with 96% ethanol solvent for 24 hours to ensure complete immersion, with stirring every 8 hours. The macerate was poured and filtered, and the residue was macerated again with the solvent for a total of three repetitions to extract all secondary metabolites. All the solvent macerates obtained were the evaporated using a rotatory evaporator, resulting, in a concentrated extract 41.5g as semi-solid.

Preparation of Concentration Series

The kecombrang flower extract was made into a 2.6% stock solution, which was the diluted to create required concentration series. The stock solution was prepared using the formula % = (g/ml) x 100%. To dissolve the extract, 16.9g was mixed with 60ml of aquadest. The concentration series of 0.1%, 0.6%, 1.1%, 1.6%, 2.1% and 2.6% were created using the dilution formula V1 x M1 = V2 x M2. In the larvicidal activity test, there were 7 treatment groups with diluted kecombrang flower extract.

RESULT

Result of the preliminary test for the research can be observed in Table 1. The mortality rate for all samples was observed at a concentration of 1.5%, while in the negative control group, no larval deaths were recorded.
The effectiveness of kecombrang flower (Etlingera elatior) as a larvicide... 

Table 1 Preliminary test of the effectiveness of kecombrang flower extract on the mortality of Aedes aegypti instar III mosquito larvae

<table>
<thead>
<tr>
<th>Concentration of Kecombrang Flower Extract</th>
<th>Total Number of Tested Larva</th>
<th>Mortality</th>
</tr>
</thead>
<tbody>
<tr>
<td>Kontrol negatif</td>
<td>20</td>
<td>0</td>
</tr>
<tr>
<td>0.5%</td>
<td>20</td>
<td>19</td>
</tr>
<tr>
<td>1,5%</td>
<td>20</td>
<td>20</td>
</tr>
<tr>
<td>1,75%</td>
<td>20</td>
<td>20</td>
</tr>
<tr>
<td>3%</td>
<td>20</td>
<td>20</td>
</tr>
<tr>
<td>5%</td>
<td>20</td>
<td>20</td>
</tr>
<tr>
<td>10%</td>
<td>20</td>
<td>20</td>
</tr>
</tbody>
</table>

Continuing from this data, probit analysis was performed to determine the LC50 and LC95 values to determine extract concentration to be used in the actual test. The LC50 and LC95 values can be seen in Table 2.

Table 2 Preliminary probit analysis test

<table>
<thead>
<tr>
<th>Concentration</th>
<th>Estimate</th>
<th>Lower Bound</th>
<th>Upper Bound</th>
</tr>
</thead>
<tbody>
<tr>
<td>LC50</td>
<td>0.334</td>
<td>0.181</td>
<td>0.511</td>
</tr>
<tr>
<td>LC95</td>
<td>0.795</td>
<td>0.588</td>
<td>1.439</td>
</tr>
</tbody>
</table>

from the data in Table 2, it can be conducted that to achieve as 50% larval mortality rate, an estimated dose of 0.034% is required, with dose range that can be used from 0.181% to 0.551%. In contrast, to achieve a 95% larval mortality rate, an estimated dose of 0.795% is needed, with a dose range that can be used from 0.588% to 1.439%.

The average mortality data for larvae in actual test can be seen in Graph 1.

**DISCUSSION**

The research result indicate that larval mortality is directly proportional to the concentration of the given extract. this is because a greater number of secondary metabolites contained within the extract, such as flavonoids, alkaloids, tannins, ans saponin enter the larvae’s bodies. The varying result can be attributed to the mosquito’s threshold for detoxifying substance that enter its body. 10

Flavonoid compounds can cause paralysis in the insect’s respiratory muscle system. Alkaloids work by inhibiting acetylcholinesterase enzymes, which can result in nerve damage and the degradation of insect cell walls. Tannins inhibit protease enzymes, and larval digestive activity decreases, causing insect to obtain less food. Saponins trigger an inflammatory process in the insect’s gastrointestinal mucosa, leading to decreased appetite. Furthermore, saponons cause fluid extravasation in the larva’s body, resulting in significant 11,12.

The result of this research show better effectiveness compared to the study conducted by Palgunadi.11 In Palgunadi’s study, a mortality rate of 99.2% was obtained at a concentration of 15%. This difference can be attributed to various factors such as extraction method, solvent use, and the geographical location of sample collection. The maceration method was chosen due to its simplicity and the use of basic equipment, preventing damage to thermolabile substances like flavonoids, alkaloids, and tannins. The use of 96% ethanols as the solvent was based on its high polarity, ensuring optimal compound extraction. This is because the active compounds in plant have nearly the same polarity as 96% ethanol.13 Geographical location affects air humidity, soil quality, rainfall, and plant age, all of which influence the secondary metabolites contained in kecombrang flowers.14

**CONCLUSIONS**

The concentrations of kecombrang flower (Etlingera elatior) extract at 0.1%, 0.6%, 1.1%, 1.6%, 2.1%, and 2.6% are
effective for use as larvicides for Aedes aegypti instar III mosquitoes.

REFERENCE: