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Effectiveness of Tembelekan Leaf Extract (*Lantana camara* L.) on Mortality of *Riptortus linearis*

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Abstract: This study aims to determine the effect, effective concentration, Lethal Concentration 50 (LC_{50}), and Lethal Time (LT_{50}) of tembelekan leaf extract (*Lantana camara* L.) on the mortality of *Riptortus linearis* and metabolic compounds of *L. camara* leaves. The method was experimental. The study used a completely randomized design (CRD) with five treatments, namely 0%, 10%, 15%, 20%, and 25%, and five replications. Each treatment contained 10 *R. linearis* test animals. The results of the ANOVA analysis showed the effect of treatment on the mortality of *R. linearis* ($p<0.05$). The concentration that showed the highest mortality was 25%. Based on the LC_{50} test, the 10.155% *L. camara* leaf extract concentration can kill *R. linearis* in more than 50% of each treatment. And based on LT_{50} with a concentration of 25%, it takes 7.681 hours to kill 50% of *R. linearis*. In *L. camara* leaf extract contains metabolite compounds, namely alkaloids with a level of 307.87 mg/L, then tannins with a group of 2.75 %b/v, then saponins with a group of 0.31 %b/v, and flavonoids with a group of 1.40 %b/v., It can be concluded that there is an influence of giving tembelekan (Lantana camara L) leaf juice on the mortality of Riptortus linearis.

Keywords: *Lantana camara* L.; Mortality; *Riptortus linearis*

Introduction

Pod-sucking bugs (*R. linearis*) are insects distributed throughout the soybean and mung bean cropping areas. Pod-sucking bugs are one of the insects that have the potential as pests because the presence of pod-sucking bugs in large numbers or abundance in a soybean planting area can cause damage and decrease soybean and mung bean crop production.

R. linearis can damage seeds in the generative phase, especially during seed formation, resulting in wrinkled, deflated seeds and even pod loss. During the pod development and seed-filling phases, pods and seeds may become deflated and dry out. At the seed ripening stage, black spots appear on the seeds, resulting in a decrease in the quality of the seeds.

Controlling these pests cannot be separated from the use of chemical insecticides or synthetic insecticides. However, synthetic insecticides can harm the environment and cause residual insecticides in the harvest. According to Prayogo (2010), the control of pod bugs carried out by farmers until now only relies on synthetic insecticides. More than 90% of farmers use synthetic insecticides in the field using doses and spray volumes that do not follow the recommendations.

To overcome this problem, other alternatives are needed by using natural ingredients or better known as biopesticides which are relatively non-poisonous to humans, animals, and other plants because they are easily decomposed, so they do not cause residues, besides that natural vegetable pesticides do not cause side effects on the environment, raw materials can be obtained quickly and cheaply. They can be made simple so that farmers can quickly adopt them. One type of plant with the potential as a vegetable pesticide is the *L. camara* (Sakan et al., 2021).

L. camara is one type of plant whose population is found in Gorontalo Province but has yet to be optimally utilized (Sulistiani et al., 2016). The utilization of the tembelakan is the main ingredient of biopesticides

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because it contains active compounds such as alkaloids, flavonoids, and triterpenoids. Previous research stated that *L.camara* contains steroids, saponins, and tannins (Mirna et al., 2023). Saponin-type steroids are one of the ingredients that can function as anti-feeding against insects and saponins that act to inhibit the work of enzymes that cause a decrease in the result of the digestive tract and the use of protein (Jumiati et al., 2023). In addition, this plant is relatively easy to obtain. Its existence is very abundant and develops more quickly (Anitasari et al., 2018).

Based on the description of the problem above, this journal aims to determine the effectiveness of the effect of giving Tembelekan (*Lantana camara* L.) leaf juice on the mortality of pod-sucking ladybirds (Riptortus linearis).

Method

Research Location

This research was conducted at the Zoology Laboratory of the Biology Department of Universitas Negeri Gorontalo to test the mortality of *R. linearis* and at the testing laboratory of LPPT-UGM (Integrated Research and Testing Laboratory) Universitas Gadjah Mada to analyze the content of secondary metabolites of the *L. camara* leaf extract. The research time began from June to August 2022.

Research Design

The research method used is Complete Randomized Design (CRD). Based on Nuraini et al. (2021), it can be used as a reference for five treatments and five replications with the following treatment arrangement: control, 10% concentration (10 ml *l. camara* leaf extract + 90 ml distilled water), concentration 15% (15 ml *l. camara* leaf extract + distilled water 85 ml), concentration 20% (20 ml *l. camara* leaf extract + 80 ml distilled water), concentration 25% (25 ml *L. camara* leaf extract + 75 ml distilled water)

Research Procedures

Preparation

L. camara leaves used are old from the tip, starting from the fourth stalk of 1000 grams, and cleaning them using running water. They were then air-dried to reduce the water content contained in tembelekan leaves. After being air-dried, *L. camara* leaves are cut into small pieces and mashed using a mortar and pestle. Then the smooth *L. camara* leaves are squeezed and filtered into a 100 ml beaker. *R. linearis* were obtained from the Bugs Seed Center in Malang, reared for acclimatization to the laboratory environment for two days, and fed mung bean pods before treatment.

Application of L. camara leaf extract R. linearis

Each container contains one concentration of *L. camara* leaf extract with concentrations namely 10%, 15%, 20%, 25%, and control (Nuraini et al., 2021). Then each replicate is given as much as 20 ml in 100 ml of diluted *L. camara* leaf extract. Each beaker contains ten *R. linearis* and is sprayed using a hand sprayer bottle containing *L. camara* leaf extract.

The mortality rate can be determined using the formula:

$$\text{Mortality} = \frac{\text{A}}{\text{B}} \times 100\ \% \quad (1)$$

Description:

A : Number of dead larvae

B : Total number of larvae

Metabolite content analysis of L. camara leaf extract

The procedure for analyzing the metabolite content of *L. camara* leaf extract is as follows: preparation of standard curve, determination of total alkaloid equivalent quinine spectrophotometric method, determination of total tannin equivalent tannic acid spectrophotometric method, standard curve of saponins from quilaja bark, procedure for determination of total flavonoid equivalent quercetin spectrophotometric method

Data Analysis

Data were collected by counting the number of *R. linearis* deaths in each beaker. Calculations were made every 2-hour interval for 24 hours. Data were recorded and presented in the form of a table. Test animals considered dead are test animals that have not responded or do not move to the stimuli given.

To determine the effect of treatment on the mortality of *R. linearis*, the data were analyzed using ANOVA after checking the normality and homogeneity tests and continued with the LSD further test if there were significant differences. Meanwhile, to determine the LC_{50} (Lethal Concentration 50%) and LT_{50} (Lethal Time 50%) values, the data that have been collected were analyzed using probit analysis. LC_{50} aims to see the optimum concentration that can kill 50% of Riptortus linearis. LT_{50} aims to see how long it takes to kill 50% of *R. linearis*.

Result and Discussion

The study results of the effect of *L. camara* leaf extract on the average mortality of *R. linearis* after being applied for 24 hours can be seen in Table 1. The average mortality of *R. linearis* after being given *L. camara* leaf extract for 24 hours showed a significant difference (P = 0.05) which means that *L. camara* leaf extract has an effect

on the mortality of *R. linearis*. The results showed that 0% concentration (P1) caused average mortality of 0.6, 10% concentration (P2) of 5.2%, 15% concentration (P3) of 6.2, 20% concentration (P4) of 7.8, and 25% concentration (P5) of 8.6. The lowest average mortality of R. linearis was shown by the control (P1) with 0% concentration of *L. camara* leaf extract. In comparison, the highest average mortality of *R. linearis* (many dead) was shown by a 25% concentration of *L. camara* leaf extract or treatment 5. Shows that *L. camara* leaf extract can cause death to *R. linearis*. It can be used as an alternative material for pest control (Hendrival et al., 2012).

Table 1. Mortality of *R. linearis* 4 Hours after Application of *L.camara* Leaf Extracts at Different Concentrations

Treatment	Mortality of R. *linearis*					Total	Average
	R1	R2	R3	R4	R5	Mortality	Mortality
P1 (0%)	1	0	1	0	1	3	0.6[a]
P2 (10%)	4	5	6	5	6	26	5.2[b]
P3 (15%)	5	6	7	6	7	31	6.2[c]
P4 (20%)	7	8	7	8	9	39	7.8[d]
P5 (25%)	8	9	8	9	9	43	8.6[d]

Based on LSD analysis, P1 treatment significantly differs from P2, P3, P4, and P5 treatments. The same results were shown by P2 and P3 treatments. Meanwhile, the P4 and P5 treatments showed insignificant differences, meaning there was no difference between the 20% and 25% concentrations. This indicates that in this study, the concentration of *L. camara* leaf extract that effectively kills *R. linearis* is 20-25%. Another study also showed that a concentration of 17% *L. camara* flower extract effectively killed mosquitoes *A. aegypti* (Wardani et al., 2022).

Mortality of *R. linearis* occurred within the first hour after *L. camara* leaf extract was applied and continued to increase until the 24th hour after application. The larval mortality rate proportionally increased as the concentration of the extract increased. These data show that the higher the *L. camara* leaf extract concentration, the higher the mortality rate (Figure 1). This is due to the content of secondary metabolite compounds from *L. camara* leaf extract and the increasing concentration and exposure of secondary metabolite compounds to *R. linearis*. *L. camara* leaf extract contains components in its volatile oil known as terpenoids, which may have caused a high mortality rate (Manait II, 2023).

This is in line with Putra et al. (2016), which shows that in the mortality of gold snails tested using four different concentrations, the results show that with each addition of insecticide concentration, there will be an increase in the percentage of gold snail mortality. Another study also found different mortality levels in *S. exigua,* given *L. camara* leaf extract with different concentrations (Mubarak, 2022). The results showed that the higher the level of mortality that occurred, the higher the concentration given (Liswarni et al., 2018).

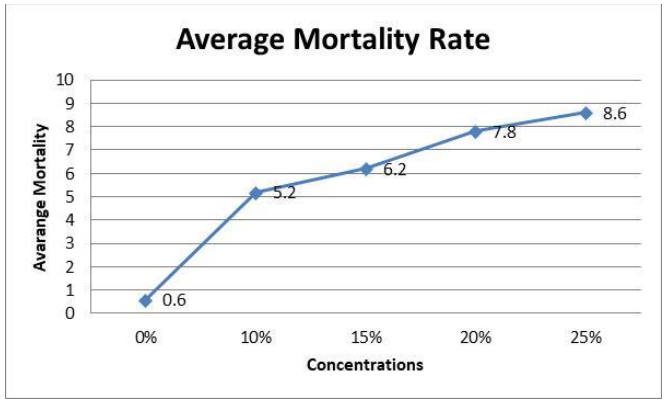

Figure 1. Average Mortality of *R. linearis* after being treated with *L. camara* leaf extract for 24 hours

LC_{50} is the concentration required to kill 50% of the *R. linearis* population. The following are the results of the probit analysis to determine LC_{50}.

Table 2. Lethal Concentrations 50% 24-Hour (LC_{50}) of *L. camara* Leaf Extract against *R. linearis*

Lethal Concentration	Concentration		
	Estimation	Lower Limit	Upper Limit
LC_{50}	10.155	6.257	12.451

Table 2 shows that to kill 50 percent of *R. linearis*, a concentration of 10.155% is needed. This means that to kill 50% of R. linearis, the concentration of *L. camara* leaf extract is 10.15% for 24 hours. In accordance with research conducted by Hamid (2021), it was found that the application of *L. camara* leaf extract affects the mortality of armyworms. In addition, the LC_{50} 24-hour value for the concentration of *L. camara* leaf extract on the mortality of armyworms was recorded at 9.762%. Indicates that at this concentration, *L. camara* leaf extract can kill armyworms with a mortality rate of more than 50%.

Meanwhile, based on Table 3, it is known that to kill 50% of *R. linearis*, it takes different times according to the concentration of tembelekan leaves. For treatment P2, with a concentration of 10% takes 23.187 hours to kill 50% of *R. linearis*. P3 concentration of 15% takes 14.396 hours to kill 50% of *R.linearis*, P4, with a concentration of 20%, takes 8.821 hours to kill 50% of *R. linearis*, and for P5 concentration of 25% takes 7.681 hours to kill 50% of *R. linearis*. Puspitalia et al. (2018) stated in his research that *L. camara* leaf water extract works relatively slowly in killing *S. exigua* larvae, presumably because *L. camara* leaf water extract is a stomach and respiratory poison.

Table 3. Lethal Time 50% 24-Hour (LT_{50}) of *L. camara* Leaf Extract against *R. linearis*

Treatments	Lethal Time	Concentration		
		Estimation	Lower Limit	Upper Limit
P2 (10%)	LT 50	23,187	19,323	30,191
P3 (15%)	LT 50	14,396	12,314	17,280
P3 (20%)	LT 50	8,821	7,473	10,205
P4 (25%)	LT 50	7,681	6,655	8,690

The death of *R. linearis* due to the treatment is caused by secondary metabolites in the natural insecticide base material in the *L. camara* leaf extract. Secondary metabolites are organic compounds produced by plants that have no direct role in growth, reproduction, and development (Salam et al., 2023). Secondary metabolites can also be by-products (intermediaries) of primary metabolism. The three main groups of secondary metabolites plants produce terpenoid compounds, phenols, and nitrogen-containing compounds such as alkaloids and glucosinolates (Mastuti, 2016). The content of secondary metabolite compounds in certain plants that are toxic to pests is then seen as a potential for using these plants as vegetable insecticides.

Table 4. The Content of Secondary Metabolite of *L. camara* Leaf Extract

Secondary Metabolites	Results
Steroids	1692.45 µg/g
Saponins	0.31 % b/v
Tannins	2.75 % b/v
Alkaloids	307.87 mg/L
Flavonoids	1.40 % b/v

In this study, based on phytochemical analysis, *L. camara* leaf extract contains secondary metabolites as shown in Table 4 above. The analysis showed that the plant has steroids, saponins, tannins, alkaloids, and flavonoids. Significant total amounts of steroids (1692.45 µg/g), saponins (0.31 % b/v), tannins (2.75 % b/v), alkaloids (307.87 mg/L), flavonoids (1.40 % b/v). The same study also reported the results of phytochemical analysis of *L. camara* leaf extracts showing the presence of alkaloids, tannins, flavonoids, and saponins (Lourenço et al., 2022).

Flavonoids have various biological activities, including vasodilation, anti-carcinogenic, anti-inflammatory, antibacterial, immune-stimulating, anti-allergic, antiviral, and radioprotective effects. In addition, flavonoids have also been shown to be better antioxidants (Sivakumar et al., 2022). Alkaloid compounds that are toxic act as stomach poisons and contact poisons. Alkaloids in the form of salts can degrade the cell membrane of the digestive tract to enter and damage cells and can also interfere with the nervous system by inhibiting the work of the enzyme acetylcholinesterase (Kurniawan et al., 2013).

Conclusion

This study concluded that *L. camara* leaf extract could effectively source secondary metabolites in killing *R. linearis*. Therefore, the use of *L. camara* leaf extract treatment as a biopesticide is worth considering.

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