

Efficacy of Turmeric Rhizome Infusion (*Curcuma longa* L.) on Mortality of *Aedes* sp. Mosquito Larvae

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Abstract: *Aedes aegypti* and *Aedes albopictus* are mosquito vectors that are thought to be the cause of Dengue Hemorrhagic Fever (DHF), especially in Indonesia, because it is a tropical climate area, and the rate of disease transmission due to mosquitoes is high. Besides that, Indonesia is known for being rich in plant species that can be used as natural larvicides. Turmeric rhizome (*Curcuma longa* L.) is a plant that contains secondary metabolites that can be used as larvicide. The purpose of this study was to test the effectiveness of turmeric rhizome infusion (*Curcuma longa* L.) on the mortality of *Aedes aegypti* and *Aedes albopictus* mosquito larvae. In this study, turmeric rhizome (*Curcuma longa* L.) infusion was used at concentrations of 25%, 50%, and 75%, as well as the positive control (abate drug) and the negative control (aquadest). The Kruskal–Wallis test results showed a probability value (p-value) of 0.001 ($p < 0.05$) in both *Aedes aegypti* and *Aedes albopictus* larvae, indicating a significant difference between treatment groups, especially between the positive control and the 75% concentration of turmeric rhizome infusion treatment. Furthermore, the Mann–Whitney U test results showed that the 75% concentration exhibited a larvicidal effect against both mosquito larval types. Turmeric rhizome infusion (*Curcuma longa* L.) was proven to have inhibitory activity against *Aedes* sp. larvae, although its effectiveness was still lower than that of chemical larvicides. However, the community considered the use of turmeric rhizome infusion safer, simpler, and easier to apply than natural larvicides. Therefore, this study can serve as a reference for developing natural larvicidal agents from turmeric rhizomes (*Curcuma longa* L.).

Keywords: *Aedes aegypti*; *Aedes albopictus*; Infusion; Larvacida; Turmeric Rhizome.

Introduction

Mosquito-borne diseases are currently endemic in more than 100 countries worldwide, and 700 million people are infected annually [1]. Mosquito-borne infectious diseases tend to occur in tropical climates because the environment, with its warmer temperatures and high humidity, is a supporting factor for the continuation of the mosquito's life cycle as a disease vector [2]. Indonesia is a tropical climate region that has a high prevalence of mosquito-borne infectious diseases [3].

Dengue Hemorrhagic Fever (DHF) is a mosquito-borne infectious disease that remains a global health problem [4]. DHF cases occur at high rates every year and contribute significantly to morbidity and mortality in various provinces, especially during the rainy season when mosquito populations increase drastically [5]. According to data from the *World Health Organization* (WHO), in 2024, 164,673 cases of DHF were reported throughout Indonesia, with 926 deaths [6].

Dengue Hemorrhagic Fever (DHF) is transmitted through the bite of the *Aedes* sp [7]. The *Aedes aegypti* mosquito is the main vector, and *Aedes albopictus* is a potential vector of dengue fever in Indonesia [7][8]. Both types of mosquitoes are characterized by their dark bodies with white stripes on the body and legs [9]. *Aedes* sp. mosquitoes will actively bite in the morning and evening. Seasonal patterns and environmental factors such as rainfall,

temperature and human activity influence the increase in cases of infection [10].

Larvicide is a type of insecticide that kills mosquitoes, particularly the larval and pupal stages [11]. Larvicide is made from imidazolium-based chemicals [9], considered effective against *Aedes aegypti* and *Aedes albopictus* larvae with a working mechanism that damages the intestinal tissue of the larvae, causing death [12]. However, the use of chemical larvicides can risk causing resistance [13]. Therefore, an alternative to chemical larvicides is needed, using natural ingredients that are safer but effective for controlling mosquito larvae and preventing resistance.

Indonesia is known for its rich natural resources that can be used as natural larvicides. Turmeric rhizome (*Curcuma longa* L.) is a plant that is easily found in Indonesia and is often used as a kitchen spice to enrich the flavor of dishes, as well as a traditional medicine ingredient [14]. Turmeric leaf infusion shows potential as a natural larvicide against Instar III *Aedes aegypti* larvae: at a concentration of 30%, it causes only 2.5% larval mortality within 24 hours, and at a concentration of 100%, after 24 hours, the mortality rate increases to 17.5% [13]. In addition, turmeric extract shows a larvicidal effect, with an LC₅₀ of 4.074 ppm. The main curcuminoid compound in turmeric rhizomes has potential as a natural larvicide [15]. The novelty of this study not only in the variation in concentration and observation time, but also in the plant parts used. While previous studies used leaves, this study

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uses turmeric rhizomes (*Curcuma longa* L.), which contain higher levels of active compounds, such as curcumin, than leaves. By using turmeric rhizomes and observing at 30, 60, 90, and 120 minute intervals at concentrations of 25%, 50%, and 75%, this study is expected to demonstrate a stronger and faster-acting larvicidal effect on *Aedes* sp. larvae.

Based on this background, the aim of this study is to determine the effectiveness of turmeric rhizome infusion (*Curcuma longa* L.) on the mortality of *Aedes aegypti* and *Aedes albopictus* mosquito larvae as a natural larvicide for mosquito control. This research focuses on the use of turmeric rhizome infusion (*Curcuma longa* L.) to facilitate community use and ensure its safety as a larvicide that inhibits mosquito larval growth.

Research Methods

Tools and Materials

The materials used in this study were *Aedes aegypti* and *Aedes albopictus* mosquito larvae, turmeric rhizome (*Curcuma longa* L.), a microscope, an objective lens, a coverslip, sterile cotton, sterile distilled water (negative control), and the abate drug (positive control). The tools used were a beaker, a glass, a hotplate, an analytical balance, a knife, a stirring rod, a Petri dish, a dropper pipette, a filter, a measuring cup, a measuring pipette, a sterile brown bottle, and a stopwatch.

Method

This is an experimental study with a posttest-only control group design, in which observations were conducted simultaneously on the control and treatment groups to determine the effect of turmeric rhizome (*Curcuma longa* L.) infusion on the mortality of *Aedes aegypti* and *Aedes albopictus* larvae.

Preparation of Turmeric Rhizome (Curcuma longa L.) Infusion

The collected turmeric rhizomes (*Curcuma longa* L.) are then sorted, washed thoroughly under running water, and air-dried. The outer skin of the turmeric rhizomes (*Curcuma longa* L.) is then peeled off with a knife. Fresh turmeric rhizomes that have been cleaned are immediately processed into an infusion by boiling. Add 250g of turmeric rhizome to 500ml of aquadest, then heat for 15 minutes, starting when the temperature reaches 90-98°C, while stirring occasionally. Then, variation concentrations of 25%, 50%, and 70% were made.

Identification of Mosquito Larvae

Mosquito larvae were identified at the fourth instar stage. *Mosquito larvae are identified at the fourth instar stage.* Differences between *Aedes aegypti* and *Aedes albopictus* larvae can be observed in the base of the thoracic hairs and the presence and shape of spines on the eighth abdominal segment. Observations were made using a microscope at 10x and 40x magnification.

Effectiveness Test of Turmeric Rhizome Infusion (Curcuma longa L.) Against Aedes aegypti and Aedes albopictus Mosquito Larvae

A total of 12 petri dishes were arranged as test containers, then each petri dish was filled with a test solution according to the treatment group, namely: Group I (positive control): 15ml of aquades with the addition of 5 drops of abate. Group II (negative control): 15ml distilled water. Group III (treatment I): turmeric rhizome infusion at a concentration of 25 %. Group IV (treatment II): turmeric rhizome infusion (50 %). Group V (treatment III): turmeric rhizome infusion at a concentration of 75 %.

Afterwards, each petri dish was filled with 20 larvae, consisting of 6 petri dishes containing *Aedes aegypti* larvae and 6 petri dishes containing *Aedes albopictus* larvae. Observations were made for 30, 60, 90, and 120 minutes. Three repetitions were performed to determine the percentage of mosquito larval mortality in response to the turmeric rhizome infusion.

Statistical Test

The results of the effectiveness test of turmeric rhizome infusion on the mortality of *Aedes aegypti* and *Aedes albopictus* mosquito larvae were analysed using the Kruskal-Wallis and Mann-Whitney U test.

Results and Discussion

Larvicide is a type of insecticide that is usually made from chemicals [9] and has the ability to kill mosquitoes, especially in the larval and pupal stages [11]. Imidazolium dan temephos is a type of chemical used as a synthetic larvicide. This compound works by damaging the cells and tissues of the midgut of larvae, thus triggering their death [12]. Although chemical larvicides are considered the most effective, these chemicals cause problems such as vector resistance [13] and ecotoxicity [16]. Therefore, an alternative to chemical larvicides is needed, using natural ingredients that are safer but effective for controlling mosquito larvae and preventing resistance.

Turmeric rhizome (*Curcuma longa* L.) is considered effective as a larvicide due to its active compounds, such as curcumin, flavonoids, and essential oils [17]. These compounds are known to have a multi-target mechanism of action, including disrupting the larval digestive system, damaging cells, and triggering oxidative stress, which leads to larval death. Other studies also report that curcumin-derived compounds have quite high larvicidal activity against *Aedes aegypti* [13] and *Aedes albopictus* [18].

In this study, the mosquito larvae were first identified to determine their species. Based on the identification results, the larvae found belonged to the species *Aedes aegypti* and *Aedes albopictus*. Morphological differences between *Aedes aegypti* and *Aedes albopictus* larvae can be observed in several characteristics, namely the base of the feathers on the thorax and the presence and shape of spines on the eighth abdominal segment [19]. As shown in Figures 1 and 2.

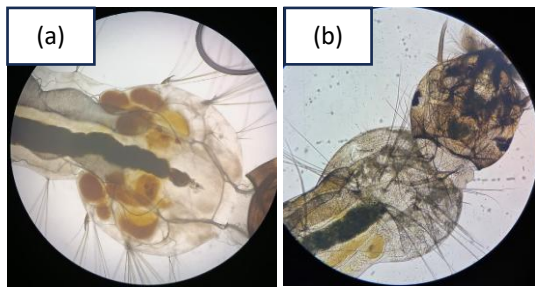


Figure 1. The morphological differences in the thoracic region: (A) *Aedes aegypti* shows well-developed spine-like structures at the bases of the setae on thoracic segments II and III, (B) *Aedes albopictus* exhibits hair bases that appear only as simple protuberances.

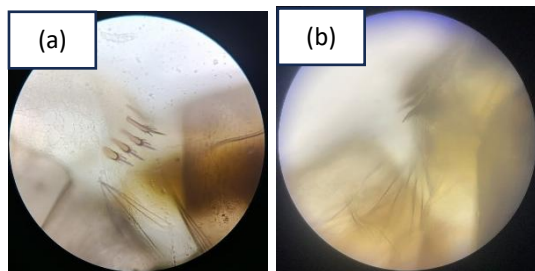


Figure 2. Morphological differences of the comb teeth in the abdomen VIII: (A) *Aedes aegypti* has a central spine and side spines like a trident, (B) *Aedes albopictus* only has spines in the middle (median spines)

Aedes aegypti and *Aedes albopictus* are mosquito species from the subgenus *Stegomyia* in the ordo *Diptera* and family *Culicidae* that act as vectors of disease in humans and are reported to have spread widely in more than 129 countries in the world [20]. Morphological differences between *Aedes aegypti* and *Aedes albopictus* larvae can be observed in several characteristics, namely the base of the feathers on the thorax and the presence and shape of spines on the eighth abdominal segment [19]. In the thoracic region of the *Aedes aegypti* mosquito larvae, there are well-developed thorn-like structures at the base of the setae on thoracic segments II and III, whereas *Aedes albopictus* exhibits hair bases that appear only as simple protuberances. Besides that, in the abdomen VIII of *Aedes aegypti* has a central spine and side spines like a trident, whereas *Aedes albopictus* only has spines in the middle (median spines). The presence of these comb teeth is a distinctive characteristic that distinguishes it from *Aedes aegypti* and *Aedes albopictus* [21].

Next, a test was conducted to determine the effectiveness of turmeric rhizome infusion (*Curcuma longa* L.). Turmeric rhizome infusion (*Curcuma longa* L.) was chosen as a larvicide in this study because it shows greater potential for community application. This is because of its economical, safe, and practical use compared to extracts. The infusion production process is relatively simple, requiring only water as a solvent and a heating method, thus eliminating the need for special equipment or potentially hazardous chemical additives. This approach is considered more suitable for household use, especially in community-based vector control efforts [22]. In addition, the abundant availability of turmeric as a raw material in Indonesia, which

is commonly used as a kitchen spice to enrich the taste of dishes, as well as an ingredient in traditional medicine [14].

The effectiveness test of Turmeric Rhizome Infusion (*Curcuma longa* L.) was carried out on *Aedes aegypti* and *Aedes albopictus* mosquito larvae at concentrations of 25%, 50%, and 75%. The test results are shown in Tables 1 and 2.

Table 1. Effectiveness test of turmeric rhizome (*Curcuma longa* L.) infusion against *Aedes aegypti* larvae

Treatment Group	n	Mean	Notasi	p-value (Kruskal-Wallis)
Control (-)	12	8.25	a	<0.001
25%	12	24.42	b	
50%	12	30.83	bc	
75%	12	34.50	c	
Control (+)	12	54.50	d	

Table 2. Effectiveness test of turmeric rhizome (*Curcuma longa* L.) infusion against *Aedes albopictus* larvae

Treatment Group	n	Mean	Notasi	p-value (Kruskal-Wallis)
Control (-)	12	9.00	a	<0.001
25%	12	24.21	b	
50%	12	28.71	bc	
75%	12	36.08	c	
Control (+)	12	54.50	d	

Based on the Kruskal–Wallis test, a p-value <0.001 was obtained, indicating a significant difference between the treatment groups in *Aedes aegypti* and *Aedes albopictus* larvae. Further Mann–Whitney test results showed significant differences between several pairs of groups, particularly between the positive control and the 75% concentration treatment. The test results are shown in Table 3.

Table 3. Mann-Whitney U test on *Aedes aegypti* and *Aedes albopictus* larvae on concentration 75% with Control Positive (+)

Treatment Group	n	Mean	p-value
Control (+)	12	18.50	<0.001
Cons 75% <i>aegypti</i>	12	6.50	
Cons 75% <i>albopictus</i>	12	6.50	

Based on the Mann–Whitney test, a p-value <0.001 was obtained, indicating a significant difference between the positive control group (abate) and the 75% turmeric rhizome infusion treatment on both types of mosquito larvae. These results indicate that the 75% turmeric rhizome infusion has larvicidal activity against mosquito larvae, but its effectiveness is still lower than that of the positive control (Abate). This can be explained because positive controls generally contain standard synthetic compounds that have more specific effects and have the potential to disrupt the physiological systems of larvae [12]. In addition, the use of natural ingredients such as infusions tends to have a lower active compound content than concentrated extracts, so their killing power is relatively more limited.

Additionally, the data were analysed using the Mann–Whitney U test to determine the most effective concentration for killing *Aedes aegypti* and *Aedes albopictus* larvae. The test results are shown in Table 4.

Table 4. Mann-Whitney U test on *Aedes aegypti* and *Aedes albopictus* larvae at a concentration 75%

Types	n	Mean	p-value
<i>Aedes aegypti</i>	12	15.25	0,051
<i>Aedes albopictus</i>	12	9.75	

The results of the study showed no significant difference in mortality between *Aedes aegypti* and *Aedes albopictus* mosquitoes at a turmeric infusion concentration of 75% ($p > 0.05$), indicating that this concentration had a similar larvicidal effect on both mosquito species. This indicates that both species have relatively similar responses to treatment, despite differences in biological tendencies. This difference in sensitivity may be caused by variations in body structure, metabolism, and the ability to adapt to the environment in each species [22].

Based on the test results, the effectiveness of infusions is generally lower than that of concentrated extracts because infusions typically contain fewer dissolved bioactive compounds. Previous studies using extracts have shown that turmeric rhizomes have potential as larvicides with an LC50 value of 4,074ppm [15]. Extraction with organic solvents is known to be able to extract active compounds such as curcumin more optimally, resulting in higher larvicidal activity [18]. However, this extraction process requires more complex techniques, is more time-consuming, and is more expensive, making it less practical for direct application by the general public. Therefore, despite its limitations in terms of potential killing power, the use of infusions remains a relevant alternative in mosquito larval control strategies that prioritize convenience, safety, and sustainability.

The fairly strong larvicidal effect of turmeric rhizome infusion at a concentration of 75% is thought to be influenced by the content of secondary metabolite compounds such as curcumin, flavonoids, and essential oils, which are known to have toxic effects on mosquito larvae [17]. Curcumin works by damaging the epithelial cells of the larval midgut through cell lysis, cytoplasmic vacuolization, and damage to the peritrophic membrane and microvilli. This damage disrupts the digestion and nutrient absorption processes in the larvae, ultimately leading to death [23]. Flavonoids act as larvicides by entering the larvae's respiratory system and causing toxic effects that can disrupt respiration [24]. Furthermore, flavonoids can inhibit the larvae's metabolic system, causing physiological imbalances, such as feeding, growth, and enzyme function, which can lead to larval death [25]. Meanwhile, essential oils work by inhibiting the enzyme acetylcholinesterase (AChE), causing mitochondrial damage, and disrupting the tissue structure of mosquito larvae, so that the physiological function of the larvae is disrupted and leads to death [26].

Overall, the results of this study indicate that turmeric rhizome infusion has potential as a natural larvicide against *Aedes aegypti* and *Aedes albopictus* larvae. Increasing the concentration increased effectiveness, but it did not compete with the positive control. Therefore, further optimization, such as increasing the concentration, is needed.

Conclusion

Based on the results of research on the efficacy of turmeric rhizome infusion (*Curcuma longa* L.) on the

mortality of *Aedes aegypti* and *Aedes albopictus* mosquito larvae, it can be concluded that turmeric rhizome (*Curcuma longa* L.) infusion with a concentration of 75% is most effective as a larvicide on the mortality of mosquito larvae.

Author's Contribution

S. Y. Kurniawan: As the first researcher, she was responsible for identifying the larvae of mosquitoes and testing the effectiveness of the Turmeric Rhizome Infusion. I.B.A. Kresnapati: assisted in statistical testing. Novitarini: assisted in preparing samples and made an infusion of the Turmeric Rhizome.

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