JKM
Sai Wawai

Jurnal Kesehatan Metro Sai Wawai
Vol. 19, No. 1, June 2026. DOI: https://doi.org/10.26630/jkmsw.v19i1.5603
Journal homepage https://ejurnal.poltekkes-tjk.ac.id/index.php/JKM
Email: jkm@poltekkes-tjk.ac.id

ISSN Online
2657-1390
ISSN Print
19779-469X

Larvicidal Potential of Red Fruit Extract (*Pandanus conoideus* Lamk.) Against Anopheles Larvae: A Pilot Study

Asrianto[1], Rina Purwati[2], Eka Sari Gasela[3], Indra Taufik Sahli[4*], Risda Hartati[5], Fajar Bakti Kurniawan[6], Afika Herma Wardani[7],Fachruddin[8]
[1,2,3,4,5,6,7] Jayapura Health Polytechnic of the Misnistry of Health
[8] Avicena Institute of Technology and Health Kendari

Corresponding author: indrataufiksahli@gmail.com

ARTICLE INFO

Article history

Submitted: 29 January 2026

Revised: 18 February 2026

Accepted: 08 May 2026

Keywords:
Anopheles; Larvacidal; Pandanus

ABSTRACT

Malaria continues to be a persistent public health issue, particularly in endemic regions such as Papua, Indonesia. Targeting mosquito larvae offers a practical point of intervention, especially in areas where breeding sites are accessible. In recent years, plant-derived compounds have gained attention as alternative larvicides, including red fruit (*Pandanus conoideus*), which is known to contain bioactive constituents with potential insecticidal properties. This study evaluated the larvicidal activity of ethanol and hexane fractions of red fruit extract against *Anopheles* larvae. A true experimental design with a completely randomised approach was applied. Extracts were prepared through ethanol maceration followed by fractionation. Larvae were exposed to five concentrations (0, 125, 250, 500, and 1000 ppm), and mortality was recorded at 60, 120, 180, and 240 minutes. Data were analysed using nonparametric tests, including the Kruskal–Wallis and Mann–Whitney U tests. Both ethanol and hexane fractions induced substantial larval mortality, with complete mortality observed at 240 minutes across all concentrations. Mortality increased progressively over time, while differences between concentrations and solvent fractions were not statistically significant ($p > 0.05$), suggesting a similar level of effectiveness under the experimental conditions. The findings indicate that red fruit extract possesses strong larvicidal activity against *Anopheles* larvae. The observed pattern suggests that exposure duration may play a more prominent role than concentration or solvent fraction in determining mortality outcomes. Further investigation is needed to explore dose–response relationships and assess performance under extended exposure and field conditions.

INTRODUCTION

Malaria remains a major public health concern, with an estimated 249 million cases and 608,000 deaths reported globally in 2022 (World Health Organization, 2023). In Indonesia, transmission persists, particularly in Papua, which contributes the highest burden of cases. Current control strategies rely heavily on vector management approaches, including insecticide-treated bed nets, indoor residual spraying, and larval control. Among these, targeting mosquito larvae is considered a practical strategy because the aquatic stage allows for direct, localised intervention (Arivoli et al., 2012). However, prolonged use of synthetic larvicides such as temephos has led to resistance, environmental contamination, and adverse effects on non-target organisms (Regis et al., 2001; Senthil-Nathan, 2020; Utami & Porusia, 2023).

These limitations have encouraged the exploration of plant-based larvicides as more sustainable alternatives. Several studies have demonstrated that plant extracts exhibit strong larvicidal activity, supported by their biodegradability and relatively lower toxicity (Kumar et al., 2023; Pavela et al., 2019). However, most studies focus on crude extracts, and comparative evaluation of different solvent fractions remains limited, particularly for indigenous plants. This

is important because solvent polarity can influence the extraction of bioactive compounds and, consequently, larvicidal effectiveness.

Red fruit (*Pandanus conoideus* Lamk.), an endemic plant from Papua, contains various bioactive compounds, including flavonoids and fatty acids, associated with insecticidal activity(Arumsari et al., 2013; Renyaan et al., 2020; Suprijono et al., 2020). Despite its traditional use, its larvicidal activity across different solvent fractions has not been widely investigated. Therefore, this study aims to evaluate the larvicidal effectiveness of ethanol and hexane fractions of red fruit extract against *Anopheles* larvae as a potential environmentally friendly vector control strategy.

METHOD

This study employed a true experimental design, using a completely randomised factorial approach, to evaluate the effects of solvent fraction and concentration on larval mortality. Red fruit (*Pandanus conoideus* Lamk.) was obtained from the Youtefa traditional market, Abepura, Jayapura. The seeds were separated, air-dried for approximately seven months, and ground into a fine powder. Extraction was performed using maceration with 96% ethanol (1:3 b/v) for three days, with daily filtration. The filtrate was concentrated using a water bath at 37°C and subsequently partitioned with n-hexane to obtain ethanol and hexane fractions.

A stock solution (1000 ppm) was prepared with Tween 80 as an emulsifier (Ayinde et al., 2020) and diluted to 500, 250, and 125 ppm.

Anopheles larvae were collected from stagnant water in a cattle farming area in Koya Koso, Jayapura. Third-instar larvae were selected and acclimatised for five hours. Larvicidal assays were conducted at five concentrations (0, 125, 250, 500, and 1000 ppm), with each treatment performed in duplicate. Ten larvae were exposed to 50 mL of the test solution. Temephos 1% was used as the positive control, and distilled water as the negative control. Mortality was recorded at 60, 120, 180, and 240 minutes. Larvae were considered dead when they showed no response to light or mechanical stimulation (Rathy et al., 2015).

This study was designed as a preliminary laboratory screening to evaluate rapid larvicidal activity; therefore, a shorter observation period (240 minutes) and a smaller sample size were used. These conditions allowed the identification of early toxic effects, although further studies using standard protocols are required.

Data were analysed using nonparametric tests due to non-normal distribution (Shapiro–Wilk test). Differences across concentrations were analysed using the Kruskal–Wallis test, and comparisons between solvent fractions were performed using the Mann–Whitney U test. A p-value < 0.05 was considered statistically significant.

RESULTS

Larval mortality increased progressively with longer exposure times in both the ethanol and hexane fractions, with all larvae ultimately reaching complete mortality at 240 minutes across all tested concentrations, as presented in Table 1. During the initial observation periods, the higher concentrations tended to produce a faster larvicidal response, resulting in greater mortality percentages compared to the lower concentrations. This finding suggests that the extracts may exhibit a concentration-dependent effect during the early stages of exposure. Nevertheless, as the duration of exposure increased, the differences in mortality rates among concentrations gradually diminished, and all treatment groups eventually achieved the same endpoint of complete larval mortality. Statistical analysis further demonstrated that neither concentration nor solvent fraction had a significant effect on larval mortality ($p > 0.05$). These results indicate that both

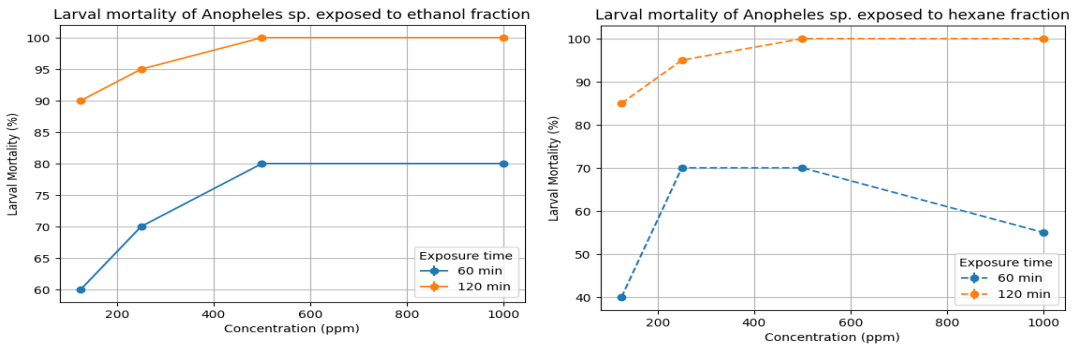
ethanol and hexane extracts possessed comparable larvicidal activity under the experimental conditions applied in this study, with exposure time appearing to play a more dominant role in determining larval mortality than concentration differences. This suggests that the effect on Anopheles larval mortality was relatively similar.

Table 1. Larval mortality (%) across concentrations and solvent fractions (n = 4)

Time (min)	Ethanol, median (min–max)	Hexane, median (min–max)	p-value (Concentration)*	p-value (Fraction)**
60	70 (60–80)	55 (40–70)	0.332	0.200
120	95 (90–100)	92.5 (85–100)	0.075	0.886
180	97.5 (95–100)	92.5 (85–100)	0.086	0.886
240	100 (100–100)	100 (100–100)	1.000	1.000

* Kruskal–Wallis test
** Mann–Whitney U test

At earlier exposure times, a slight difference between fractions was observed, with the ethanol fraction showing a faster initial effect.



A B

Figure 1. Larval mortality (%) of Anopheles sp. exposed to (A) ethanol fraction and (B) hexane fraction at different concentrations after 60 and 120 minutes. Data are presented as median values (n = 4). Later time points (180 and 240 minutes) showed a plateau in mortality and were therefore not included.

Figure 1 shows the pattern of larval mortality at 60 and 120 minutes, with differences among concentrations and solvent fractions still evident. At later observation times (180 and 240 minutes), mortality values approached or reached a plateau, leading to minimal variation across treatments and reducing their ability to distinguish treatment effects.

DISCUSSION

The results of this study demonstrate that both the ethanol and hexane fractions of red fruit extract induced substantial mortality in *Anopheles* larvae, with mortality increasing consistently over time and reaching 100% at 240 minutes. Although the ethanol fraction showed slightly higher mortality at earlier observation points, this difference diminished with longer exposure, resulting in comparable outcomes between both fractions. This pattern suggests that the larvicidal effect of the extract is strongly influenced by exposure duration rather than by solvent fraction alone.

Despite the clear increase in mortality, statistical analysis did not show significant differences across concentrations or between fractions ($p > 0.05$). This finding may be explained

by the relatively uniform response observed across all treatments. The consistently high mortality suggests that even the lowest concentration tested may have already reached an effective biological threshold. Once such a threshold is achieved, further increases in concentration are less likely to produce measurable differences, as the biological system has already responded maximally.

In addition to this threshold effect, exposure time appears to have played a dominant role in shaping the observed outcomes. Larval mortality increased progressively and ultimately reached 100% in all groups, indicating that prolonged exposure enabled sufficient accumulation of active compounds to disrupt essential physiological functions. Under these conditions, differences related to concentration or solvent polarity may become less apparent, as the overall effect converges toward a similar endpoint.

The findings of this study are consistent with previous research, which reports that plant-based larvicides can exhibit comparable effectiveness across different solvent extracts when sufficient levels of bioactive compounds are present. For instance, (Dakum et al., 2021) observed that different solvent extracts of *Hyptis suaveolens* produced similarly high larval mortality, with only minor variation in LC_{50} values. Likewise, (Leandro et al., 2023) reported that extracts of *Himatanthus drasticus* increased larval mortality across multiple fractions, although differences between solvents were relatively small at higher concentrations. These studies support the idea that larvicidal activity may reach a plateau, beyond which differences between treatments become less distinguishable.

However, contrasting findings have also been reported. (da Silva et al., 2020) demonstrated that the hexane fraction of *Croton lundianus* exhibited significantly higher larvicidal activity compared to other fractions, suggesting that certain bioactive compounds may be preferentially extracted in non-polar solvents. Similarly, (Chan et al., 2022) reported clear concentration-dependent effects in *Ocimum basilicum*, with higher concentrations resulting in significantly higher larval mortality. Compared to these studies, the absence of significant differences in the present study may indicate that the active compounds in red fruit extract are relatively evenly distributed between ethanol and hexane fractions, or that the concentration range tested was not sufficiently wide to reveal distinct differences.

Another factor that may have contributed to the lack of statistical significance is the relatively small number of replicates (n = 4), which may have limited the statistical power to detect subtle variations between treatments. In biological assays with inherently variable responses, increasing the number of replicates often improves the ability to identify meaningful differences. Therefore, future studies with larger sample sizes and broader concentration ranges are needed to characterise the dose–response relationship better.

Overall, the findings indicate that red fruit extract possesses strong larvicidal activity, with effectiveness that appears consistent across solvent fractions under the conditions tested. This suggests that both ethanol and hexane extracts may be suitable for further development. However, additional studies are required to optimise extraction methods, identify the most active compounds, and evaluate performance under field conditions, where environmental variability may influence larvicidal efficacy.

This study has several limitations. The number of larvae and replicates was relatively small, and the observation period was limited to short-term exposure. In addition, LC_{50} and LC_{90} values were not determined because mortality reached 100%, preventing the establishment of a dose-response curve. These limitations may affect the generalizability of the findings.

CONCLUSION

Both the ethanol and hexane fractions of the red fruit (*Pandanus conoideus*) extract demonstrated strong larvicidal activity against *Anopheles* larvae, with complete mortality

observed after sufficient exposure time. However, no significant differences were detected between concentrations or solvent fractions, indicating comparable effectiveness under the experimental conditions. These findings suggest that red fruit extract has potential as a plant-based larvicide. However, further work is needed to refine its application, particularly regarding its concentration range and field validation.

AUTHOR'S DECLARATION

Authors' contributions and responsibilities

First author: Conceptualization, Writing original Draft.
Second to Seventh author: Investigation, Data Curation.
Eighth author : Validation, Formal Analysis and editing

Funding
Personal expenses

Availability of data and materials
All data and supporting materials for this study are available and can be requested directly from the corresponding author.

Competing interests
The authors declare no competing interests.

ACKNOWLEDGEMENT

The authors express their appreciation to all parties who have provided facilities, support, and constructive feedback throughout the preparation of this research. Their contributions have greatly assisted in completing this study to the best possible standard

REFERENCES

Arivoli, S., Ravindran, K. J., & Samuel, T. (2012). Larvicidal efficacy of plant extracts against the malarial vector Anopheles stephensi Liston (Diptera: Culicidae). *World Journal of Medical Sciences*, *7*(2), 77-80. https://doi.org/10.5829/idosi.wjms.2012.7.2.62134

Arumsari, N. I., Riyanto, S., & Rohman, A. (2013). Some physico-chemical properties of red fruit oil (Pandanus conoideus Lam) from hexane and chloroform fractions. *A A*, *1*, 25. https://www.semanticscholar.org/paper/Some-Physico-chemical-Properties-of-Red-Fruit-Oil-(-Arumsari-Riyanto/e31d2e0167f634926077bdf14d8d14aa6f2786e7

Ayinde, A., Morakinyo, O., & Sridhar, M. (2020). Repellency and larvicidal activities of Azadirachta indica seed oil on Anopheles gambiae in Nigeria. *Heliyon*, *6*(5). DOI: 10.1016/j.heliyon.2020.e03920

Chan, C. A., Ho, L. Y., & Sit, N. W. (2022). Larvicidal activity and phytochemical profiling of sweet basil (Ocimum basilicum L.) leaf extract against Asian Tiger mosquito (Aedes albopictus). *Horticulturae*, *8*(5), 443.DOI: 10.3390/horticulturae8050443

da Silva, V. E. S., da Silva Rocha, A. R. F., Fernandes, V. M. P., da Conceição, H. N., & Lago, E. C. (2020). Larvicide potential of genus Croton L.(Euphorbiaceae) in biological control of Aedes aegypti L.(Diptera: Culicidae). *Acta Scientiarum. Biological Sciences*, *42*, 1-7. https://doi.org/10.4025/actascibiolsci.v42i1.52755

Dakum, Y., Amajoh, C., Ombugadu, A., Istifanus, G., Agwom, F., Joseph, S., Jwanse, I., Lapang, P., Kopdorah, S., & Pam, D. (2021). Larvicidal efficacy and gc-ms analysis of Hyptis suaveolens leaf extracts against anopheles species. *International Journal of Biochemistry Research and Review*, *30*(1), 8-19. https://doi.org/10.9734/IJBCRR/2021/v30i130241

Kumar, P., Shakya, R., Kumar, V., Kumar, D., Chauhan, R., & Singh, H. (2023). Chemical constituents and strong larvicidal activity of Solanum xanthocarpum among selected plants extracts against the malaria, filaria, and dengue vectors. *Journal of vector borne diseases*, *60*(1), 18-31. https://doi.org/10.4103/0972-9062.361177

Leandro, C., Azevedo, F., Cândido, E., & Alencar, C. (2023). Phytochemical prospection and larvicidal bioactivity of the janaguba (Himatanthus drasticus) Mart. Plumel (Apocynaceae) latex against Aedes aegypti L.(Diptera: Culicidae). *Brazilian Journal of Biology*, *83*, e270143. https://doi.org/10.1590/1519-6984.270143

Pavela, R., Benelli, G., Pavoni, L., Bonacucina, G., Cespi, M., Cianfaglione, K., Bajalan, I., Morshedloo, M. R., Lupidi, G., & Romano, D. (2019). Microemulsions for delivery of Apiaceae essential oils—Towards highly effective and eco-friendly mosquito larvicides? *Industrial Crops and Products*, *129*, 631-640. https://doi.org/10.1016/j.indcrop.2018.11.073

Rathy, M., Sajith, U., & Harilal, C. (2015). Larvicidal efficacy of medicinal plant extracts against the vector mosquito Aedes albopictus. *International Journal of Mosquito Research*, *2*(2), 80-82.

Regis, L., Silva-Filha, M. H., Nielsen-LeRoux, C., & Charles, J.-F. (2001). Bacteriological larvicides of dipteran disease vectors. *TRENDS in Parasitology*, *17*(8), 377-380. DOI: 10.1016/S1471-4922(01)01953-5

Renyaan, A., Suprayitno, E., & Hariati, A. (2020). Nutrient content and endemic red fruit oil (Pandanus austrosinensis) fatty acid profile in Papua Indonesia. Journal of Physics: Conference Series. https://doi.org/10.1088/1742-6596/1665/1/012019

Senthil-Nathan, S. (2020). A review of resistance mechanisms of synthetic insecticides and botanicals, phytochemicals, and essential oils as alternative larvicidal agents against mosquitoes. *Frontiers in physiology*, *10*, 1591. https://doi.org/10.3389/fphys.2019.01591

Suprijono, M., Sujuti, H., Kurnia, D., & Widjanarko, S. (2020). Absorption, distribution, metabolism, excretion, and toxicity evaluation of Papua red fruit flavonoids through a computational study. IOP Conference Series: Earth and Environmental Science, https://doi.org/10.1088/1755-1315/475/1/012078

Utami, A. W., & Porusia, M. (2023). Kajian Literatur Pengaruh Insektisida Nabati Dan Insektisida Sintetik Terhadap Kematian Larva Nyamuk Aedes Aegypti. *Jurnal Kesehatan Masyarakat*, *11*(2), 168-189. https://doi.org/10.14710/jkm.v11i2.37721

World Health Organization. (2023). *World malaria report 2023*.