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# ATIVIDADE LARVICIDA DO EXTRATO ETANÓLICO DAS FOLHAS DE *PIPER ARBOREUM* AUBL. (PIPERALES: PIPERACEAE) CONTRA *AEDES AEGYPTI* LINNAEUS, 1762 (DIPTERA, CULICIDAE)

LARVICIDAL ACTIVITY OF ETHANOLIC EXTRACT OF *PIPER ARBOREUM* AUBL. (PIPERALES: PIPERACEAE) LEAVES AGAINST *AEDES AEGYPTI* LINNAEUS, 1762 (DIPTERA, CULICIDAE)

ACTIVIDAD LARVICIDA DEL EXTRACTO ETANÓLICO DE HOJAS DE *PIPER ARBOREUM* AUBL. (PIPERALES: PIPERACEAE) FRENTE A *AEDES AEGYPTI* LINNAEUS, 1762 (DIPTERA, CULICIDAE)

**Tamires de Moura Lima**[1]
**Kaio da Silva Bandeira**[2]
**Marcos Martins Lima**[3]
**Odair José Queiroz da Silva**[4]
**Ranielly Araújo Nogueira**[5]
**Maria Gabriela Sampaio Lira**[6]
**João Gustavo Mendes Rodrigues**[7]
**Rafael Costa Leite**[8]
**Thiago Ferreira Soares**[9]
**Guilherme Silva Miranda**[10]

## ABSTRACT

*Aedes aegypti* is one of the main vectors of human arboviruses, thus is essential to discover new alternative tools for its control. Therefore, this study aimed to evaluate the larvicidal effects of the ethanolic extract of *Piper arboreum* leaves (EEPA) against *Aedes aegypti*. The extract was prepared in a 70% ethanol extractive solution and was rota-evaporated to obtain the crude extract. In the larvicide assay the third instar larvae (n = 20) were exposed to varying EEPA concentrations (25–200 µg/mL) for 24 h. The larvae were then transferred to fresh glass cups containing only chlorine-free water to assess the mortality rates over 72 h. In addition, larval morphology was evaluated in the experimental groups to determine alterations. Finally, the toxicity of EEPA was assessed in tests with *Artemia salina.* Our results showed that the larval mortality ranged from 17–100% in a dose-dependent manner after 72 h. The highest concentrations (150 and 200 µg/mL) were more effective than 25 µg/mL and the negative control group. The lethal concentration capable of killing 50% larvae (LC50) obtained for EEPA was 80.54 µg/mL (95%CI: 60.81–106.7 µg/mL). Morphological analyses revealed damage to the chitin layer of larvae exposed to EEPA, particularly at higher concentrations. The toxicity test of EEPA on *A. salina* showed an LC50 of 58 µg/mL (95%CI: 45–70.5µg/mL), which indicates elevated toxicity. Therefore, our results demonstrate that EEPA has moderate larvicidal activity against *A. aegypti*; however, further studies are needed to reduce its toxicity against non-target organisms.

## KEYWORDS

Arboviruses; plant extract; vector control; secondary metabolites.

## RESUMO

*Aedes aegypti* é um dos principais vetores de arboviroses humanas, sendo essencial descobrir novas alternativas para seu controle. Portanto, este estudo teve como objetivo avaliar o efeito larvicida do extrato etanólico das folhas de *Piper arboreum* (EEPA) contra o *Aedes aegypti*. O extrato foi preparado em solução extrativa de etanol 70% e rota-evaporado para obtenção do extrato bruto. No ensaio larvicida, larvas de terceiro instar (n = 20) foram expostas a concentrações variadas de EEPA (25–200 µg/mL) por 24 h. As larvas foram então transferidas para béqueres de vidro contendo apenas água sem cloro para avaliar as taxas de mortalidade ao longo de 72 horas. Além disso, a morfologia larval foi avaliada nos grupos experimentais para determinar alterações. Por fim, a toxicidade do EEPA foi avaliada em testes com *Artemia salina*. Nossos dados demonstraram que a mortalidade larval variou de 17 a 100%, de maneira dose-dependente, após 72 horas. As concentrações mais altas (150 e 200 µg/mL) foram mais eficazes que 25 µg/mL e o grupo controle negativo. A concentração letal capaz de matar 50% das larvas (CL50) obtida para EEPA foi de 80,54 µg/mL (IC 95%: 60,81–106,7 µg/mL). Análises morfológicas revelaram danos à camada quitinosa das larvas expostas ao EEPA, principalmente em concentrações mais elevadas. O teste de toxicidade EEPA em *A. salina* mostrou uma CL50 de 58 µg/mL (IC 95%: 45–70,5 µg/mL), o que indica elevada toxicidade. Portanto, nossos resultados demonstram que o EEPA tem atividade larvicida moderada contra *A. aegypti*; no entanto, mais estudos são necessários para reduzir sua toxicidade contra organismos não-alvo.



## PALAVRAS-CHAVES

Arboviroses; extrato vegetal; controle vetorial; metabólitos secundários.



## RESUMEN

*Aedes aegypti* es uno de los principales vectores de arbovirus humanos, y es fundamental descubrir nuevas alternativas para su control. Por lo tanto, este estudio tuvo como objetivo evaluar los efectos larvicidas del extracto etanólico de hojas de *Piper arboreum* (EEPA) contra *Aedes aegypti*. El extracto se preparó en una solución de extracción con etanol al 70% y se evaporó rotatoriamente para obtener el extracto crudo. En el ensayo larvicida, las larvas de tercer estadio (n = 20) fueron expuestas a concentraciones variables de EEPA (25–200 µg/mL) durante 24 h. Luego, las larvas fueron transferidas a vasos de vidrio que contenían solo agua libre de cloro para evaluar las tasas de mortalidad durante 72 h. Además, se evaluó la morfología larvaria en los grupos experimentales para determinar alteraciones. Finalmente, se evaluó la toxicidad del EEPA en pruebas con *Artemia salina*. Nuestros resultados demostraron que la mortalidad larvaria varió de 17 a 100% de manera dosis-dependiente después de 72 h. Las concentraciones más altas

(150 y 200 µg/mL) fueron más efectivas que 25 µg/mL y el grupo control negativo. La concentración letal capaz de matar el 50% de las larvas (CL50) obtenida para EEPA fue 80,54 µg/mL (IC del 95%: 60,81–106,7 µg/mL). Los análisis morfológicos revelaron daño a la capa de quitina de las larvas expuestas a EEPA, particularmente a concentraciones más altas. La prueba de toxicidad de EEPA en *A. salina* mostró una CL50 de 58 µg/mL (IC del 95%: 45–70,5 µg/mL), lo que indica una alta toxicidad. Por lo tanto, nuestros resultados demuestran que EEPA tiene una actividad larvicida moderada contra *A. aegypti*; sin embargo, se necesitan más estudios para reducir su toxicidad contra organismos no objetivo.

## PALABRAS CLAVE

Arbovirus; extracto de plantas; control de vectores; metabolitos secundarios.

## 1 INTRODUCTION

*Aedes aegypti* Linnaeus, 1762 is considered one of the main vectors of human arboviruses (viruses transmitted by arthropods), such as Dengue, Zika, Chikungunya, and Yellow Fever (TERRA *et al*., 2017; SCOLARI *et al*., 2019; WHO, 2024). Among them, Dengue fever is the most important arbovirus infection to which approximately half of the world population is exposed in more than 80 countries (AKINSULIE; IDRIS, 2024). Indeed, its incidence has increased eight-fold over the past two decades (WHO, 2022). This increase is associated with factors such as globalization, the adaptation of vectors to growing urbanization, and environmental changes (GOULD *et al*., 2017; CHIKEZIE *et al*., 2024).

Owing to the absence of effective vaccines and specific treatments for most diseases transmitted by *A. aegypti,* the main alternative for preventing the spread of arboviruses is to control these vectors (MOYES *et al*., 2017; MORAIS *et al*., 2020). Thus, insecticides based on synthetic compounds such as organophosphates, pyrethroids, and carbamates have been used for many years. However, owing to the uncontrolled and continuous use of these insecticides, resistance has rapidly emerged in some strains of *A. aegypti* (MELO-SANTOS *et al*., 2010; MOYES *et al*., 2017; ZULFA *et al*., 2022). Because of the production of various secondary metabolites, plants have gained focus of the studies seeking novel non-toxic and biologically active compounds against *A. aegypti* (SILVÉRIO *et al*., 2020), which can help prevent the spread of resistance in these mosquitoes and preserve environmental health.

The Piperaceae family is one of the main groups of plants studied with biotechnological potential, with the genus *Piper* being the most representative, with approximately 700 species (PESSINI *et al*., 2003; LARA JÚNIOR *et al*., 2012). Among the species found in Brazil, *P. arboreum* (known as 'Pimenta de macaco') has a wide distribution, occurring from the northern to the southern region (GUIMARÃES *et al*., 2015). Some biological effects of *P. arboreum* have been previously described, such as antimalarial (YIMTCHUI *et al*., 2024), antibacterial, antioxidant (KRISHANA *et al*., 2024), and anthelmintic (SILVA ALVES *et al*., 2020); however, the larvicidal action of *P. arboreum* against *A. aegypti* has not

yet been evaluated. Therefore, this study aimed to describe the larvicidal effects and toxicity levels on non-target organism of the ethanolic extract of *Piper arboreum* (EEPA) leaves against *A. aegypti*.

## 2 METHODS

### 2.1 PLANT MATERIAL AND ETHANOLIC EXTRACT PRODUCTION

The leaves of *Piper arboreum* used to produce the ethanol extract were collected in the morning (9:00 AM) in the city of São Luís, Maranhão, Brazil (02°33' 38.5"S and 044°14'19.6' W). The plant species was confirmed by specialists from the Rosa Mochel Herbarium at the State University of Maranhão (UEMA) and deposited under voucher number 4929. The extract was prepared as previously described by Silva Alves *et al*. (2020), obtaining a yield of 16%. Briefly, the plant material was collected, cleaned, dried and powdered. The powder was macerated with a 70% hydroethanolic solution, a traditional extraction used in Brazilian folk medicine (RODRIGUES *et al*., 2020) and mixed every 12 h for a total of 48 h at a ratio of 1:5 (w/v). The extract was first filtered five times and then concentrated under reduced pressure to obtain a dry extract (NEIVA *et al*., 2014). The extract was stored under refrigeration (4 °C) until use. The presence of phytocompounds in ethanolic leaf extracts of *P. arboreum* was performed by High-Performance Liquid Chromatography (HPLC), according to the previous published data (SILVA ALVES *et al*., 2020). This research was registered in the National System for the Management of Genetic Heritage and Associated Traditional Knowledge (registration number AB9E2EC).

### 2.2 COLLECTION EGGS OF *AEDES AEGYPTI*

The eggs of *Aedes* spp. were captured using ovitraps set up in the peri-urban area of houses located in São Raimundo das Mangabeiras, according to Noleto *et al*. (2020). To confirm the *A. aegypti* species, 10% of the eggs from each collection point were stimulated to hatch, and L3 larvae were morphologically evaluated according to Forattini (2002). Only the batches of eggs with 100% confirmation of *A. aegypti* were used in the bioassays. In this study, we used larvae of wild origin because they more precisely simulate real natural conditions to test plant extracts (LUZ *et al*., 2022).

### 2.3 LARVICIDAL BIOASSAY

The larvicidal effect of the ethanol extract of *P. arboreum* (EEPA) was evaluated according to the methodology proposed by the World Health Organization (WHO, 2005), with slight modifications. Briefly, a total of 20 third instar larvae (L3) were exposed to concentrations of 25, 50, 100, 150, and 200 µg/mL of EEPA in a glass beaker containing a final volume of 500 mL of chlorine-free water. For the negative control, the same number of larvae were placed in 500 mL of chlorine-free water. In the positive control, the larvae (n = 20) were maintained in contact with Pyriproxifen (Sumilarv®) at a concentration of 10 µg/mL, as recommended by the manufacturer. The mortality rate was calculated as the percentage of dead larvae compared to the total number of larvae evaluated. These data were used to calculate the lethal concentration capable of killing 50% of the larval population (LC50).

To determine the level of action of EEPA against *A. aegypti* larvae, we used the classification established by Komalamisra *et al*. (2005). This classification is based on LC50 values, as follow describe: LC50 < 50 µg/mL = strong activity; LC50 50–100 µg/mL = moderate activity; LC50 100–750 µg/mL = low activity; and LC50 >750 µg/mL = inactive. At the end of the larvicidal bioassay, larvae from each group were fixed in 70% ethanol and evaluated under an optical microscope to verify possible morphological damage. The experiment was performed in triplicate (technical replicates) and repeated three times (biological replicates).

### 2.4 TOXICITY ASSAY IN *ARTEMIA SALINA*

A toxicity test on the marine microcrustacean *A. salina* was performed according to the protocol proposed by Meyer *et al*. (1982). Ten nauplii were used at each tested concentration (25, 50, 100, 150, and 200 µg/mL), in six-well cell culture plates (Flat Bottom Plate - SARSTEDT, Inc. USA). Mortality rate was evaluated after 24 h using a stereoscopic microscope. Mortality data were used to calculate the LC50 and verify the level of toxicity of the extract, using the following classification: high toxicity (<100 µg/mL), moderate toxicity (100–500 µg/mL), low toxicity (500–1000 µg/mL) and non-toxic (>1000 µg/mL) (MEYER *et al*. 1982; RAJABI *et al*. 2015). The bioassay was performed in triplicate and repeated twice.

### 2.5 STATISTICAL ANALYSIS

Data normality was verified using the Shapiro–Wilk test. The means of the number of dead larvae were compared between groups using analysis of variance (ANOVA) and Bonferroni post-hoc tests. LC50 was calculated using nonlinear regression from the interpolation of mortality data (%) compared to the tested dose (on a logarithmic scale). Statistical significance was set at $p<0.05$. To perform these tests, we used GraphPad Prism software version 8 (Prism Software, Irvine, California, USA).

## 3 RESULTS

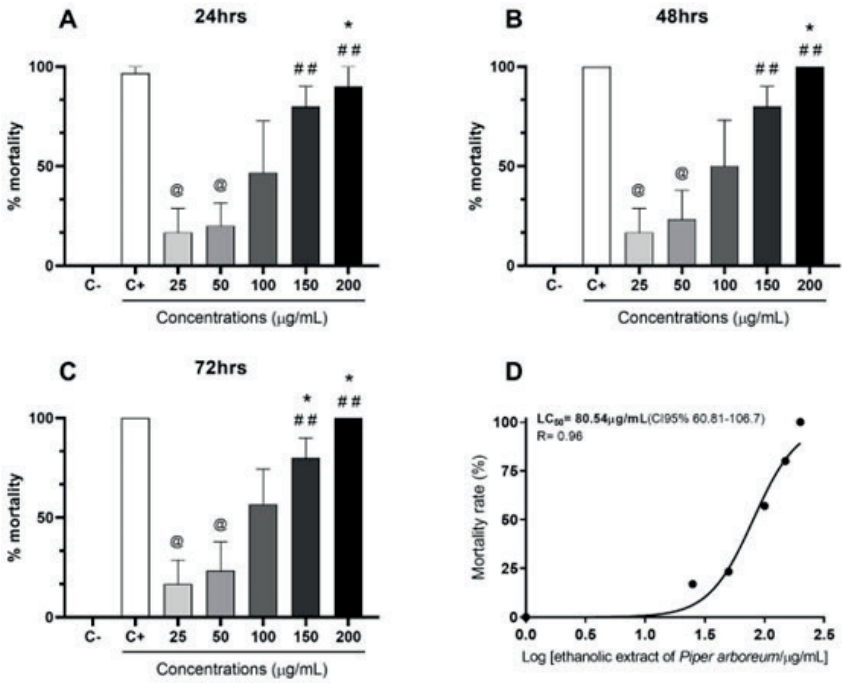
*Aedes aegypti* larvae exposed to different concentrations of *P. arboreum* ethanolic extract exhibited dose-dependent mortality. During the first 24 h, the larval mortality ranged from 17–90% in the test groups. Furthermore, the two highest concentrations (150 and 200 µg/mL) induced significantly higher mortality than the negative control group and showed similar results to those of the positive control group. The larvae mortality observed at a concentration of 200 µg/mL was also statistically higher than that recorded at the lowest concentration used (25 µg/mL) (Figure 1A).

The biological effects of EEPA were enhanced within 48 h. Larval mortality was 17–100% in the test groups. Concentrations of 150 and 200 µg/mL yielded a statistically higher larval mortality rate than the negative control group but similar to the positive control group. However, the 200 µg/mL concentration also induced higher mortality in *A. aegypti* larvae compared to the 25 and 50 µg/mL concentrations (Figure 1B).

At 72 h, the larval mortality remained between 17 and 100%. Furthermore, the two highest concentrations remained the most effective when compared to the negative control group and showed similar efficacy to the positive control group (Figure 1C). However, the concentrations of 150 and 200 µg/mL induced higher mortality than those of 25 and 50 µg/mL (Figure 1C). In all the time points evaluated, the positive control group was more effective than the two lower concentrations of EEPA (25 and 50 µg/mL).

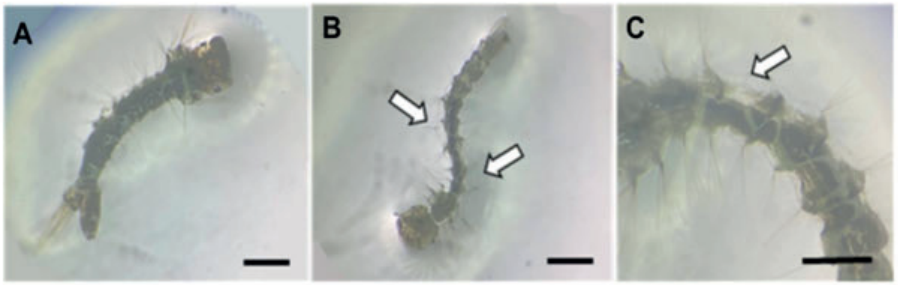
Owing to the better efficacy observed after 72 h, this point was used to calculate the EEPA LC50., which was found to be 80.54 µg/mL (95% CI: 60.81–106.7 µg/mL) (Figure 1D). Therefore, these data classify our extract with moderate larvicidal activity, according to Komalamisra *et al*. (2005).

**Figure 1 -** The mortality rate of *Aedes aegypti* larvae exposed to different concentrations of ethanolic extract from *Piper arboreum* leaves. Statistically significant ($p<0.05$) compared to the negative control group (#), positive control group (@) and 25 – 100µg/mL groups (*).




Complementarily, we also evaluated the morphology of *A. aegypti* larvae exposed to varying EEPA concentrations. Our results demonstrated that the larvae in the control group did not exhibit morphological damage (Figure 2A). However, in the experimental group (Figures 2B, C), a greater frequency of damage to the chitin layer of the larvae (mechanical barrier) was observed, particularly at higher concentrations.

**Figure 2** - Representative photographs of morphological features of *Aedes aegypti* L3 larvae exposed to varying concentrations of ethanolic extract of *Piper arboreum* leaves. Morphology without change in larvae from the control group (A). Presence of morphological damage to the chitin layer (arrows) in experimental group larvae (B and C). The images were observed under a stereomicroscope. Bar = 1 mm.




In our tests using *A. saline*, we found that after 24 h of the experimental procedure, mortality varied between 80% and 100% in a dose-dependent manner. Thus, the LC50 was 58 µg/mL (95% CI: 45–70.5 µg/mL), which classifies our extract as highly toxic, according to Meyer *et al*. (1982) and Rajabi *et al*. (2015).

## 4 DISCUSSION

*Aedes aegypti* mosquitoes are a global problem, as they are vectors of several arboviruses of medical importance, such as Dengue, Zika, Chikungunya, and Yellow Fever (ZARA *et al*., 2016; MATTHEWS, 2019; WHO, 2024); therefore, controlling these vectors is essential. Thus, our study demonstrates for the first time that the ethanolic extract of *Piper arboreum* leaves have moderate biological activity against *A. aegypti* and could be a potential bioproduct to control this mosquito in specific environments.

We showed that EEPA induced larval mortality ranging from 17–100%, with an LC50 of 80.54 µg/mL. According to Komalamisra *et al*. (2005), this LC50 classified the EEPA with moderate larvicidal activity. Based on the recent literature review (LIMA *et al*., 2024), the LC50 of bioproducts derived from *Piper* against *Aedes* larvae was lower than 400 µg/mL. In this review (LIMA *et al*., 2024), the methanolic extract of *P. ribesioides* twigs showed the lower LC50 (1174.34 µg/mL) (KUMRUNGSEE *et al*., 2018). On the other hand, the ethanolic extract of *P. hispidum* leaves has the best larvicidal effect (LC50 = 0.169µg/mL) (PORTO *et al*., 2017). In addition, a previous study showed that the hexane and hydroethanolic extract of *P. cubeba* fruits presented an LC50 of 185.84 ppm (µg/mL) and 191.1 ppm, respectively, against *A. aegypti* larvae (GOMES *et al*., 2023). In contrast, lower LC50 values (34.87 µg/mL) were observed after 3 h of analysis for the methanolic extract of *P. nigrum* leaves (LIJA-ESCALINE *et al*., 2015). Thus, these studies confirmed that LC50 values can vary according to the extraction method, vegetable parts, and *Piper* species (LIMA *et al*., 2024).

The larvicidal action observed in our study may be associated with morphological damage to *A. aegypti* larvae. We found that larvae treated with EEPA at different concentrations showed morphological damage to their external chitin layers compared with the control group. Indeed, extracts from the fruits of *P. cubeba* and the essential oil of *P. capitarianum* have been shown to induce internal and external morphological damage in *A. aegypti* larvae (FRANÇA *et al*., 2021; GOMES *et al*., 2023). The chitin layer that covers the body of mosquito larvae protects against dehydration, infection, and physical damage; therefore, the survival of these insects is associated with this chitin layer (GOMES *et al*., 2023). Hence, its structural deformity induced by EPPA in our study may be associated with the death of mosquito larvae.

The morphological damage observed in the present study may be related to secondary metabolites of EEPA. In our previous study (SILVA ALVES *et al*., 2020), we demonstrated that EEPA predominantly contained amides, flavonoids, and phenolic compounds. Studies have indicated that phenolic compounds and flavonoids associated with other substances, such as triterpenes and tannins, may be associated with inhibiting the growth of insects (MITCHELL *et al*., 1993; KIM *et al*., 2000). Furthermore, hinokinin extracted from fruits of *P. cubeba* is capable of inducing morphological damage in *A. aegypti* larvae (GOMES *et al*., 2023). However, more studies are needed to better elucidate the compounds present in EEPA, so that it is possible to find the likely bioactive substance and its more precise mechanism of action responsible for the death of *A. aegypti* larvae that we found in this study.

Finally, natural products must have toxic effects observed in non-target organisms before being tested in nature. In the present study, we used a bioassay with *Artemia salina,* as it is a species that presents a clear response to small variations in the quality of the environment (MEYER *et al*., 1982; NUNES *et al*., 2006; RIBEIRO *et al*., 2019); therefore, it is an excellent bioindicator. Thus, our data demonstrated that EEPA exhibits high toxicity against *A. salina*, limiting its use in field-based experimental studies. However, further research is necessary to better evaluate EEPA's toxicity in other biological models. Additionally, isolating the major compounds from EEPA or exploring alternative botanical extraction methods could help reduce its toxicity.

## 5 CONCLUSION

In summary, our results demonstrated that EEPA has moderate biological activity against *A. aegypti* larvae. In addition, we verified that the probable mechanism of action of EEPA is morphological damage to the larvae. However, EEPA in this current formulation has remarkable toxicity. Therefore, further studies are necessary to explore new extraction methods that may reduce EEPA toxicity and to identify the primary larvicidal substances present in this extract that exhibit no adverse effects against non-target organisms.

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## REFERENCES


AKINSULIE, O.C., IDRIS, I. Global re-emergence of dengue fever: the need for a rapid response and surveillance. **The Microbe**. v.4. p.100107. 2024.

CHIKEZIE, F. M. *et al*. Impacts of changing climate on arthropod vectors and diseases transmission. **Niger J Entomol**. v.40, n.1, p.179–92, 2024.

FORATTINI, O. P. **Culicidologia Médica. Identificação, Biologia, Epidemiologia**. Editora Universidade de São Paulo, 2th ed. São Paulo, Brasil. 860 pp., 2002.

FRANÇA, L. P. *et al*. *Piper capitarianum* essential oil: a promising insecticidal agent for the management of *Aedes aegypti* and *Aedes albopictus*. **Environ Sci pollut Res Int**. v. 28, n. 8, p. 9760-9776, 2021.

GOMES, A. C. *et al*. Larvicidal potential of extracts and isolated compounds from *Piper cubeba* fruits against *Aedes aegypti* (Diptera: Culicidae) larvae. **Nat Prod Res**. v. 37, n. 16, p. 2787-2794, 2023.

GOULD, E. *et al*. Emerging arboviruses: Why today? **One health**. v. 4, p. 1-13, 2017.

GUIMARÃES, E. F. *et al*. **Piperaceae in Lista de Espécies da Flora do Brasil.** Jardim Botânico do Rio de Janeiro, 2015. Accessed on: 04 January 2024. Available at: https://floradobrasil2015.jbrj.gov.br/jabot/floradobrasil/FB12848

KIM, J. S. *et al*. Inhibition of alpha-glucosidase and amylase by luteolin, a flavonoid. **Biosci Biotechnol Biochem**. v. 64, n. 11, p. 2458-2461, 2000.

KOMALAMISRA, N. *et al*. Screening for larvicidal activity in some Thai. **Southeast Asian J Trop Med and Public Health**. v. 36, n. 6, p.1412-1422, 2005.

KRISHANA, G. M. *et al*. Sustainable synthesis of zinc oxide nanoparticles using *Piper betle* petiole leaf extract: Antibacterial, antioxidant, and cytotoxic potential. **Results Chem.** v. 9, p.101646, 2024.

KUMRUNGSEE, N. *et al*. Bioefficacy of *Piper ribesioides* (Piperaceae: Piperales) extracts and isolated piperine against *Aedes aegypti* (Diptera: Culicidae) larvae and their impact on detoxification enzymes. **Biopestic Int.** v. 14, n. 1, p. 25-32, 2018

LARA JÚNIOR, C. R. *et al*. Antimicrobial activity of essential oil of *Piper aduncum* L. (Piperaceae). **J Med Plants Res**. v. 6, n. 21, p. 3800-3805, 2012.

LIJA-ESCALINE, J. *et al*. Physiological and biochemical effects of botanical extract from *Piper*

*nigrum* Linn (Piperaceae) against the dengue vector *Aedes aegypti* Liston (Diptera: Culicidae). **Parasitol Res**. v. 114, n. 11, p. 4239-4249, 2015.

LIMA, M. M. *et al*. *Piper* plants as an alternative to the control of *Aedes aegypti* and *Aedes albopictus*: an integrative review. **Interfaces Cient Saúde Amb**, v. 9, n. 3, p. 610-631, 2024.

LUZ, T. R. S. A. *et al*. Seasonal variation in the chemical composition and larvicidal activity against *Aedes aegypti* L. of essential oils from Brazilian Amazon. **Exp Parasitol.** v. 243, p. 108405, 2022

MATTHEWS, B.J. *Aedes aegypti*. **Trends Genet**. v. 35, n. 6, p. 470-471, 2019.

MELO-SANTOS, M.A. *et al*. Resistance to the organophosphate temephos: mechanisms, evolution and reversion in an *Aedes aegypti* laboratory strain from Brazil. **Acta Trop**. v. 113, n. 2, p. 180-189, 2010.

MEYER, B.N. *et al*. Brine shrimp: a convenient general bioassay for active plant constituents. **Planta med**. v. 45, n. 5, p. 31-34,1982.

MITCHELL, M.J. *et al*. Effects of plant flavonoids and other allelochemicals on insect cytochrome P-450 dependent steroid hydroxylase activity. **Insect Biochem Mol Biol**. v. 23, n. 1, p. 6571,1993.

MORAIS, H. L. M. D. N. *et al*. Hydroalcoholic extract of *Caryocar brasiliense* Cambess. leaves affect the development of *Aedes aegypti* mosquitoes. **Rev Soc Bras Med Trop**. v. 53, p. e20200176, 2020.

MOYES, C. L. *et al*. Contemporary status of insecticide resistance in the major *Aedes* vectors of arboviruses infecting humans. **PLoS Negl Trop Dis**. v. 11, n. 7, p. e0005625, 2017.

NEIVA, V. A. *et al*. Plant species used in giardiasis treatment: ethnopharmacology and in vitro evaluation of anti-*Giardia* activity. **Rev Bras Farmacogn**. v. 24, p. 215–224, 2014.

NOLETO, J. V. *et al*. Use of ovitraps for the seasonal and spatial monitoring of *Aedes* spp. in an area endemic for arboviruses in Northeast Brazil. **J Infect Dev Ctries**. v. 14, n. 4, p. 387-393, 2020.

NUNES, B. S. *et al*. Use of the genus *Artemia* in ecotoxicity testing. **Environ Pollut.** v. 144, n. 2, p. 453-462, 2006.

PESSINI, G. L. *et al*. Análise farmacognóstica de *Piper regnellii* (Miq.) C. DC. var. pallescens (C. DC.) Yunck: aspectos botânicos e enfoque físico-químico preliminar. **Lat Am J Pharm**. v. 22, p. 209-216, 2003.

PORTO, K. R. *et al*. Screening of plant extracts and fractions on *Aedes aegypti* larvae found in the state

of Mato Grosso do Sul (Linnaeus, 1762) (Culicidae). **An Acad Bras Ciênc**. v. 89, p. 895-906, 2017.

RAJABI, S. *et al*. *Artemia salina* as a model organism in toxicity assessment of nanoparticles. **DARU J Pharm Sc**. v. 23, n. 1, p. 1-6, 2015.

RIBEIRO, E.A. *et al*. *Aveloz-Euphorbiatirucalli*: Toxicidade da planta. **Braz J Nat Sci**. v. 1, p. 1-9, 2019.

RODRIGUES, J. G. M. *et al*. The immunomodulatory activity of *Chenopodium ambrosioides* reduces the parasite burden and hepatic granulomatous inflammation in *Schistosoma mansoni*-infection. **J Ethnopharmacol.** v. 264, p. 113287, 2020.

SCOLARI, F.*et al*. *Aedes* spp. and their microbiota: A review. **Front Microbiol**. v. 10, p. 2036, 2019.

SILVA ALVES, R. R. *et al*. Antiparasitic effects of ethanolic extracts of *Piper arboreum* and *Jatropha gossypiifolia* leaves on cercariae and adult worms of *Schistosoma mansoni*. **Parasitology**. v.147, n.14, p. 1689-1699, 2020.

SILVÉRIO, M. R. S. *et al*. Plant natural products for the control of *Aedes aegypti*: The main vector of important arboviruses. **Molecules**. v. 25, n. 15, p. 3484, 2020.

TERRA, M. R. *et al*. *Aedes Aegypti* e as Arboviroses Emergentes no Brasil. **Uningá Rev**. v. 30, n. 3, p. 52-60, 2017.

WHO - World Health Organization. **Dengue and Severe Dengue**. 2022. Available from: https://www.who.int/news-room/factsheets/detail/dengue-and-severe-dengue. Access in: 12 june 2025

WHO - World Health Organization. Guidelines for laboratory and field testing of mosquito larvicides. WHO, Geneva, Switzerland, WHO/CDS/WHOPES/GCD PP/1.3, 2005. Available from: https://www.who.int/publications/i/item/WHO-CDS-WHOPES-GCDPP-2005 .13. Access in: 04 January. 2024.

WHO - World Health Organization. Vector-Borne Disease. 2024. Available from: https://www. who.int/news-room/fact-sheets/detail/vector-borne-diseases. Access in: 12 June 2025.

YIMTCHUI, M. T. *et al*. Bioassay-directed isolation of antiplasmodial alkaloids and lignans from the dry seeds of *Piper guineense* Schumach. & Thonn (Piperaceae) and their cytotoxicity. **S Afr J Bot.** v.173, p. 225-231, 2024.

ZARA, A. L. S. A. *et al*. Estratégias de controle do *Aedes aegypti*: uma revisão. **Epidemiol Serv Saúde**. v. 25, n. 2, p. 391-404, 2016.

ZULFA, R. *et al*. Updating the insecticide resistance status of *Aedes aegypti* and *Aedes albopictus* in Asia: a systematic review and meta-analysis. **Trop med infect dis.** v. 7, n. 10, p. 306, 2022.5

1 Graduada em Ciências Biológicas. Programa de Pós-Graduação em Ciências Agrárias, Universidade Estadual do Maranhão, MA, Brasil; ORCID: 0000-0002-4779-9361. E-mail: tamiresmouralima2016@gmail.com

2 Graduado em Ciências Biológicas. Programa de Pós-Graduação em Ecologia e Conservação da Biodiversidade, Departamento de Biologia, Universidade Estadual do Maranhão, São Luís, Brasil; ORCID: 0000-0002-4506-242X. E-mail: kaiobiolog202@gmail.com

3 Graduado em Ciências Biológicas. Instituto Federal de Educação, Ciência e Tecnologia do Maranhão, Departamento de Educação, Campus São Raimundo das Mangabeiras, São Raimundo das Mangabeiras, MA, Brasil; ORCID: 0000-0003-0383-2067. E-mail: martinslimamarcos@gmail.com

4 Graduado em Ciências Biológicas. Mestre em Biologia Parasitária.Programa de Pós-Graduação em Biologia Parasitária, Centro de Biociências, Universidade Federal do Rio Grande do Norte, RN, Brasil. ORCID: 0009-0004-9161-1081. E-mail: odaiirqueiroz541@gmail.com

5 Graduada em Ciências Biológicas, Mestre em Ciências da Saúde. Programa de Pós-Graduação em Ciências da Saúde, Universidade Federal do Maranhão, São Luís, Brasil; ORCID: 0000-0002-9470-0918. E-mail: ranielly.an@hotmail.com

6 Graduada em Ciências Biológicas, Mestre em Ciências da Saúde. Programa de Pós-Graduação em Ciências da Saúde, Universidade Federal do Maranhão, São Luís, Brasil; ORCID: 0000-0003-0380-2167. E-mail: gabrielasampaiolira@yahoo.com.br

7 Graduado em Ciências Biológicas, Doutor em Parasitologia. Programa de Pós-Graduação em Ecologia e Conservação da Biodiversidade, Departamento de Biologia, Universidade Estadual do Maranhão, São Luís, Brasil; ORCID: 0000-0003-0920-4977. E-mail: gustavorodrigues_98@hotmail.com

8 Graduado em Ciências Biológicas. Programa de Pós-Graduação em Biodiversidade e Conservação, Universidade Federal do Piauí, Floriano, Brasil. ORCID: 0000-0003-3486-9639. E-mail: rafael.loreto12@gmail.com;

9 Graduado em Ciências Biológicas, Mestre em Biologia Animal. Instituto Federal de Educação, Ciência e Tecnologia do Maranhão, Departamento de Educação, Campus São Raimundo das Mangabeiras, MA, Brasil; ORCID: 0000-0002-0325-6605. E-mail: thiago.soares@ifma.edu.br

10 Graduado em Ciências Biológicas, Doutor em Parasitologia. Instituto Federal de Educação, Ciência e Tecnologia do Maranhão, Departamento de Educação, Campus São Raimundo das Mangabeiras, MA, Brasil; mirandagsbio@gmail.com; 0000-0003-3763-2776.

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