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BIOLOGICAL CONTROL OF *Aedes aegypti mosquito* WITH NATURAL PRODUCTS: A SYSTEMATIC REVIEW AND META-ANALYSES OF THE LITERATURE

Raphael da Silva Affonso
Faculdade Anhanguera do Rio Grande (FARS), Rio Grande – RS, Brazil

Lucas Santos de Sousa
Faculdade Anhanguera de Brasília (FAB) – Unidade Taguatinga Shopping – QS 1-40 – Taguatinga Sul, Brasília – DF, Brazil

Rosângela Maria Rodrigues
Parasitology Laboratory, Institution: "Universidade Federal de Jataí", Jataí, Goiás, Brazil

Ana Cristina Rodrigues da Cruz
Institution: "Universidade Federal de Viçosa" (UFV) – Department of Entomology, Viçosa, MG, Brazil

Giovanna M. C. Lemos Caramaschi
Faculdade Anhanguera de Brasília (FAB) – Unidade Taguatinga Shopping – QS 1-40 – Taguatinga Sul, Brasília – DF, Brazil

Joselita Brandão de Sant'Anna
Faculdade Anhanguera de Brasília (FAB) – Unidade Taguatinga Shopping – QS 1-40 – Taguatinga Sul, Brasília – DF, Brazil

Larissa Leite Barboza
Faculdade Anhanguera do Rio Grande (FARS), Rio Grande – RS, Brazil

Raylene Andrade Oliveira
Postgraduate Course in Medical Sciences, medicine course, Institution: "Universidade de Brasília", Brasília, Distrito Federal, Brazil

Patrícia Gomes de Assis
Laboratory of Medical Parasitology and Biological Vectors, medicine course, Institution: "Universidade de Brasília", Campus Darcy Ribeiro, Asa Norte, Brasília, DF, Brazil

Joselita Eleuza Rodrigues Machado
Nutrition course, da Faculdade Anhanguera de Brasília, Águas Claras, DF, Brasília, DF, Brazil

Abstract: Since a long time ago, *Aedes aegypti* (Diptera: Culicidae) is part of the catalog of the greatest threats to public health in the world, due to its ability to transmit important arboviruses, such as dengue, chikungunya, zika and yellow fever. Due to its high powers of dissemination and resistance, the mosquito still survives in the face of the usual methods aimed at its eradication, especially in underdeveloped countries. **Goal:** to develop a systematic review of the literature on the control of the A. aegypti mosquito, with an emphasis on natural products. **Methodology:** references were obtained through the following international databases: PubMed, SciELO, Bireme and Google Scholar. In the search, the keywords were used: *A. aegypti*; Control; Natural products; Arboviruses; Vector. The incorporation of references in this review followed the criteria: a) select articles with text available in full and free of charge, such as: theses, dissertations, course conclusion works, books on entomology and medicinal plants that have specificities related to the theme; a) disregard the year of publication (no time frame); c) opt for works written in the following languages: English, Spanish and Portuguese (BR). **Conclusions:** related to the traditionally used control methods of A. aegypti, plant extracts, such as essential oil from cloves, are a viable alternative, with proven insecticidal potential, without causing damage to the environment and human health, in addition to being economically viable. However, it is essential that new and detailed studies be carried out, so that the use of natural products is, in fact, put into practice, preferably on a large scale.
Keywords: *A. aegypti*; Vectors; Arboviruses; Combat methods; plant extracts.

INTRODUCTION

According to data from the World Health Organization (WHO), pathologies transmitted by vectors represent 17% of the global burden of infectious diseases (CANNON, et al., 2021). Mosquitoes are among the main transmitters of these diseases and, despite their fragile and harmless appearance, they are directly responsible for most of the millions of human deaths that occur every year (). In 2015, only malaria, transmitted by mosquitoes: *Anopheles* spp., reached 212 million cases worldwide (429,000 deaths) against 2.4 million cases of snakebite envenomation (100,000 deaths) and 98 cases of shark attacks (6 deaths) (WHO, 2013, 2016a; UNIVERSITY OF FLORIDA, 2017; MAIER, et al, 2019).

Vector-borne diseases represent a major threat to global health, as they are caused by parasites, transmitted to humans especially by insects. The main vector-borne global diseases to humans is extensive and includes: malaria, dengue, lymphatic filariasis, schistosomiasis, Chikungunya, onchocerciasis, Chagas disease, leishmaniasis, Zika, yellow fever and Japanese encephalitis. These diseases have a global distribution, with a higher incidence in less developed countries, where income concentration and poverty predominate. It is estimated that 80% of the world's population live in risk areas under threat of infection by at least one important vector in the transmission of diseases, and that more than half live with the threat of infection by two or more vectors (WHO, 2014; WHO, 2017).

In Brazil, the main mosquito vectors of diseases are sandflies, transmitters of leishmaniasis.; *Anopheles* spp.; and *A. aegypti*, which stands out for transmitting different diseases, such as dengue, chikungunya, zika and yellow fever. Epidemics led by *A. aegypti* have been reported since 1986. These facts show that public health policies have faced difficulties in definitively controlling these vectors for decades (BRAGA; VALE, 2007; UNIVERSITY OF FLORIDA, 2017; JOHNSON, et al., 2020).

Thus, the motivation for carrying out this work stemmed from the panorama presented above, with the fundamental objective of building a systematic review of the literature on the biological control of the mosquito: *A. aegypti*, with an emphasis on natural products.

METHODOLOGY

A literature review research on biological mosquito control was carried out: *A. aegypti*, with natural products.

For this purpose, a search was made for scientific articles in the following international databases: *National Library of Medicine* (PubMed), *Scientific Eletronic Library Online* (SciELO), Regional Library of Medicine (Bireme), and Google Scholar (*Google Scholar*). In the research, the descriptors were used: *Aedes aegypti,* Control, Natural products, Arboviruses, Vector.

From an advanced search, in each database, these descriptors were combined using the "AND" or e operator.

The incorporation of references in this review followed the criteria: a) select articles with text available in full and free of charge, such as: theses, dissertations, course conclusion works, books on entomology and medicinal plants that have specificities related to the theme; b) disregard the year of publication (without chronological demarcation); c) opt for works written in the following languages: English, Spanish and Portuguese (BR).

Experience reports and duplicate articles were excluded. After the aforementioned steps, the articles were read by title and abstract, to verify the compatibility between the information and the theme proposed here. The articles that presented agreement were read and included in the collection of references.

RESULTS

Considering the selected references as a parameter, the following were outlined: the sanitary importance and the main morphophysiological, anatomical, taxonomic and behavioral characteristics belonging to the *A. aegypti*, creating parallels with similar species; and also, with the insertion of statistical data, the entire retrospective of the mosquito that took place in Brazilian territory. In addition, conventional and alternative methods of combating the insect were discussed in depth, with emphasis on the essential oil extracted from cloves, which had been the subject of successful past studies.

EPIDEMIOLOGY OF DISEASES TRANSMITTED BY *A. aegypti* IN THE WORLD AND IN BRAZIL

The most relevant vector and metaxenic diseases (when the vector not only carries the etiological agent, but is also a mandatory element for its maturation or multiplication) in Brazil are: dengue, malaria, schistosomiasis, leishmaniasis, yellow fever, filariasis and Nasturtium. Among these, dengue and malaria programs receive the most government resources (NEVES, 2006; TAUIL, 2006; SOCIEDADE BRAZILEIRA DE PARASITOLOGIA, 2017; LEE, et al., 2018).

The relationship between Brazil and the epidemics associated with metaxenic diseases, especially those transmitted by mosquitoes, goes back a long way. Between 1928 and 1929, in Rio de Janeiro, there was a large urban yellow fever (YF) epidemic, which resulted in 478 deaths. In the following decades, from 1930 to 1940, Brazil took part in a large campaign to eradicate the mosquito: *A. aegypti* that took place across the American continent, encouraged by the Rockefeller Foundation. Finally, in 1955, the country was considered a free territory of *A. aegypti*, having its last outbreak extinguished that year, in the rural area of the municipality of Santa Terezinha, Bahia. However, in the following years, the fight against the mosquito was neglected, especially in countries in the south-central region of the American continent, such as: Venezuela, Guyana, Cuba, Haiti, Colombia and the Dominican Republic. This fact, added to failures in epidemiological surveillance by the Brazilian authorities and the disorderly growth of cities, led to a new invasion of this vector in the country in 1967, starting in the state of Pará and radiating throughout Brazil, especially in the state of Rio de Janeiro. January. Since then, the fight against the mosquito has been carried out using methods that do not provide a new eradication, as observed in 1955 (CHIEFFI, 1985; COSOLI; ROTRAUT; TEIXEIRA, 1988; CONSOLI; OLIVEIRA, 1994; BRAGA; VALLE, 2007).

In 1985, literature data warn about the risk of negligence with *A. aegypti*, which could generate new outbreaks of urban yellow fever and even a large epidemic of dengue, with the possibility of the emergence of new, more severe serotypes of the disease (CHIEFFI, 1985).

DISEASES TRANSMITTED BY *A. aegypti* IN BRAZIL

In retrospect, based on data published by HEALTH MINISTRY (MS) in the last two years (2020 and 2021), it is clear that there has been a drastic reduction in cases of dengue, Chikungunya and Zika. However, this is not necessarily due to the effectiveness of combating the mosquito vector, but rather to the occurrence of the pandemic caused by the SARS-CoV-2 virus (Coronavirus), whose spread was devastating, causing millions of cases and deaths, overshadowing the diseases transmitted by the virus *A. aegypti*. Despite this, mid-2021 and early 2022 bring substantial numbers of diseases transmitted by this mosquito. (HEALTH MINISTRY, 2021; HEALTH MINISTRY, 2022).

According to data from the literature, by mid-2021, there were about 348,508 probable cases of dengue registered in Brazil, with a reduction of 57.4%, compared to the year 2020, with 362 cases per 100,000 inhabitants, in the Central- West with the highest index. The other regions presented, in descending order, the following numbers: South (207.6 cases per 100 thousand inhabitants), Southeast (177.7 cases per 100 thousand inhabitants), North (129.4 cases per 100 thousand inhabitants) and Northeast (76.2 cases per 100,000 inhabitants), with a total of 100 confirmed deaths (MINISTRY OF HEALTH, 2021).

And also in 2021, nationwide, there were 36,242 probable cases of Chikungunya with 17 cases per 100,000 inhabitants. Also, for these diseases there was a reduction of 18.7% in the incidence, compared to the year 2020, with the Northeast being the region with the highest cases of infections, with about 30 cases per 100,000 inhabitants. The Southeast region ranked second, with 20 cases per 100,000 inhabitants, and the North in third, with approximately three cases per 100,000 inhabitants, and 15 deaths (HEALTH MINISTRY, 2021).

Finally, they registered about two thousand probable cases of Zika, with 0.9 cases per 100 thousand inhabitants, and a reduction of 43.3%, compared to 2020, with no deaths (HEALTH MINISTRY, 2021).

In the year 2022, until the month of June, there were about 1,036,505 cases of dengue registered, with a rate of 486 cases per 100,000 inhabitants. Compared to the same month of 2021, there was a significant increase of 191.3%. Similarly to that year, the Midwest region gained prominence, with a rate of 1,473 cases per 100,000 people. The other areas presented the numbers: South (884 cases per 100,000 inhabitants), Southeast (394.4 cases per 100,000 citizens), North (202 cases per 100,000 people), and Northeast (225.3 cases per 100,000 inhabitants), and with about 438 deaths (MINISTRY OF HEALTH, 2022).

Comparing the same period of 2021 with 2022, the incidence of Chikungunya cases increased radically (91.5%), with 98,540 records (46.2 cases per 100,000 people). With 140 cases per 100,000 inhabitants, the Northeast region has the highest rate of patients, above the Midwest, with 25.8 cases per 100,000 people, and the North, with 19 cases per 100,000 inhabitants, and 18 deaths reported (MINISTRY OF HEALTH, 2022).

Until the month of June 2022, 4,839 cases of Zika were detected, corresponding to 2.3 cases per 100 thousand citizens, showing a growth of 102.1%, in relation to the previous year, however, no death was notified (MINISTÉRIO DA HEALTH, 2022).

ANATOMICAL AND PHYSIOLOGICAL FEATURES OF *A. aegypti*

A. aegypti is an insect of the order Diptera and family Culicidae, which includes 95 genera divided into 3,700 species. Among them: *Culex*, which is the transmitter of Nile fever (CHATTERJEE, et al., 2021), type B encephalitis (CHATTERJEE, et al., 2021), and filariasis due to: *Wuchereria bancrofti* (LUPENZA, et al., 2021), *Anopheles*, the transmitter of malaria (ADEDEJI, et al., 2020), and Aedes the transmitter of yellow fever, dengue, Chikungunya and Zika (SERVICE, 1993; LOUNIBOS; KRAMER, 2016; VAIRO, et al., 2019; ONG, et al, 2021).

The name Aedes comes from the Greek: hateful, detestable, and the species: *A. aegypti* derives from latin: *aegyptus*, which had its origin in Ethiopia, Africa, and described, for the first time, by Carolus Linnaeus (1762), in Egypt. In the period of the great navigations, *A. aegypti* m quickly migrated to the old world and, later, together with the colonizers and their slave ships, it reached the Americas. Currently, they are described as cosmopolitan mosquitoes, with greater occurrence in the subtropical and tropical regions of the world (CONSOLI; OLIVEIRA, 1994; BESERRA, et al., 2006).

A. aegypti when adult measures between 4 and 6 mm, and has a small spherical head, from which antennae with a long flagellum emerge. It has sensory structures in the oral apparatus, responsible for balance, known as palps. They have an acute abdomen, with no spiracular bristles, and the anal and genital orifices are visible in the eighth segment. It is a dark insect with white stripes, and at the base of the dorsal segment, over the mesonotum, it has a characteristic design, which may or may not be evident, in the form of a "lyre" whose "strings" have narrower silver-white elements in two fine lines (NELSON, 1986; ALMEIDA, 2011).

From the thorax project three pairs of legs, two respiratory holes and the pair of membranous wings, narrow and long, with ribs covered in scales, as well as the dumbbells (second pair of modified wings), which, during flight, help in the balance. The male is distinguished from the female by: being smaller, having longer palps, having plumier antennae and feeding on sap and nectar. The difference in size is due to the fact that the female is responsible for the production and storage of eggs, and also for the accumulation of blood from one or more meals (NELSON, 1986; ALMEIDA, 2011; SOUSA, et al., 2022).

Feeding of *A. aegypti*

Male and female mosquitoes have energy metabolism, so they need carbohydrates from flowers, fruits and saps. However, the accumulation of triglycerides and glycogen, which are more difficult to obtain from plant sources, is crucial for the development of some potential activities, such as reproduction. Thus, they need a blood meal from the females, without which their reproductive capacity would be compromised, that is, the development of eggs. Thus, only females have hematophagous characteristics. They are fast and persistent suckers, a fact that reinforces their characteristic of vector of a series of diseases (SERVICE, 1993; PERDOMO, et al., 2021).

In most Aedes species, to perform the first egg laying, the female needs at least two blood meals. However, in other reproductive cycles, posture is usually performed after each meal. The species A. aegypti and others can promote multiple meals, replacing sugary food and, therefore, contributing to increase the longevity of females, as they obtain a greater amount of nutrients from the blood (COSOLI; OLIVEIRA, 1994; PERDOMO, et al., 2021).

Females have a diurnal habit, with a predilection for the early hours of the morning (6:00 am to 8:00 am) and late afternoon (4:00 pm to 6:00 pm), when humidity and temperature are more conducive to feeding and oviposition. The radius of action of A. aegypti is about 100 meters from the egg laying site, however, there are records of dispersion of up to 800 meters (COSOLI; OLIVEIRA, 1994; REITER, 1996; PERDOMO, et al., 2021). After landing, the females select the bite site and introduce the proboscis, a piercing-sucking mouthpart with high piercing capacity. During the sucking process, saliva, which contains hundreds of digestive proteins, is expelled and inoculated through a formed channel. This secretion has vasodilator substances, whose function is to increase the local blood supply, anticoagulants and antiplatelet agents, to prevent blood clotting and thus the obstruction of the mouth canal of the mosquito during the meal. The contact between insect saliva and human blood is crucial for the introduction of viruses, or even other etiological agents, into the host organism. The allergenic characteristics, inherent to the bites, are also given by the action of saliva at that moment (ALMEIDA, 2011; SUN, et al., 2020).

Reproduction of *A. aegypti*

They are popularly known as stilt, were classified as stenogamous, however, studies show their participation in swarms in nature (CONSOLI, OLIVEIRA, 1994; LANG, et al., 2018). Swarms can be composed of more than one species, and the factors for their formation consist of the most varied possible, such as: luminosity, physical references (a pole or a tree, for example), physiological state of the mosquitoes, air currents, among others. others (SERVICE, 1993).

The species of *A. aegypti* and *Culex quinquefasciatus* (popularly known as stilt or muriçoca), were classified as stenogamous, however, studies show their participation in swarms in nature (CONSOLI, OLIVEIRA, 1994; LANG, et al., 2018). Swarms can be composed of more than one species, and the factors for their formation consist of the most varied possible, such as: luminosity, physical references (a pole or a tree, for example), physiological state of the mosquitoes, air currents, among others. others (SERVICE, 1993).

Copulation can occur before or after the ingestion of the first blood meal, being more common before. Intraspecific mating is rare, but such a cross has been reported between: *A. albopictus* and *A. aegypti* (NASCI, et al., 1989).

Oviposition of *A. aegypti*

At the end of copulation, the spermatozoa are stored in the spermathecae, so that they can gradually be used to fertilize the eggs when they are being deposited. Male gametes have long-lasting viability. Studies indicate that the species: *A. fluviatilis* performed up to 15 spawnings fertilized by spermatozoa stored before the first spawning. There are also indications that in some species of mosquitoes, the type of blood ingested can influence the size of the spawn (CLEMENTS, 1963; FORATTINI et al., 1989).

Inside the female's abdomen, when the eggs reach maturity, they are directed to the oviduct, which initiates a series of contractions, with the aim of expelling them. Fertilization takes place in the oviduct, and immediately after, oviposition occurs. However, retention of eggs may occur due to the absence of favorable conditions for their development. For mosquitoes of the genus Aedes, fertility declines vigorously when eggs are retained for more than 15 days (CONSOLI; OLIVEIRA, 1994). Physical trauma, such as decapitation, removal of wings, poisoning or even mechanical shocks can, in some species, cause premature oviposition, even in the absence of water (SERVICE, 1993).

Site selection by the female to promote oviposition is of significant relevance in determining the distribution of species in nature. This process is directly influenced by chemical factors such as the indole molecule that is identified as attractive for oviposition (BIESSMANN, et al., 2010; AFFONSO, 2012; AFFONSO, et al., 2013), and physical factors such as: intensity or absence light, wavelength reflected by the surfaces, and the color of the place may represent an attraction for mosquitoes, temperature, degree of salinity and biological factors such as the presence of plants or microorganisms (REITER, 1996).

Given its exposure during the process, oviposition is a critical moment in the female's life cycle, which can present a laying volume of between 50 and 500 eggs, released in the so-called laying cycles. The genus Aedes lays eggs close to the liquid environment, that is, on the edges of water puddles, and the larval form will only hatch from the egg when it is hit by nearby water. The eggs remain viable for long periods of desiccation, for months or years (FORATTINI, 2002).

The eggs are elliptical in shape and initially white in color, which later changes to black when in contact with oxygen, over a period of approximately two hours. The embryos, under adequate environmental conditions, complete the embryonic development, that is, the diapause of the egg, with subsequent hatching after submersion in the liquid medium, with a decrease in the oxygen supply, in the period of 2 to 3 days on average, starting to the larval stage, which is strictly aquatic. Classically, it is assumed that mosquitoes prefer to lay their eggs in a place with clean water, however, there is a wide variety of breeding sites that also have polluted water (GRANDE, et al., 2006; BARRETO, et al., 2006); IOC/FIOCRUZ, 2017).

Larval stage of *A. aegypti*

The larvae that emerge after hatching from the eggs have four stages: L1, L2, L3 and L4, which can be distinguished by the successive shedding of the exoskeleton (ecdysis) that favor the growth of the larva. The average period of the entire larval stage ranges from 5 to 10 days. The larvae, composed of head, thorax and abdomen, feed on organic waste, in general, by filtering the water, but they can crush, bite or even scrape surfaces or other bodies. The larval forms of *A. aegypti, they* exhibit photophobia, which promotes a characteristic serpentine movement (in an "S") of flight to more distant places from the surface, serving as a parameter for the identification of the larvae (FORATTINI, 1986; FORATTINI, et al., 1998).

The last immature stage of the mosquito's life cycle is the pupa, which, like the larval stage, is also aquatic. This stage consists of a transition period to the winged (adult) phase of the mosquito, lasting about 1 to 3 days. The pupae remain breathing on the surface of the container where they were in the larval phase, and when there is movement of water, they flee. At this stage, food intake does not occur, as the insect's metabolism is only focused on its complete maturation. During this period, some larval organs are eliminated, as well as the formation of new essential structures for adult development. The male pupae are smaller and hatch before the female ones (TAVEIRA, et al., 2001).

Sensory activity of *A. aegypti*

A. aegypti has 2 large eyes composed of vision units, between 421 and 492 units, called ommatidia (SNOW, 1971; TEIXEIRA, 1988). These eyes cover the head at an angle of 225°. The yellow-green (420 to 620 nm) and ultraviolet (340 to 370 nm) regions of the light spectrum correspond to the regions of greatest light sensitivity for mosquitoes (SNOW, 1971).

Mosquito attraction

Mosquitoes can be attracted by visual, sound, thermal and olfactory stimuli. It is from these mechanisms that they locate their targets, however, the details involved in this sensory scope are still not fully understood (TAKKEN; KLINE, 1989; TAKKEN; KNOLS, 1999; RIBAS; CARREÑO, 2010; AFFONSO, et al., 2012; TSITSANOU, et al., 2012).

According to Clements (1963), male mosquitoes can be attracted by the vibratory frequency of the beating of the wings of females of the same species. Anatomical portions, such as the flagellum and the "Johnston's organ", are responsible for capturing and amplifying the sound of mosquitoes, respectively.

In principle, the molecules are carried by the air and penetrate the cuticular pores of mosquitoes, and then they are captured by odorant agent binding proteins (PLO) and transported to olfactory receptors located in the dendritic membranes of neurons that are housed in the antennae, more specifically in the sensory cords, where they are detected (THOMSEN; CHRISTENSEN, 2006; BIESSMANN, et al., 2010; TSITSANOU, et al., 2012; AFFONSO, et al., 2012; AFFONSO, et al., 2013).

PLOs are the first line of smell recognition in the mosquito organism. Among the 60 categories of PLOs encoded in the genome of: *Anopheles gambiae,* the *Ag*OBP$_1$ is more abundant on the antennae of females compared to males. The profusion is reduced when females feed on blood. In this sense, the ability of females to detect a potential target for feeding is associated with PLOs (BIESSMANN, et al., 2010; AFFONSO, et al., 2012; TSITSANOU, et al., 2012).

The natural binder of *Ag*OBP$_1$ has not yet been identified, however, it is known that this dimeric protein has an unusual cavity, which is a tunnel that connects its two monomers. The global chain has six helices connected by loops and with a hydrophobic pocket similar to other PLOs (WOGULIS, et al., 2006).

The polymer formed by ethylene glycol, PEG (polyethylene glycol) is able to make hydrophobic contacts with the cavity of the *Ag*OBP$_1$ and some polar contacts mediated by water molecules. Polar atoms from the side chains of residues Tyr10, Ser79, His111 and Trp114 are exposed at the protein binding site, as well as the carbonyl of Phe123 and the amino group of Val125, and the protein is opened to the solvent in the space created by the helices 1, 3 and 4. This door is opened by residues Leu15, Ala18, Leu22, Ala62, Val64 and Leu73 (WOGULIS, et al., 2006).

Together with the DEET (N, N-diethyl-3-methylbenzamide) molecule, Tsitsanou, et al. (2012) crystallized AgOBP1 and performed a molecular dynamics (DM) study. The results show that, despite the extensive hydrophobic cavity that connects the two protein monomers, the DEET molecule has an affinity for the marginal region of the tunnel, close to the connection between the two monomers.

The DEET molecule makes 57 van der Waals interactions, mostly with non-polar atoms. In addition to these interactions, there is a hydrogen bond between the carboxylic oxygen of DEET and a water molecule, which, in turn, interacts with the nitrogen in the ring of Trp114 and with Gly 92 or Cys 95, alternately (ABD-ELLA, et al. al., 2015).

The potential for interaction between the major compounds of the extract of *S. aromaticum* and the protein: *Ag*OBP$_1$, in a previous work (AFFONSO, et al., 2013), docking and molecular dynamics (DM) studies on the crystallographic structure of AgOBP1 showed that eugenyl acetate and eugenol had interactions with AgOBP1 comparable to those of DEET. The results led to the proposition that eugenol and eugenyl acetate have the same mechanism of action as DEET, being potential repellents against mosquitoes that have PLOs proteins (ABDEL-RAHMAN, et al., 2004).

The protein: *Ag*OBP$_1$ available in the protein data bank (PDB), under the code 2ERB, shows 82% similarity with the mosquito protein OBP1: *A. aegypti* and also a 91% identity among the DEET binding site residues. This similarity value, especially in the region corresponding to the binding tunnel, between residues 59 and 119, is sufficient for the inhibition results obtained for AgOBP1 to be extrapolated for AaOBP1

(LEITE, et al., 2009; MAO, et al., 2010; PROTEIN DATA BANK, 2017; UNIPORT, 2017).

COMBATTING THE MOSQUITO

Until 2001, and with the aim of eradicating the mosquito from Brazil, the Federal Government created a series of plans and projects, none of which achieved the expected success. In the aforementioned year, after the failure of the A. aegypti Eradication Plan (PEA), launched in 1996, it was decided to change strategy and focus only on controlling this vector, to the detriment of its eradication. Thus, the Dengue Control Action Intensification Plan (PIACD) emerged, which evolved into the current National Dengue Control Plan (PNCD) (VALLE; BRAGA, 2007; FIGUEIRÓ, et al., 2010).

When talking about reducing mosquito damage, the term “vector control” is most appropriate. This control is an important challenge in underdeveloped countries, which have a low allocation of resources for this purpose (VALLE; BRAGA, 2007; FIGUEIRÓ, et al., 2010).

In the model proposed by the PNCD, community agents and agents to combat endemic diseases (arms of the State) act together with the local population, promoting chemical and mechanical vector control actions. These actions focus on the detection and removal of potential mosquito breeding sites, via an active search, in particular, for stagnant water reservoirs (mechanical control); in the use of pathogens or predators that naturally reduce the population of larvae, without harming the environment or human health (biological control); and the use of chemical products capable of eliminating larval (larvicides) or adult (insecticides or adulticides) forms, characterizing chemical control (ZARA, et al., 2016).

Other strategies (Table 1) have been adopted as a way to increase the effectiveness of mosquito control. The following stand out: the release of transgenic male mosquitoes (ARAÚJO, et al., 2015; CARVALHO, et al., 2015); sterilization by small doses of gamma radiation on male mosquitoes (FERREIRA, et al., 2008; ALPHEY, et al., 2010); and biological control using bacteria: *Wolbachia*,

Technology	Mechanism	Benefits	Limitations
Abordagem eco-bio-social	Social participation in vector control using mechanical tools.	Eliminates the use of insecticides.	Depends on the involvement of various sectors of society.
Compostos naturais	Larvicidal and bacterial activity.	Alternative to chemical control; uses safer insecticides.	There is a need for efficacy and cost-effectiveness studies compared to chemical control.
Wolbachia	The bacterium causes sterility and reduces the transmission of arboviruses.	Self-sustainable; no sexing of mosquitoes; does not use insecticides and radiation.	Climate differences, mosquito release protocols, level of urbanization and human density can limit the invasive potential of insects at release sites.
Mosquitos transgênicos	Production of lethal genes, sterilization of mosquitoes, or introduction of a gene that reduces or blocks disease transmission.	Reduces the lifespan of mosquitoes; no use of radiation.	Replacement by wild mosquito population may occur over time.
Irradiação	Sterilization of insects by irradiation.	Reduces the risk of replacement by wild mosquitoes and transmission of pathogens.	Same technology limitations with the use of *Wolbachia*.

Table 1. New strategies used in the control of biological vectors of infectious diseases.

Adapted from ZARA, et al., 2016.

which halves the lifetime of the mosquito and prevents the transmission of viruses: DENV, CHIKV and ZIKV (MCMENIMAN, et al., 2009; YEAP, et al., 2011).

Insecticidal and repellent agents

Synthetic insecticidal agents are widely used to control vectors that attack agriculture, livestock and humans. However, the perennial and abusive use of these compounds causes damage to human health and the environment. This fact causes health authorities to create bureaucratic barriers in the use and registration of synthetic compounds with insecticidal characteristics (JUMBO, 2013; MEKALI, et al., 2013; WAKIL, et al., 2013).

Organophosphates (OF), along with pyrethroids (PI), make up the chemical class most used as an insecticide agent. However, many authors have verified significant side effects of OP in humans (JUMBO, 2013), such as: Alzheimer's disease (ZAGANAS, et al., 2013), diabetes (EVERETT; MATHESON, 2010), damage to the immune and endocrine systems (CORSINI, et al., 2013; MOSTAFALOU, ABDOLLAHI, 2013), in addition to the known activity on acetylcholinesterase (AChE) and butyrylcholinesterase (BChE) enzymes (KAVVALAKIS; TSATSAKIS, 2012). PI, in turn, are immunotoxic to humans (ZHANG, et al., 2010), and inhibit the action of progesterone (MCKINLAY, et al., 2008).

In 1946, the United States Department of Agriculture, together with the armed forces, developed DEET or N-diethyl-3-methylbenzamide, a substance with low toxicity, with only 43 reports of intoxication in the last 5 decades, which can be used on the skin as an insect repellent. Today, about 30% of the North American population uses formulations containing DEET, increasing the protection time, depending on the concentration (KATZ, et al., 2008; AFFONSO, et al., 2012). DEET's mechanism of action consists of providing a vapor barrier that prevents the insect from coming into contact with the skin, due to the offensive odor exhaled and perceived only by the vector, due to its low boiling point (KATZ, et al., 2008; World Health Organization, 2009).

The concomitant use of DEET with retinoid products, which are widely used in the composition of creams and lotions for the treatment of acne, for example, can lead to an increase in the toxicity of this substance. However, the use in sunscreens leads to an increase of up to 6 times in the systemic absorption, generating a greater chance of intoxication by DEET (KATZ, et al., 2008).

Other studies claim that DEET causes a decrease in the protection factor of sunscreens, thus offering a false sense of security against ultraviolet rays (KATZ, et al., 2008). This result is relevant, mainly due to the use of this interaction (DEET and sunscreens) in endemic regions of malaria and other diseases transmitted by mosquitoes, such as the Amazon region (RIBAS; CARREÑO, 2010).

Data from the literature suggest that continuous exposure to DEET leads to sensorimotor deficits associated with changes in the cholinergic system, part of the central nervous system. According to the authors, there was a significant reduction in ache activity after treatment with DEET (ABOU-DONIA, et al., 2004).

Picaridin, icaridine or 1-methylpropyl 2-(2-hydroxyethyl)-1-piperidine carboxylate, is a synthetic compound derived from pepper that exhibits insect repellent activity comparable to the activity of DEET. The use of this substance was approved in Brazil and the United States (RIBAS; CARREÑO, 2010). Marketed since 1973, picaridin acts by promoting a strong excitation of the insect's central nervous system and blocking the circulation of sodium in nerve cells, by inhibiting the synthesis of ATP, AChE and the γ-amino butyric acid receptor (GABA), causing complete paralysis of the insect (KATZ, et al., 2008; RIBAS; CARREÑO, 2010). This action, especially on ache, becomes the key to explaining the insecticidal effect of the molecules used against insects.

Considering that the mechanism of action occurs in direct contact with the insect, icaridin is not applied on the skin, but in agriculture for pest control or sprayed against lice. It has toxicity only at high doses, with neurotoxic effects, in addition to being odorless, little irritating to the skin and not damaging plastic materials, allowing it to be stored in containers based on this material (KATZ, et al., 2008; RIBAS; CARREÑO, 2010).

Another substance widely used in repellent formulations currently marketed, including by the Brazilian Army, is permethrin, a synthetic compound belonging to the PI class. This chemical has an extensive spectrum of action against insects, in addition to rapid absorption, distribution and excretion with low toxicity to humans and animals. Its mechanism of action is associated with blocking sodium channels and antagonizing GABA receptors in insects (MELO, et al., 2008).

Natural agents

Until the beginning of the 21st century, only 1% of the insecticides used in the world were of botanical origin. However, the tendency is for the observed percentage to increase in the coming decades, as discussions related to sustainability, especially the environmental one, are increasingly consolidated. This considerably increases the possibilities of plant extracts being used as a base in the production of insecticides (ROZMAN, et al., 2007).

In general, molecules of plant origin are volatile, that is, they easily change their physical state, which causes some impediment to the large-scale use of these compounds, as they have low residual power and toxicity ranging from low to moderate. Compared to synthetic substances, natural ones have a lower cost (LUCCA, 2009; AFFONSO, et al., 2012). In this context, some authors suggest a neurotoxic action of essential oils on insects, due to a multitude of factors (COATS, et al., 1991; ENAN, 2001; KOSTYUKOVSKY, et al., 2002a; PRICE; BERRY, 2006; HUIGNARD, et al., 2008; SOUSA, et al., 2022).

Some authors suggest that, in addition to the octopaminergic action, the toxic activity of essential oils in insects may be related to the inhibition of ache and cytochrome: P_{450} dependent monooxygenase (KOSTYUKOVSKY, et al., 2002b). Price and Berry (2006) demonstrated the neural activity of eugenol in ache (JUMBO, 2013), which was also pointed out by Corbel et al., (2009) as an important target to be inhibited by DEET during its insecticidal activity.

Monoterpenes such as eugenol are found in abundance in essential oils. Some studies associate these lipophilic compounds with neurotoxic activities that reach the *A. aegypti* (LUCCA, 2009; GAO, et al., 2009; AFFONSO, 2012; AFFONSO, et al., 2014).

1. Syzygium aromaticum (Indian clove)

Oils derived from natural products that have the greatest potential as insect repellents are citronella, clove, verbena, cedarwood, lavender, pine, cinnamon, rosemary, basil, pepper, and allspice (TAJUDDIN, et al, 2004). Indian clove extract was discussed in a previous work carried out by our research group, as a new and potential low-cost repellent, therefore accessible to low-income populations, which are the most affected by diseases transmitted by insects (AFFONSO, 2012; AFFONSO, et al., 2013; AFFONSO, et al., 2014; SOONWERA; PHASOMKUSOLSIL, 2016).

The Indian clove tree belongs to the myrtaceae family (Myrtaceae), identified by the name of *Syzygium aromaticum* [L] Merr. et Perry. Historical reports on the use of this plant, in addition to being a culinary spice, report repellency, antimicrobial, antifungal, anti-ulcer, anesthetic, anti-inflammatory, antitumor, antidiabetic and aphrodisiac activities (RABÊLO, 2010; RANA, et al., 2011; RADÜNZ, et al., 2019; KAČÁNIOVÁ, et al., 2021).

In a recent work (AFFONSO, et al., 2014) developed a methodology by gas chromatography coupled to a flame ionization detector (GC-DIC), to quantify and qualify the compounds extracted from a sample of macerated cloves, in a way static for 4 days. The results showed that the eugenol and eugenyl acetate molecules are the ones present in higher concentrations in the ethanolic extracts of the flower buds of the *S. aromaticum*. This result corroborates the data described in the literature, regarding the chemical composition of the volatile compounds extracted from *S. aromaticum*: leaves, stems and flower buds, which show that eugenol and eugenyl acetate are the main components of the extract, as shown in Table 2 (PAWAR; THAKER, 2006; PRASHAR, et al., 2006; SANTORO, et al., 2007; SCHERER, et al., 2009; GAO, et al., 2009; OLIVEIRA, et al., 2009; POLITEO, et al., 2010; AFFONSO, et al., 2014).

2. Eugenol insecticidal activity

Molecular modeling results suggest that eugenol can be classified as an active molecule, with larvicidal activity on: *A. aegypti* (SANTOS, et al., 2009). In other molecular modeling studies, it was observed that eugenol and eugenyl acetate have a strong molecular interaction with the AgOBP1 protein, which is present in females of the genus: *Anopheles*, being responsible for triggering the feeding process of hematophagous arthropods vectors of malaria (AFFONSO, 2012; AFFONSO, et al., 2013).

Products containing clove essential oil, in concentrations of 10 or 20% and 10% of makaen (*Limonella zanthoxylum*) have been

Authors	Composed (%)				
	β-C*	**α-H****	**Eugenol**	**AE*****	**Humulen**
PAWAR; THAKER, 2006.	1,35	-	47,64	-	-
PRASHAR, et al., 2006.	13,00	-	78,00	-	-
SANTORO, et al., 2007.	8,20	-	86,34	3,58	0,83
SCHERER, et al., 2009.	10,98	1,26	83,75	-	-
GAO, et al., 2009.	1,39	0,20	88,59	5,62	0,28
OLIVEIRA, et al., 2009.	0,64	-	88,38	10,98	-
POLITEO, et al., 2010.	1,20	0,10	91,20	7,40	-
AFFONSO, et al., 2014.	-	-	89,00	11,00	-

β-C*: β- Caryophyllene; α-H**: α-Humulene; AE***: eugenyl acetate.

Table 2. Contents of compounds extracted from the extract of *S. aromaticum*, according to different authors, using gas chromatography.

tested and shown repellency against: *A. aegypti* and *Culex quinquefasciatus*. However, there was no synergism, but the mixture reduces the cost and increases the safety of the product (TRONGTOKIT, et al., 2004).

The repellent activity of carvacrol, thymol and eugenol, extracted by hydrodistillation from *Monarda fistulosa L*, used in bioassays showed that these molecules have repellent activity for *A. aegypti* (TABANCA, et al., 2013).

Other studies evaluated the larvicidal effects of seven essential oils against *A. aegypti, Anopheles dirus* and *C. quinquefasciatus* (PHASOMKUSOLSIL; SOONWERA, 2013), and the results showed that the essential oil of *S. aromaticum* was the most effective against all immature stages of the three mosquito species. These results corroborate with other studies that showed the larvicidal action of *S. aromaticum* (TEHRI; SINGH, 2015).

Table 3 shows the insecticidal activity of cloves and the compound eugenol for *A. aegypti* and other insect species.

In view of the above, homemade ethanolic extract of clove flower buds (*S. aromaticum*) can be used effectively and safely as a mosquito repellent: *A. aegypti*, in addition, studies by molecular modeling of the main components of this extract, such as: eugenol and eugenyl acetate, contributed to a better understanding of the repellency mechanism, with this they can be used to propose the structuring of new and more efficient repellents that are economically less expensive.

DISCUSSION

The method proposed by CCD proved to be adequate for the quantification and qualification of the eugenol and eugenyl

Authors	Reported activity
PLARRE, et al., 1997.	The mixture of clove oil and citronella oil proved to be effective in the short term against "clothes moth" (*Tineola bisselliella*).
PRICE; BERRY, 2006.	Eugenol is toxic against two species of cockroaches, *Periplaneta americana* and *Blaberus discoidalis*.
LUCCA, 2009.	The methanolic and hexanic extracts of *S. aromaticum* showed repellency against the corn weevil (*Sitophilus zeamais*).
CHOI, et al., 2010.	Efficacy of eucalyptus and clove oils used concomitantly against malathion and permethrin resistant lice.
ZERINGÓTA, et al., 2013.	The repellent activity of eugenol was considered promising against *Rhipicephalus microplus* (bull tick).
JUMBO, 2013.	Clove and cinnamon essential oils have an insecticidal effect against *Acanthoscelides obtectus*.
REIS, et al., 2014.	Eugenol was the best of the 5 compounds tested as an insecticide against bean weevil (*Callosobruchus maculatus*).
HUANG, et al., 2015.	Eugenol showed insecticidal and repellent effectiveness against: fire ant (*Solenopsis invicta*).
IWAMATSU, et al., 2016.	The mixture of eugenol and β-caryophyllene is the most effective for repelling body lice.

Table 3. Insecticidal activities of cloves and their components against different insect species.

acetate components in the ethanolic extract of cloves, making it one more analytical technique to be used in the evaluation of extracts from this plant. The low cost of the technique associated with the precision, selectivity, robustness and easy applicability of the proposed method, qualifies the CCD as an attractive technique for future analysis.

In this review study confirmed that the ethanolic extract of *S. aromaticum*, prepared according to the popular use (4 days of static maceration), it is effective as a repellent, with 63% repellency for: *A. aegypti*, against 20% of the positive control for DEET, which is present in commercial repellents. It is worth noting that the concentration of eugenol in the 4-day extract is 83.7 times lower than that of DEET, suggesting that the repellency potential of eugenol is much greater than that of DEET.

It was possible to observe that the male gender of the mosquito is more than the female gender by the clove repellent. This difference is significant and may be associated with a greater physiological release of lactic acid from male skin.

Regarding the anticholinesterase activity, eugenol and DEET showed similar inhibitory powers, while the ethanolic extract was able to inhibit the enzyme only at higher concentrations, since DEET does not interact with DmAChE, but interacts with EcBChE, thus, the Human toxicity observed for DEET is likely to be observed in the extract due to its low potency of ache inhibition.

Due to the versatility of the *S. aromaticum* widely reported in the scientific literature, the extract and its major compounds: eugenol and eugenyl acetate were tested based on the antibacterial activity cited in different works, with the 10-day extract being the only one that showed significant antibacterial activity against *S. aureus* e *E. coli*. Eugenol showed a halo of inhibition against both bacteria tested, being similar to the antibiotic used as a positive control (amoxicillin + potassium clavulanate). Thus, eugenol appears as the main antimicrobial agent present in the ethanolic extract.

In molecular modeling studies, six out of eleven molecules proposed as potential new repellents showed molecular docking energies with AgOBP1, more favorable than eugenyl acetate and DEET. However, DM studies have shown that two of them (E7 and DE1) can destabilize the protein, although they remain in the binding site. It is inferred, therefore, that the six selected molecules are potential leads for the development of new repellents. Additional studies are important for the isolation of these molecules, as they have synthetic viability.

FINAL CONSIDERATIONS

With regard to methods of controlling the *A. aegypti* conventionally used, vegetable extracts, such as the essential oil from cloves, are a viable alternative, with proven insecticidal potential, without causing damage to the environment and human health, in addition to being economically viable. However, it is imperative that new and detailed studies be carried out, so that the use of natural products is effectively put into practice, so that laboratory limits are exceeded, and that these substances are used in greater proportions.

REFERENCES

ABD-ELLA, A.; STANKIEWICZ, M.; MIKULSKA, K.; NOWAK, W.; PENNETIER, C.; GOULU, M.; FRUCHART-GAILLARD, C.; LICZNAR, P.; APAIRE-MARCHAIS, V.; LIST, O.; CORBEL, V.; SERVENT, D.; LAPIED, B. The repellent DEET potentiates carbamate effects via insect muscarinic receptor interactions: An alternative strategy to control insect vector-borne diseases. *PLOS ONE*, v. 10, n. 5, p. e0126406, 2015.

ABDEL-RAHMAN, A.; ABOU-DONIA, S. M.; EL-MASRY, E. M.; SHETTY, A. K.; ABOU-DONIA, M. B. Stress and combined exposure to low doses of pyridostigmine bromide, deet, and permethrin produce neurochemical and neuropathological alterations in cerebral cortex, hippocampus, and cerebellum. *Journal of Toxicology and Environmental Health*, Part A, v. 62, n. 2, p. 163–192, 2004.

ABOU-DONIA, M.; DECHKOVSKAIA, A. M.; GOLDSTEIN, L. B.; ABDEL-RAHMAN, A.; BULLMAN, S. L.; KHAN, W. A. Co-exposure to pyridostigmine bromide, DEET, and/or permethrin causes sensorimotor deficit and alterations in brain acetylcholinesterase activity. *Pharmacol Biochem Behav*., v. 77, n. 2, p. 253–62, 2004.

AFFONSO, R. DA S. 2012. *Quantificação do extrato etanólico do cravo da índia utilizado como repelente caseiro e estudos por ancoramento molecular de seus principais componentes*. Dissertação de Mestrado, Instituto Militar de Engenharia, 57p.

AFFONSO, R. DA S.; GUIMARÃES, A. P.; OLIVEIRA, A. A.; SLANA, G. B. C. A.; FRANÇA, T. C. C. Applications of molecular modeling in the design of new insect repellents targeting the odorant binding protein of *Anopheles gambiae*. *Journal of the Brazilian Chemical Society*, v. 24, n. 3, p. 473–482, 2013.

AFFONSO, R. S.; LESSA, B.; SLANA, G. B. C. A.; BARBOZA, L. L.; DE ALMEIDA, F. V.; LIMA, A. L. S.; DE SOUZA, F. R.; FRANÇA, T. C. C. Quantificação e caracterização dos principais componentes do extrato etanólico de cravo-da-Índia *Syzygium aromaticum* [1] Merr. et Perry. *Revista Virtual de Quimica*, v. 6, n. 5, p. 1316–1331, 2014.

AFFONSO, R. S.; RENNÔ, M. N.; SLANA, G. B. C. A.; FRANÇA, T. C. C. Aspectos químicos e biológicos do óleo essencial de cravo da Índia. *Revista Virtual de Quimica*, v. 4, n. 2, p. 146–161, 2012.

ALMEIDA, A. P. G. Os mosquitos (Diptera, Culicidae) e a sua importância médica em Portugal: Desafios para o século XXI. *Acta Médica Portuguesa*, v. 24, n. 6, p. 961–974, 2011.

ALPHEY, L.; BENEDICT, M.; BELLINI, R.; CLARK, G. C.; DAME, D. A.; SERVICE, M. W.; DOBSON, S. Sterile-insect methods for control of mosquito-borne diseases: An analysis. *Vector-Borne and Zoonotic Diseases*, v. 10, n. 3, p. 295–311, 2010.

ARAÚJO, H.; CARVALHO, D.; IOSHINO, R.; COSTA-DA-SILVA, A.; CAPURRO, M. *Aedes aegypti* control strategies in Brazil: Incorporation of new technologies to overcome the persistence of dengue epidemics. *Insects*, v. 6, n. 2, p. 576–594, 2015.

BESERRA, E. B.; JÚNIOR, F. P. C.; SANTOS, J. W.; SANTOS, T. S.; FERNANDES, C. R. M. Biologia e exigências térmicas de *Aedes aegypti* (L.) (Diptera : Culicidae) provenientes de quatro regiões bioclimáticas da Paraíba. *Neotropical Entomology*, v. 35, n. 6, p. 853–860, 2006.

BIESSMANN, H.; ANDRONOPOULOU, E.; BIESSMANN, M. R.; DOURIS, V.; DIMITRATOS, S. D.; ELIOPOULOS, E.; GUERIN, P. M.; IATROU, K.; JUSTICE, R. W.; KRÖBER, T.; MARINOTTI, O.; TSITOURA, P.; WOODS, D. F.; WALTER, M. F. The *Anopheles gambiae* odorant binding protein 1 (AgamOBP1) mediates indole recognition in the antennae of female mosquitoes. *PLOS ONE*, v. 5, n. 3, 2010.

BRAGA, I. A.; VALLE, D. *Aedes aegypti*: inseticidas, mecanismos de ação e resistência. *Epidemiologia e Serviços de Saúde*, v. 16, n. 4, p. 279–293, 2007.

CARVALHO, D. O.; MCKEMEY, A. R.; GARZIERA, L.; LACROIX, R.; DONNELLY, C. A.; ALPHEY, L.; MALAVASI, A.; CAPURRO, M. L. Suppression of a field population of *Aedes aegypti* in Brazil by sustained release of transgenic male mosquitoes. *PLOS Neglected Tropical Diseases*, v. 9, n. 7, p. e0003864, 2015.

GAO, X.; WANG, W.; WEI, S.; LI, W. Review of pharmacological effects of *Glycyrrhiza radix* and its bioactive compounds. *Zhongguo Zhong yao za zhi*, v. 34, n. 21, p. 2695–700, 2009.

CHIEFFI, P. P. Algumas questões decorrentes da reintrodução do *Aedes aegypti* no Brazil. *Cadernos de Saúde Pública*, v. 1, n. 3, p. 385–387, 1985.

CHOI, H.-Y.; YANG, Y.-C.; LEE, S. H.; CLARK, J. M.; AHN, Y.-J. Efficacy of spray formulations containing binary mixtures of clove and eucalyptus oils against susceptible and pyrethroid/malathion-resistant head lice (Anoplura: Pediculidae). *Journal of medical entomology*, v. 47, n. 3, p. 387–391, 2010.

CLEMENTS, A. N. The physiology of mosquitoes: International series of monographs on pure and applied biology: Zoology, Vol. 17, 1963.

COATS, J. R.; KARR, L. L.; DREWES, C. D. Toxicity and neurotoxic effects of monoterpenoids. *Naturally Occurring Pest Bioregulators*, v. 2, p.305–316, 1991.

CONSOLI, R. A. G. B.; OLIVEIRA, R. L. 1994. *Principais mosquitos de importância sanitária no Brazil*. 20 ed. Rio de Janeiro, 1994, 228p.

CORBEL, V.; STANKIEWICZ, M.; PENNETIER, C.; FOURNIER, D.; STOJAN, J.; GIRARD, E.; DIMITROV, M.; MOLGÓ, J.; HOUGARD, J. M.; LAPIED, B. Evidence for inhibition of cholinesterases in insect and mammalian nervous systems by the insect repellent deet. *BMC Biology*, v. 7, n. 1, p. 47, 2009.

CORSINI, E.; SOKOOTI, M.; GALLI, C. L.; MORETTO, A.; COLOSIO, C. Pesticide induced immunotoxicity in humans: A comprehensive review of the existing evidence. *Toxicology*, v. 307, p. 123–135, 2013.

COSOLI, ROTRAUT A. G. B.; TEIXEIRA, M. A. Influence of larval and pupal products on the oviposition behaviour of *Aedes fluviatilis* (Lutz) (Diptera: Culicidae). *Memórias do Instituto Oswaldo Cruz*, v. 83, n. 2, p. 213–218, 1988.

DOS SANTOS, R. B.; JUNIOR, V. L.; CASTRO, E. V. R. Uso de métodos quimiométricos e mecânico- quanticos na análise de terpenóides e fenilpropanóides. *Orbital*, v. 1, n. 4, p. 273–288, 2009.

ENAN, E. Insecticidal activity of essential oils: octopaminergic sites of action. *Comparative Biochemistry and Physiology*, v. 130, n. 3, p. 325–337, 2001.

EVERETT, C. J.; MATHESON, E. M. Biomarkers of pesticide exposure and diabetes in the 1999 – 2004 national health and nutrition examination survey. *Environment International*, v. 36, n. 4, p. 398–401, 2010.

FERREIRA, C. P.; YANG, H. M.; ESTEVA, L. Assessing the suitability of sterile insect technique applied to *Aedes aegypti*. *Journal of Biological Systems*, v. 16, n. 4, p. 565–577, 2008.

FIGUEIRÓ, A. C.; SÓTER, A. P.; BRAGA, C.; HARTZ, Z. M. A.; SAMICO, I. Análise da lógica de intervenção do programa nacional de controle da dengue. *Revista Brazileira de Saúde Materno Infantil*, v. 10, n. Suplemento 1, p. S93–S106, 2010.

FORATTINI, O. P. *Aedes* (*Stegomyia*) *Alboptctus* (Skuse) identification in Brazil. *Revista Saúde Pública*, v. 20, n. 3, p. 244–5, 1986.

FORATTINI, O. P. 2002. *Culicidologia Médica*. 1 ed. São Paulo, 2002, 860 p.

FORATTINI, O. P.; GOMES, A. C.; NATAL, D.; KAKITANI, I.; MARUCCI, D. Preferências alimentares de mosquitos Culicidae no Vale do Ribeira, São Paulo, Brazil. *Revista de Saude Publica*, v. 23, n. 1, p. 9–19, 1989.

FORATTINI, O. P.; KAKITANI, I.; MARQUES, G. R. A. M.; BRITO, M. Novos encontros de anofelíneos em recipientes artificiais. *Revista Saúde Pública*, v. 32, n. 6, p. 598-599, 1998.

HUANG, C. L.; FU, J. T.; LIU, Y. K.; CHENG, D. M.; ZHANG, Z. X. The insecticidal and repellent activity of soil containing cinnamon leaf debris against red imported fire ant workers. *Sociobiology*, v. 62, n. 1, p. 46–51, 2015.

HUIGNARD, J.; LAPIED, B.; DUGRAVOT, S.; MAGNIN-ROBERT, M.; KETOH, G. K. Modes d'action neurotoxiques des derives soufrés et de certaines huiles essentielles et risques lies à leur utilization. In: Regnault-Roger, C., Philogène, B. J. R. & Vincent, C. (Eds.). Biopesticides d´Origine Végétale, (2nd ed.). *Les nouveautés Tec & Doc*, p. 219-231, 2008.

IOC/FIOCRUZ. *Dengue vírus e vetor.* Disponível em: (http://www.ioc.fiocruz.br/dengue/textos/oportunista.html). Acesso em: 10/1/2017.

JUMBO, L. O. V. 2013. *Atividade inseticida e de repelência de óleos essenciais de cravo e canela sobre o caruncho Acanthoscelides obtectus (Say)*. Dissertação de Mestrado, Universidade Federal de Viçosa, Viçosa, Minas Gerais, 57p.

KATZ, T. M.; MILLER, J. H.; HEBERT, A. A. Insect repellents: Historical perspectives and new developments. *Journal of the American Academy of Dermatology*, v. 58, n. 5, p. 865–871, 2008.

KAVVALAKIS, M. P.; TSATSAKIS, A. M. The atlas of dialkylphosphates; assessment of cumulative human organophosphorus pesticides' exposure. *Forensic Science International*, v. 218, n. 1–3, p. 111–122, 2012.

KOSTYUKOVSKY, M.; RAFAELI, A.; GILEADI, C.; DEMCHENKO, N.; SHAAYA, E. Activation of octopaminergic receptors by essential oil constituents isolated from aromatic plants: Possible mode of action against insect pests. *Pest Management Science*, v. 58, n. 11, p. 1101–1106, 2002a.

KOSTYUKOVSKY, M.; RAFAELI, A.; GILEADI, C.; DEMCHENKO, N.; SHAAYA, E. Activation of octopaminergic receptors by essential oil constituents isolated from aromatic plants: possible mode of action against insect pests. *Pest Management Science*, v. 58, n. 11, p. 1101–1106, 2002b.

LUCCA, P. S. R. 2009. *Potencial inseticida de extratos de funcho, erva-doce, cravo-da-india e do preparado homeopático para controle de pulgão em couve*. Dissertação de Mestrado, Universidade Estadual do Oeste do Paraná, Paraná, Rio Grande do Sul, 75p.

MCKINLAY, R.; PLANT, J. A.; BELL, J. N. B.; VOULVOULIS, N. Endocrine disrupting pesticides: Implications for risk assessment. *Environment International*, v. 34, n. 2, p. 168–183, 2008.

MCMENIMAN, C. J.; LANE, R. V.; CASS, B. N.; FONG, A. W. C.; SIDHU, M.; WANG, Y. W.; O'NEILL, S. L. Stable introduction of a life-shortening *Wolbachia* infection into the Mosquito *Aedes aegypti. Science*, v. 323, n. 5910, p. 141–144, 2009.

MEKALI, J.; NAGANAGOUD, A.; KAPASI, M.;. Management of rhyzopertha dominica fab. Under modified atmospheric condition in stored. *Entomological Studies*, v. 2, n. 2278–5566, p. 34–43, 2013.

MELO, R. M. P. DOS S.; FERNANDES, J. I.; VIEIRA, V. P. C.; RIBEIRO, F. A.; BOTELHO, M. C. S. N.; VEROCAI, G. G.; SCOTT, F. B. Eficácia do piretróide permetrina no controle de *Psoroptes ovis* (Hering, 1838) (Acari: Psoroptidae) em coelhos (*Oryctolagus cuniculus*) naturalmente infestados. *Revista Brazileira de Parasitolologia Veterinária*, v. 17, n. 1, p. 55–58, 2008.

HEALTH MINISTRY. *Monitoramento dos casos de arboviroses urbanas causados por vírus transmitidos pelo mosquito Aedes (dengue, Chikungunya e Zika), semanas epidemiológicas 1 a 21, 2021*. 2021. Vol 52. Brasília (DF), 2021, 11p.

HEALTH MINISTRY. *Monitoramento dos casos de arboviroses até a semana epidemiológica 21 de 2022*. 2022. Vol 53. Brasília (DF), 2022, 14p.

MOSTAFALOU, S.; ABDOLLAHI, M. Pesticides and human chronic diseases: Evidences, mechanisms, and perspectives. *Toxicology and Applied Pharmacology*, v. 268, n. 2, p. 157–177, 2013.

NASCI, R. S.; HARE, S.; SCOTT, F. Interespecific mating between Louisiana strains of *Aedes albopictus* y *Aedes aegypti* in the field and laboratory. *Quality*, v. 5, n. 3, p. 416–421, 1989.

NELSON, M. J. 1986. *Aedes Aegypti: Biologia y Ecologia*. 1 ed. Washington D.C., 1986, 62p.

NEVES, D. P. 2006. *Parasitologia Dinâmica*. 2 ed. São Paulo, 2006, 474p.

OLIVEIRA, R. A.; REIS, T. V.; SACRAMENTO, C. K.; DUARTE, L. P.; OLIVEIRA, F. F. Constituintes químicos voláteis de especiarias ricas em eugenol. *Brazilian Journal of Pharmacognosy*, v. 19, n. 3, p. 771–775, 2009.

PAWAR, V.; THAKER, V. *In vitro* efficacy of 75 essential oils against *Aspergillus niger*. *Mycoses*, v. 49, n. 4, p. 316–323, 2006.

PHASOMKUSOLSIL, S.; SOONWERA, M. Efficacy of thai herbal essential oils against three immature stages of *Aedes aegypti* (Linn), *Anopheles dirus* (Peyton and Harrison) and *Culex quinquefasciatus* (Say). *Journal of Herbal Medicine*, v. 2, n. 2, p. 25–35, 2013.

PLARRE, R.; PÖSCHKO, M.; PROZELL, S.; FRANK, A.; WOHLGEMUTH, R.; PHILLIPS, J. K. Effects of oil of cloves and citronellol, two commercially available repellents, against the webbing clothes moth *Tineola bisselliella* Hum. (Lepidoptera: Tineidae). *Anzeiger für Schädlingskunde, Pflanzenschutz, Umweltschutz*, v. 70, n. 3, p. 45–50, 1997.

POLITEO, O.; JUKIC, M.; MILOS, M. Comparison of chemical composition and antioxidant activity of glycosidically bound and free volatiles from clove (Eugenia caryophyllata Thunb.). *Journal of Food Biochemistry*, v. 34, n. 1, p. 129–141, 2010.

PRASHAR, A.; LOCKE, I.; EVANS, C. Cytotoxicity of clove (*Sygyzim aromaticum*) oil and its major components to human skin cell. *Cell Proliferation*, v. 39, n. 4, p. 241–248, 2006.

PRICE, D. N.; BERRY, M. S. Comparison of effects of octopamine and insecticidal essential oils on activity in the nerve cord, foregut, and dorsal unpaired median neurons of cockroaches. *Journal of Insect Physiology*, v. 52, n. 3, p. 309–319, 2006.

PROTEIN DATA BANK. PDB. Disponível em: (https://www.rcsb.org/pdb/explore/remediatedSequence.do?structureId=3K1E). Acesso em: 28/8/2017.

RABÊLO, W. F. 2010. *Caracterização química, toxidade e avaliação da atividade antibacteriana do óleo essencial do cravo da Índia (Syzygium aromaticum)*. Tese de Doutorado, Universidade Federal do Maranhão, Maranhão, 79p.

RANA, I. S.; RANA, A. S.; RAJAK, R. C. Evaluation of antifungal activity in essential oil of the *Syzygium aromaticum* (L.) by extraction, purification and analysis of its main component eugenol. *Brazilian Journal of Microbiology*, v. 42, n. 4, p. 1269–1277, 2011.

REIS, S. L.; MANTELLO, A. G.; ROSSETE, È. A.; CARDOSO, A. M.; BELEBONI, R. O. Insecticidal and repellent activity of typical monoterpenes from plant essential oils against *Callosobruchus maculatus* (Fabr. 1775). *BMC Proceedings*, v. 8, n. Suplemento 4, p. P115, 2014.

REITER, P. Oviposition and dispersion of *Aedes aegypti* in an urban environment. *Le Bulletin de la Société de Pathologie Exotique*, v. 89, n. 2, p. 120–2, 1996.

RIBAS, J.; CARREÑO, A. M. Avaliação do uso de repelentes contra picada de mosquitos em militares na Bacia Amazônica. *Anais Brazileiros de Dermatologia*, v. 85, n. 1, p. 33–38, 2010.

ROZMAN, V.; KALINOVIC, I.; KORUNIC, Z. Toxicity of naturally occurring compounds of Lamiaceae and Lauraceae to three stored-product insects. *Journal of Stored Products Research*, v. 43, n. 4, p. 349–355, 2007.

SANTORO, G. F.; CARDOSO, M. G.; GUIMARÃES, L. G. L.; MENDONÇA, L. Z.; SOARES, M. J. *Trypanosoma cruzi*: Activity of essential oils from *Achillea millefolium* L., *Syzygium aromaticum* L. and *Ocimum basilicum* L. on epimastigotes and trypomastigotes. *Experimental Parasitology*, v. 116, n. 3, p. 283–290, 2007.

SCHERER, R.; WAGNER, R.; DUARTE, M. C. T.; GODOY, H. T. Composição e atividades antioxidante e antimicrobiana dos óleos essenciais de cravo-da-índia, citronela e palmarosa. *Revista Brazileira de Plantas Medicinais*, v. 11, n. 4, p. 442–449, 2009.

SERVICE, M. W. 1993. *Mosquito ecology: field sampling methods*. 2 ed. Liverpool, 1993, 988p.

SNOW, W. F. The spectral sensitivity of *Aedes aegypti* (L.) at oviposition. *Bulletin of Entomological Research*, v. 60, n. 4, p. 683, 1971.

SOUSA, L. S.; CRUZ, A. C. R.; MAGIOLE, B. R.; RESENDE, M. M. G.; MACHADO, E. R. Efetividade da atividade larvicida de óleos essenciais frente ao *Aedes aegypti*. In: SILVA, C. D. D.; FERREIRA, H. R. P. *Entomologia: diversidade e evolução dos insetos*. Paraná: Atena, 2022. p. 14-41.

SOCIEDADE BRAZILEIRA DE PARASITOLOGIA. *Glossário de parasitologia*. Disponível em: (http://www.parasitologia.org.br/estudos_glossario_D.php). Acesso em: 8/8/2017.

TABANCA, N.; BERNIER, U. R.; ALI, A.; WANG, M.; DEMIRCI, B.; BLYTHE, E. K.; KHAN, S. I.; BASER, K. H. C.; KHAN, I. A. Bioassay-guided investigation of two Monarda essential oils as repellents of yellow fever mosquito *Aedes aegypti*. *Journal of Agricultural and Food Chemistry*, v. 61, n. 36, p. 8573–8580, 2013.

TAKKEN, W.; KLINE, D. L. Carbon dioxide and 1-octen-3-ol as mosquito attractants. *Journal of the American Mosquito Control Association*, v. 5, n. 3, p. 311–6, 1989.

TAKKEN, W.; KNOLS, B. G. J. Odor-mediated behavior of Afrotropical malaria mosquitoes. *Annual Review of Entomology*, v. 44, n. 1, p. 131–157, 1999.

TAUIL, P. L. Perspectivas de controle de doenças transmitidas por vetores no Brazil. *Revista da Sociedade Brazileira de Medicina Tropical*, v. 39, n. 3, p. 275–277, 2006.

TAVEIRA, L. A.; FONTES, L. R.; NATAL, D. 2001. *Manual de diretrizes e procedimentos no controle do Aedes aegypti*. Ribeirão Preto, 2001, 104p.

TEHRI, K.; SINGH, N. The role of botanicals as green pesticides in integrated mosquito management – A review. *International Journal of Mosquito Research*, v. 2, n. 1, p. 18–23, 2015.

THOMSEN, R.; CHRISTENSEN, M. H. MolDock: A new technique for high-accuracy molecular docking. *Journal of Medicinal Chemistry*, v. 49, n. 11, p. 3315–3321, 2006.

TRONGTOKIT Y , RONGSRIYAM Y, KOMALAMISRA N, KRISADAPHONG P, A. C. Laboratory and field trial of developing medicinal local Thai plant products against four species of mosquito vectors. *The Southeast Asian Journal of Tropical Medicine and Public Health*, v. 35, n. 2, p. 325–333, 2004.

TSITSANOU, K. E.; THIREOU, T.; DRAKOU, C. E.; KOUSSIS, K.; KERAMIOTI, M. V.; LEONIDAS, D. D.; ELIOPOULOS, E.; IATROU, K.; ZOGRAPHOS, S. E. *Anopheles gambiae* odorant binding protein crystal complex with the synthetic repellent DEET: Implications for structure-based design of novel mosquito repellents. *Cellular and Molecular Life Sciences*, v. 69, n. 2, p. 283–297, 2012.

UNIPORT. *UniPortKB*. Disponível em: (http://www.uniprot.org/). Acesso em: 28/8/2017.

UNIVERSITY OF FLORIDA. *Shark attacks hit all-time high in 2015*. Disponível em: (http://news.ufl.edu/articles/2016/02/shark-attacks-hit-all-time-high-in-2015.php). Acesso em: 12/7/2017.

VALLE, D.; BRAGA, I. A. *Aedes aegypti* : histórico do controle no Brazil. *Epidemiologia e Serviços de Saúde*, v. 16, n. 2, p. 113–118, 2007.

WAKIL, W.; RIASAT, T.; LORD, J. C. Effects of combined thiamethoxam and diatomaceous earth on mortality and progeny production of four Pakistani populations of *Rhyzopertha dominica* (Coleoptera: Bostrichidae) on wheat, rice and maize. *Journal of Stored Products Research*, v. 52, p. 28–35, 2013.

WHO. 2009. *Guidelines for efficacy testing of mosquito repellents for human skin*. Nova York, 2009, 37p.

WHO, 2017. *Animal bites* (http://www.who.int/mediacentre/factsheets/fs373/en/). Acesso em: 12/7/2017.

WHO, 2014. *Media centre World Health Day 2014: Preventing vector borne diseases* (http://www.who.int/mediacentre/news/releases/2014/small-bite-big-threat/en/). Acesso em: 12/7/2017.

WHO, 2016. *World malaria report 2016*. 2016ª (https://www.who.int/publications/i/item/9789241511711). 12/7/2017.

WHO, 2016. *Zika situation report* (http://www.who.int/emergencies/zika-virus/situation-report/4-march-2016/en/). Acesso em: 12/7/2017.

WHO, 2017. *Media centre malaria* (http://www.who.int/mediacentre/factsheets/fs094/en/). Acesso em: 12/7/2017.

WHO, 2017. *Global vector control response 2017–2030* (https://www.who.int/publications/i/item/9789241512978). Acesso em: 12/7/2017.

WOGULIS, M.; MORGAN, T.; ISHIDA, Y.; LEAL, W. S.; WILSON, D. K. The crystal structure of an odorant binding protein from *Anopheles gambiae*: Evidence for a common ligand release mechanism. *Biochemical and Biophysical Research Communications*, v. 339, n. 1, p. 157–164, 2006.

YEAP, H. L.; MEE, P.; WALKER, T.; WEEKS, A. R.; O'NEILL, S. L.; JOHNSON, P.; RITCHIE, S. A.; RICHARDSON, K. M.; DOIG, C.; ENDERSBY, N. M.; HOFFMANN, A. A. Dynamics of the "Popcorn" *Wolbachia* infection in outbred *Aedes aegypti* informs prospects for mosquito vector control. *Genetics*, v. 187, n. 2, p. 583–595, 2011.

ZAGANAS, I.; KAPETANAKI, S.; MASTORODEMOS, V.; KANAVOURAS, K.; COLOSIO, C.; WILKS, M. F.; TSATSAKIS, A. M. Linking pesticide exposure and dementia: What is the evidence? *Toxicology*, v. 307, p. 3–11, 2013.

ZARA, A. L. S. A.; DOS SANTOS, S. M.; FERNANDES-OLIVEIRA, E. S.; CARVALHO, R. G.; COELHO, G. E. Estratégias de controle do *Aedes aegypti*: Uma revisão. *Epidemiologia e Serviços de Saúde*, v. 25, n. 2, p. 1–2, 2016.

ZERINGÓTA, V.; SENRA, T. O. S.; CALMON, F.; MATURANO, R.; FAZA, A. P.; CATUNDA-JUNIOR, F. E. A.; MONTEIRO, C. M. O.; CARVALHO, M. G.; DAEMON, E. Repellent activity of eugenol on larvae of *Rhipicephalus microplus* and *Dermacentor nitens* (Acari: Ixodidae). *Parasitology Research*, v. 112, n. 7, p. 2675–2679, 2013.

ZHANG, Y.; ZHAO, M.; JIN, M.; XU, C.; WANG, C.; LIU, W. Immunotoxicity of pyrethroid metabolites in an in vitro model. *Environmental Toxicology and Chemistry*, v. 29, n. 11, p. 2505–2510, 2010.