


GENETICS AND ECOLOGY OF *Aedes spp.* IN THE STATE OF MATO GROSSO <https://doi.org/10.56238/sevened2024.037-073>**Lenicy Lucas de Miranda Cerqueira¹, Cristina Márcia de Menezes Butakka², Sandra Mariotto³ and Rosina Rosina Djunko Miyazaki⁴.****ABSTRACT**

This chapter is a fundamental contribution to the understanding and management of one of the main vectors of diseases in Brazil. It is the result of a collaborative effort of a research group that has explored in depth the complex interaction between the genetics and ecology of *Aedes spp.*, with a special focus on the specific conditions of the state of Mato Grosso – MT, Brazil. The analysis discusses the genetic diversity and ecological adaptations of *Aedes spp.*, based on data collected directly in the territory of Mato Grosso. It presents studies from the last ten years, which reveal how environmental and genetic factors influence the population dynamics and vector capacity of these mosquitoes. First, it deals with the biology of the *Aedes spp.* mosquito, which makes a bibliographic review of the state of knowledge type in the time period from 2013 to 2023, on the subject in our state. It then discusses climate change and one health, in the face of arboviruses, and details the methods used to map the genetic diversity of *Aedes spp.* populations in Mato Grosso, aiming to clarify genetic variability and the possible implications for insecticide resistance and the spread of diseases. As future perspectives, the chapter also addresses the implications of the results obtained for future control and prevention strategies, discussing integrated approaches, including genomic studies, that can be adopted to mitigate the impacts of these diseases in the state, expanding knowledge about the vector.

Keywords: Mosquitoes. Arboviruses. Public health.

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INTRODUCTION

BIOLOGY OF *Aedes* spp.

Aedes (Stegomyia) aegypti (Linnaeus, 1762) and *Aedes (Stegomyia) albopictus* (Skuse, 1894) are the main vectors of highly pathogenic viruses for humans, such as Dengue (DENV), Chikungunya (CHIKV) and Zika (ZIKV) (Gómez, *et al.*, 2022; Allan *et al.*, 2023), which cause febrile, hemorrhagic and neurological diseases and continue to be a major threat to global public health. The high ecological plasticity, opportunistic dietary patterns and the versatility in the use of urban and natural breeding sites of these vectors have favored their dispersion and adaptation in tropical, subtropical and even temperate zones. Due to the lack of available treatments and vaccines, mosquito population control is the most effective way to prevent arboviruses (Gómez, *et al.*, 2022).

Both *A. aegypti* and *A. albopictus* are diurnal mosquitoes and, in their larval stage, live in clean, still water or in any container that accumulates clean water. Their morphological appearance is very similar and with the naked eye it is easy to confuse the two species of mosquitoes, because they have black and white streaked bodies and legs. *A. aegypti* is characterized by the presence of a set of silvery-white scales and a lyre on the thorax (mesonot) and *A. albopictus* has a longitudinal median band of longitudinal silvery-white scales on the back, in addition to being darker than *A. aegypti*. *A. albopictus* has a single straight line in the center of the thorax (Figure 1).

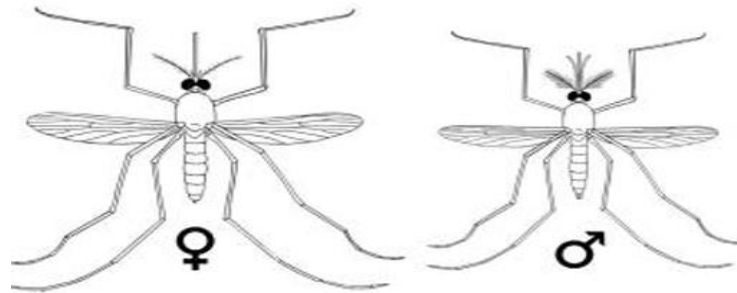
Figure 1 - Difference between the two vector species of arboviruses



Source: Florida Medical Entomology (1999). <https://fmel.ifas.ufl.edu/research-areas/invasion-biology-of-Aedes-albopictus/>.

Gonçalves (2016) showed that the female and the male are differentiated, morphologically (sexual dimorphism), by some characteristics such as the antenna and the abdomen. The female has a hairy antenna and a more robust abdomen, while the male has a feathery antenna and a slender abdomen (Figure 2), as well as longer palps.

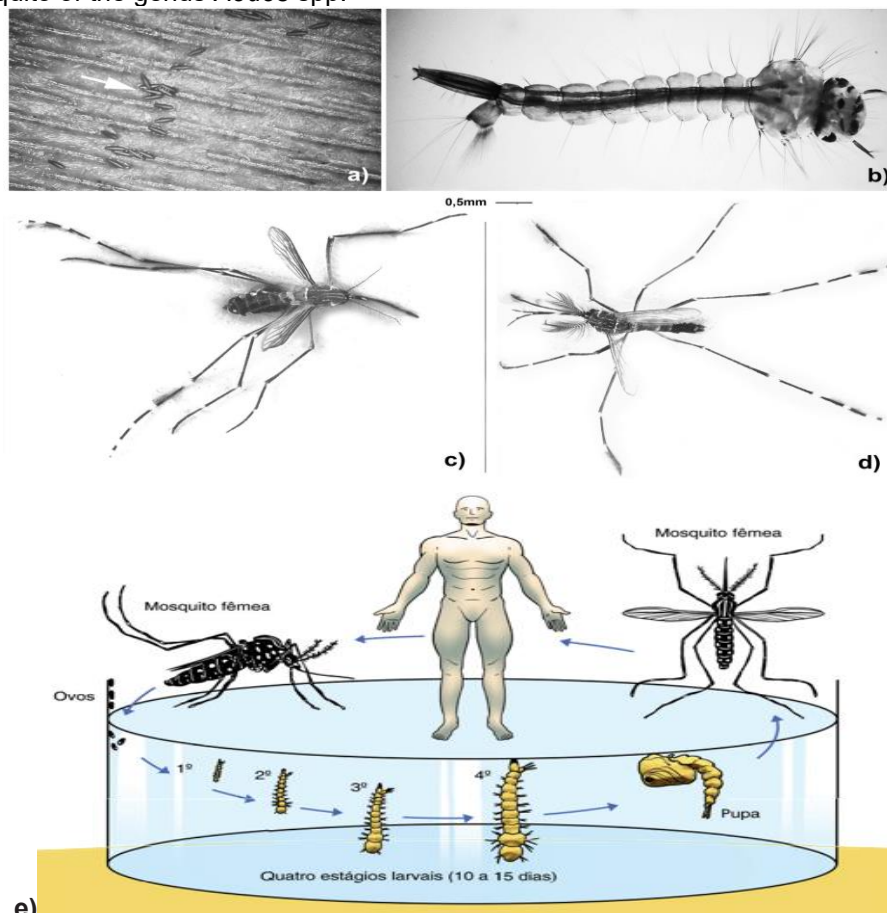
Figure 2 - Sexual dimorphism of the *A. aegypti* mosquito



Source: Lorenz *et al.*, (2018).

The female mosquito, after biting a person, lays the eggs on the wall of the container. The eggs evolve through four larval stages and a pupal stage, when they then transform into adult male and female mosquitoes (Figure 3). Within 24 hours of turning into mosquitoes, males and females fly to mate. The female, from then on, needs blood for the eggs to mature (anthropophilia) and the male feeds exclusively on vegetable juices, and the mosquito lives an average of 30 to 35 days.

Figure 3 - Stages of development of *Aedes* spp.: a) eggs, b) larva, c) adult female and d) adult male e) Life cycle of the mosquito of the genus *Aedes* spp.



Note: Construction by the authors.



Oviposition usually takes place in the late afternoon and the pregnant female prefers dark and shaded environments with clean water to lay the eggs.

We found larvae and pupae in various breeding sites such as bottles and in disposable packaging, cans, tires, vases, cisterns, slabs, uncovered water tanks and others. It also requires attention under potted plants where they accumulate water and are considered potential breeding grounds. Other important environments are pools without cleaning and without weekly treatment and clogged gutters.

CLASSIFICATION OF *Aedes* spp.

Ordem - Diptera (Di=dois, pteros=asas)
Family - Culicidae
Subfamily - Culicinae
Tribe - *Aedini*
Genus - *Aedes*
Subgenus - *Stegomyia*
Species – *Aedes aegypti*
Espécie - *Aedes albopictus*
Aedes (Stegomyia) aegypti (Linnaeus, 1762)
Aedes (Stegomyia) albopictus (Skuse, 1894)

Aedes and *Anopheles* species thrive in the same urban environments, and urgent action is needed to integrate disease surveillance and generate innovative vector control tools for urban populations, to protect the many millions of people at risk (Allan, 2023). The integration of different compatible and effective vector control strategies considers the available technologies and regional characteristics as a viable method to try to reduce mosquito infestation and the incidence of arboviruses transmitted by them (Zara *et al.*, 2016).

It is worth noting the occurrence of populations of *A. aegypti* resistant to organophosphates commonly used as larvicides and insecticides (Montella *et al.*, 2007). This mechanism is known as metabolic resistance and the increased activity of detoxifying enzymes, such as esterases circulating in the vector's body, which contributes to the elimination or inactivation of insecticides (Brasil, 2006).

Insecticide resistance can be thought of as a process of accelerated evolution of a population that responds to intense selective pressure, with the consequent survival of individuals who have alleles that confer resistance. This resistance is pre-adaptive, the result of many mutations (Braga; Valle, 2007). It is very important to understand their mechanisms in order to contribute to the progress of methods capable of overcoming them. When we identify the mechanisms of resistance related to *A. aegypti*, there is a minimization of the evolution of the infection picture and consequent reduction of the impact of resistance to humans (Rosa, 2016).



In general, the resistance they stimulate is due to amplification of the number of genes or increased activity by the action of regulatory mechanisms, which generate an increase in the synthesis of the corresponding product (Rodríguez *et al.*, 2002). Consequently, this adaptive trait in a set of selected genes favors the maintenance of the insect capable of reproducing in an environment exposed to insecticides (Belinato & Martins, 2016).

It is suggested that insecticide resistance may be pertinent to the high genetic differentiation of Brazilian populations of *A. aegypti* and this may reflect in important vector differences and populations from different areas of Brazil should be treated as independent epidemiological units (Ayres *et al.*, 2003).

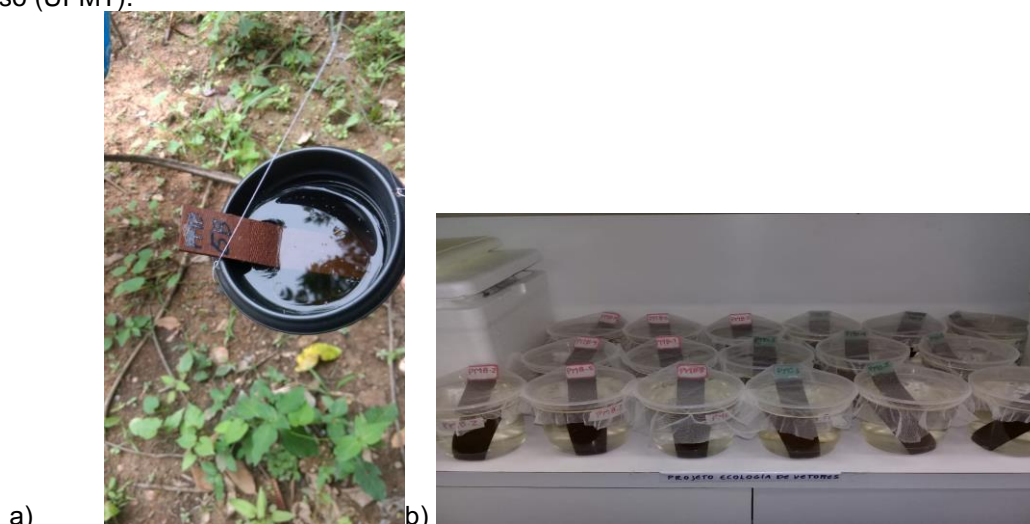
GEOGRAPHICAL DISTRIBUTION

It has a wide geographical distribution, and predominates in tropical and subtropical areas located between the parallels of latitudes 45° North and 40° South (Forattini, 2002). The genus *Aedes* is common to oviposition outside the liquid medium, but close to it or in potentially floodable places. The place for oviposition is one of the main factors responsible for the distribution of mosquitoes in breeding sites and their subsequent dispersal in different geographic areas (Tilak *et al.*, 2004). As a result, the geographic displacement of human beings favors and increases the risk of carrying pathogens from other locations, causing the emergence or reemergence of diseases (Lima-Camara, 2016).

In her reproductive cycles, after each oviposition, the female will respond to the attractive stimuli of a host, which gives the mosquito its epidemiological role in the transmission of diseases (Natal, 2002), especially dengue, Zika, Chikungunya and yellow fever, in addition to the vector having a great capacity to adapt to urban environments (Brasil, 2005). This fact instigates research for new methods of vector control, whose larvae explain the degree of adaptability to drastically altered areas resulting from the destruction of natural habitats, and later, invasion into environments occupied by man.

Its control through ovitraps (egg capture, Figure 4a) is a promising strategy that depends on the scope and time of use of the traps in the field, as developed and efficiently employed by Miyazaki *et al.* (2009), Depoli (2016) and Oliveira *et al.* (2020). After collecting the ovitraps, the material is sent to the Entomology Laboratory of the Federal University of Mato Grosso (UFMT) for the development of the biological cycle (Figure 4b).

Figure 4- a) Ovitrap. b) Development of mosquitoes at the Entomology Laboratory of the Federal University of Mato Grosso (UFMT).



Note: Construction by the authors.

In favorable conditions of humidity and temperature, the development of the mosquito embryo is completed in 48 hours. The resistance to desiccation also allows the eggs to be transported over great distances in dry containers.

Larval density, high fertility and fertility rates, short life cycle, adaptive capacity to environmental changes, and egg resistance to waterlessness are factors that determine the survival, distribution, and density of vectors (Custódio *et al.*, 2019). The abrupt transformation of natural environments (Ferreira Filho *et al.*, 2017) consequently due to anthropogenic changes in the landscape interferes with its proliferation for its rapid population growth, and seems to play an important role in the dynamics of its abundance (Butakka *et al.*, 2022).

STATE OF KNOWLEDGE ABOUT THE GENUS *Aedes* spp. IN THE STATE OF MATO GROSSO

A survey was carried out in the following Brazilian catalogs: 1) Capes Journals; 2) CAPES Catalog of Theses and Dissertations (CTDC), ⁵3) Brazilian Digital Library of Theses and Dissertations (BDTD).⁶

In the Capes Periodicals, the descriptor "*Aedes*" and "Mato Grosso" and specific filters for the period were used, seeking to measure the number of productions related to this theme in the time period from 2013 to 2024 (last eleven years) 47 academic publications were located. After reading the titles and abstracts of these, they were filtered

⁵ Platform for the search and dissemination of the country's scientific production, founded by the Ministry of Education (MEC), available at <https://catalogodeteses.capes.gov.br/catalogo-teses/#/>.

⁶ Platform for searching and disseminating research carried out in the country, developed by the Brazilian Institute of Information in Science and Technology, available at <https://bdt.d.ibict.br/vufind/>.



to 23 publications in which we sought to identify studies that addressed *Aedes* spp. in the state of Mato Grosso in themes focused on general biology, genetics, ecology and health education.

Considering the high importance and need for studies on *Aedes* spp. in the state of Mato Grosso, and based on the studies stored in the Capes periodical portal, we found that the number of scientific productions is still small, requiring more studies to boost the theme in the state and encourage the development of research at various levels.

In the CTDC, when we used only the keyword "*Aedes*", in the CTDC, 2,360 records were generated. When we used the keywords "*Aedes*" and "Mato Grosso" only 02 works returned. Thus, it was decided to use only the keyword "*Aedes*" and the filter "educational institution", with the following options: "Federal University of Mato Grosso (UFMT)", "University of the State of Mato Grosso (UNEMAT)" and "University of Cuiabá" (UNIC), Federal Institute of Mato Grosso and University Center of Várzea Grande (UNIVAG).

When applying the time lapse from 2013 to 2023, the number of dissertations and theses selected for reading and analysis of the abstracts were: 10 for UFMT, 05 for UNIC. Subsequently, in the phase corresponding to pre-analysis, among the productions cited in the research universe, these were selected after reading the abstracts because they were studies on *Aedes* sp. in the state of Mato Grosso, with 12 master's dissertations and 03 doctoral theses. When used as options, the other higher education institutions in the state of Mato Grosso did not return any work.

There is a small number of studies that discuss research with a molecular data approach of *Aedes* spp. In this sense, it is relevant to investigate the directions of these productions, thus contributing to a reflection on the difficulties and importance of this theme.

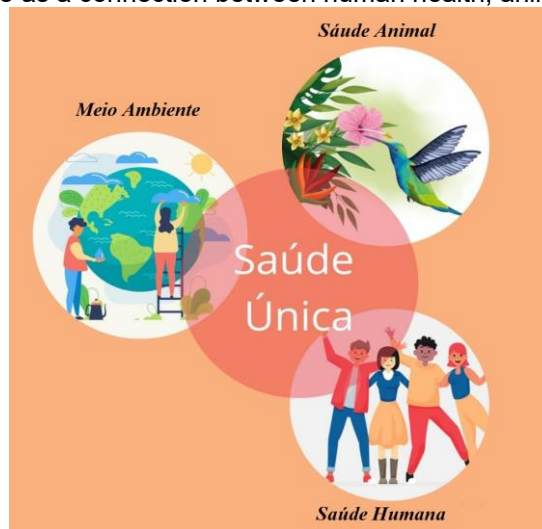
These findings indicate the need for advances and new research. It is understood that the mapping of the theme involved allows the opening of discussions and debates about the large area in Biological Sciences and how the productions have been evidenced in recent years.

CLIMATE CHANGE AND SINGLE HEALTH IN THE FACE OF ARBOVIRUSES

The concept of One Health establishes the connection between human health, animal health and the environment (Figure 5), integrated in a systemic and interdependent way that allows us to exist, coexist and at the same time "evolve" (Lobo *et al.*, 2021). The relationship between man and nature can be understood through One Health with the integration between different knowledge and areas of knowledge in an interdisciplinary and

collaborative way, with a holistic approach to the health-disease process (Cruz-Silva *et al.*, 2023).

Figure 5 – One Health Scheme as a connection between human health, animal health and the environment.



Source: Adaptation of the Environmental Education Portal. Gov. São Paulo (2021).

One Health as an interdisciplinary approach to health promotion must be contemplated with an understanding that human, animal and environmental health are inseparable. The One Health approach arises from the need to respond quickly to the problems arising from the high demographic density of the population and urbanization, the invasion of habitats for the intensification of extractivism and the use of resources (Monteiro *et al.*, 2024). Environmental Education with a focus on One Health promotes awareness and attitudes aimed at its promotion and quality of life (Cruz-Silva *et al.*, 2023).

The Brazilian Constitution of 1988, article 225, establishes the right of everyone to an ecologically balanced environment, an essential good for a healthy quality of life, imposing its defense and preservation on the Government and the community (Constitution of the Federative Republic of Brazil, 1988, update of amendments, 2016). Global climate change is producing direct effects and significant impacts on human health, especially in urban areas. Its effects can lead to exposure to extreme conditions such as natural disasters, hurricanes, droughts, and floods, increased spread of vector-borne diseases such as mosquitoes (Galati *et al.*, 2015), and increased disease transmission. This trend will be accentuated with the increase in temperatures on the planet, directly or indirectly to health, since it is the complex relationship between the physical environment of the atmosphere and the social one, with effects on the dispersion of diseases (Nunes & Mendes, 2012).

Climate variability, understood as an intrinsic property of the terrestrial climate system, is responsible for natural oscillations in the weather patterns observed at the local, regional and global levels. There are several climatic factors, such as relative humidity and



temperature that favor the reproduction and survival of vectors of infectious agents, such as the *Aedes aegypti* mosquito (Confalonieri, 2003), essentially high temperatures in cities (Degallier, 2010). In view of the alarming numbers of sick people, it is necessary to better understand the influence of climate change on the prevalence of vector-borne diseases. Obtaining data is necessary for future decision-making by the agencies responsible for the area of prevention and combat against the mosquito, as well as the civil defense itself.

The accelerated expansion of the fossil fuel industry, deforestation and burning of forests have aggravated global warming and resulted in more favorable climatic conditions for the reproduction and survival of pathogens and vectors (Mordecai *et al.*, 2017). Climate change may accelerate the cycles of arboviruses and expand their geographical distribution areas. Considered a vector that disseminates the Dengue, Zika and Chikungunya viruses, *A. aegypti* has become a threat to public health especially in urban areas, where dengue transmission is strongly influenced by a range of biological and environmental aspects, specifically the high temperatures and formation of heat islands, associated with the precarious conditions of marginalized populations in these environments. It is essential to know its seasonal and geographical range and its relationship with the urban climate (Mordecai *et al.*, 2017). The recent report by the *Intergovernmental Panel on Climate Change* (IPCC), published on February 27, 2022, points to the increased risk of arboviruses, particularly dengue, with longer seasons and a wider geographic distribution in Asia, Europe, Central and South America, and sub-Saharan Africa, potentially putting billions more people at risk by the end of the century (Prophiro, 2022).

The mosquito infestation will always be more intense due to accumulated water, which favors the hatching of mosquito eggs, as well as the lack of basic sanitation, the presence of open ditches are causes of soil contamination and the advance of transmitted diseases. Without infrastructure, the health chaos overloads public health systems and a scenario that prevents, in the short term, the proposition of actions aimed at the eradication of the transmitting vector (Galati *et al.*, 2015).

DATA ON DENGUE, ZIKA AND CHIKUNGUNYA IN MATO GROSSO

By observing the Epidemiological Scenario of Urban Arboviruses: Dengue, Chikungunya and Zika in the state of Mato Grosso, we can perceive the importance of surveillance, prevention and control actions, supporting decision-making at the various levels of management (Table 1).



Table 1 - Number of probable cases of Urban Arboviruses from Epidemiological Week 1 to 30/2024, MT, Brazil

Arboviroses urbanas					
No. Notified cases		No. Probable cases		No. Confirmed cases	Sorotipo DENV
Dengue fever	63.783	40.602	34.643	1, 2 and 4	
Zika	1.654	1.041	256	...	
Chikungunya	18.948	17.315	15.844	...	

Source: SINAN – Epidemiological Surveillance SES-MT-2024 * No. Subject to change due to online database feeding.

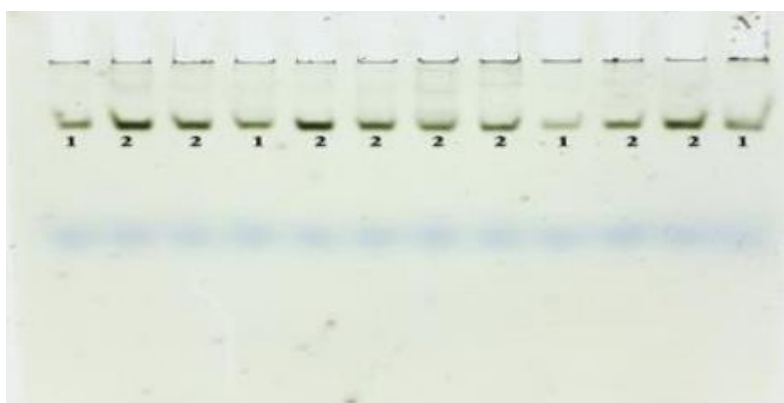
Historically, the peaks of dengue epidemics usually occur between the months of March and April, but this year, changes in temperature and in the rainy season caused by global warming and the serotypes of the disease in circulation drove the increase in cases earlier. Alternatively, the study by Souza *et al.* (2018) in a population in Bahia, indicated that in addition to determinants such as garbage collection, unhealthy housing conditions and the absence of public policies are determinants for the occurrence of cases. Environmental Education is a preponderant factor in the control of zoonosis.

INSECTICIDE RESISTANCE AND ESTERASE ENZYME EXPRESSION

The differential expression between larvae and adults may be relevant for the monitoring of resistance, when it is necessary to compare bioassays and biochemical assays. Esterases of the fourth larval stage of *A. aegypti* populations were identified by Paiva (2006) as the main mechanism that confers resistance to organophosphates, related to gene overexpression. This resistance increased with the increase in the level of the esterases. Research on resistance to these chemical components was also developed by Campos & Andrade (2003).

The analysis of esterase polymorphism occurs by using polyacrylamide gel. Samples are stained with alpha naphthyl acetate and RR-Salt, diluted in acetone and phosphate buffer, then placed in an oven at about 37°C, with no light for one hour. For the identification and analysis of the bands, weak bands (1) and overexpressed bands (2) are considered normal (Figure 6).

Figure 6- Polyacrylamide gel from *Aedes aegypti* mosquitoes stained with RR-Salt and Alpha esterase. Identification of esterase enzyme bands by band intensity, normal-weak bands (1) and overexpressed-dark bands (2).



Source: Lima *et al.* (2021).

Lima *et al.* (2021) concluded that, despite the significant presence of normal expressed alleles, there is evidence of esterase enzyme polymorphism in natural populations of *A. aegypti* in Chapada dos Guimarães - MT. The analysis also revealed the presence of overexpressed alleles, indicating that this population has insects with potential for metabolic resistance. In addition, it was observed that the seasonal phases directly influence the constitution of the population of these organisms. External environmental factors affect the selection of organisms, promoting the overexpression of the esterase enzyme and making it difficult to control the vector. Esterase facilitates the detoxification of the body, making it resistant to chemical substances such as insecticides used to combat the vector, which represents a concern for the management of these populations.

A genetic sequencing model was performed by Anderson *et al.* (2024), to mitigate insecticide resistance through the formation of alleles in a novel gene expression system. These authors tested gonial homologous cell sequences (bgcn) of *A. aegypti* to express multiplexing in the sgRNA inserted in the kynurenine 3-monooxygenase (kmo) gene. In a study on ecogenotoxicology developed by Rodrigues *et al.* (2005) in the municipality of Campo Verde – MT, it was observed that the average heterozygosity index of the esterase enzyme in two populations of insects of another family, the drosophilids, was higher in the natural ecosystem, which indicated an influence of the agroecosystem on the genetic structure of their communities, possibly narrowing the genetic base. Such data showed that insects can be used as sentinel organisms to assess environmental impacts.

Studies of insecticide resistance have shown that it is due to three main types of mechanisms: (a) reduction of insecticide penetration, due to changes in the insect cuticle; (b) increased metabolism of the insecticide by the action of esterase, monooxygenases or glutathione enzymes – transferases; and (c) by modification of the insecticide target (Bisset



et al., 2006). The literature also records a behavioral resistance mechanism, in which insects avoid contact with places that contain the toxic substance.

The effective success of a monitoring program for this vector requires a detailed study of the larvae resistance mechanisms, as well as the factors that regulate their proliferation, which include adaptation to the diversity of breeding sites.

ENVIRONMENTAL EDUCATION – FOCUSING ON SINGLE HEALTH ISSUES

The multidisciplinary essence of Environmental Education is revealed as the theme is related to the concepts of ecologically balanced environment, common use, quality of life, conservation and preservation of the environment, and sustainability (Cruz-Silva *et al.*, 2023). There are numerous possibilities for applying educational practices that are environmentally adjusted to contemporary issues, such as urban densities, pandemics, epidemics, and climate emergencies and their consequences (Monteiro *et al.*, 2024).

Environmental Education and Health are closely related, as a socially and ecologically balanced environment is capable of ensuring a healthy quality of life (Machado, 2018). This approach has the ability to stimulate the analysis of existing socio-environmental problems, as well as more ethical practices and attitudes. Environmental Education (EE) fundamentally involves encouraging the moderate use of natural resources, in order to understand the role of society in the environment. The relationship between man and nature must be resumed in a more sustainable way in view of the complexity surrounding this relationship that involves One Health (Monteiro *et al.*, 2024).

The development of activities that rescue environmental heritage should constitute a goal for environmental preservation, associated with animal welfare and human health. The One Health approach is a path to scientific improvement, which enables long-term socio-environmental and economic balance and well-being. These indicators prioritize One Health and the factors that affect the health situation of a region, not only the ecological aspect, but also by incorporating the political, cultural, and historical dimensions, among others (Limongi and Oliveira, 2020).

In our reality, in the face of arboviruses in the State of Mato Grosso and in Brazil, contributing to environmental education and working with actions to prevent the vector and transmission of diseases such as dengue, it is essential to have a social mobilization with school communities and the population, seeking to create habits of health surveillance in the public and private sectors. Knowledge can generate new paradigms to discuss the disposal of solid waste that can accumulate water, such as cleaning gutters, water tanks, caring for screens in drains and the correct recycling of materials. Mosquito control and



prevention measures can be intensified, as well as reflections on environmental impacts can be proposed to implement safe measures to eliminate mosquito breeding sites.

PUBLIC HEALTH POLICIES

The absence of good urban planning results in inadequate housing conditions, as well as the lack of public policies in basic sanitation services, access to drinking water and the management and disposal of solid waste, among the determining factors for the permanence of arboviruses and other urban diseases.

Faria *et al.* (2022) carried out an analysis of eight reference government documents in order to investigate, through public policies already implemented in Brazil, how basic sanitation is addressed in guiding instruments for the control of arboviruses in the country.

They found that aspects related to communication and social mobilization, vector control and public management are the most addressed topics in the documents analyzed. However, the components of basic sanitation, the management and drainage of rainwater and sanitary sewage were not addressed in the fight against arboviruses, which may contribute to the low effectiveness of intervention and, therefore, such contradiction needs to be overcome by existing public policies in Brazil.

To combat arboviruses, educational actions to combat *A. aegypti* contribute to improving the knowledge and behavior of communities, sensitizing the population to manage the environments they inhabit. Dias *et al.* (2022) proposed health education as an element implemented jointly and articulated with other vector combat strategies. Positive results were associated with the use of educational strategies such as: reduction of mosquito habitats; elimination of breeding sites; reduction of vector infestation; expansion of knowledge, attitudes and practices around vector control that contributed to making it empowered communities.

However, the health sector, by itself, cannot solve the complexity of the factors that favor the proliferation of the dengue vector, such as the *A. aegypti* mosquito, because dengue control encompasses a global intervention process, whose approach goes beyond the health sector. Educational interventions around diseases transmitted by *A. aegypti* are strategic actions that should be implemented in a participatory and continuous manner, with active methods, which have demonstrated more satisfactory results in health promotion, in contrast to the vertical teaching models that have demonstrated limitations in motivating the population to control vectors.

Faced with this scenario, the State Health Department of the Federal District launched a Plan to combat dengue and other arboviruses (2024-2027), subsidized by the



national regulatory framework, in view of the epidemiological and entomological situation in recent years, especially to adopt intersectoral measures and reduce the population's risk vulnerability. However, this plan identifies the ento-epidemic situation, health education and communication actions within the regions, but it does not yet involve other sectors, such as urban planning for rainwater drainage and sanitary sewage with technological innovations, notably in the outskirts of Brazilian cities.

Investing in environmental education and raising awareness among the population is necessary to influence the political process and increase the recognition that we must better understand the relationships between One Health, social relations and the integrity of ecosystems, help in planning in which prevention should be invested and succeed on an ecologically healthier planet. The association of Environmental Education and Health, together with the approach to these themes, has the ability to "encourage the analysis of existing socio-environmental problems, stimulate more ethical practices and attitudes, and enable the development of activities that rescue environmental heritage and encourage preservation", as pointed out by Machado (2018).

The control of disease vectors in areas that have shown rapid urbanization is necessary not only for health actions, but also the implementation of public policies that integrate the mobilization of society, improvements in housing, sanitation and actions to avoid further deforestation (Machado, 2013). It is important to evaluate urban health and environmental quality as a concept intertwined in the aspects of the spatial, biological, social and economic urban environment (Ribeiro & Vargas, 2015). Thus, it is recommended that managers and representatives of civil society frequently dialogue with all segments of the community to identify which factors are preponderant for vector control. They should represent priorities for an environment free of vector-borne diseases, collective mobilizations, investments in health, safety, leisure, infrastructure, especially basic sanitation (Dias *et al.*, 2022).

This intersectoral dialogue between civil society and communities should be a priority in health surveillance, with an emphasis on environmental sanitation and guidance on the correct disposal of garbage, as this can generate a large number of potential breeding sites (Rocha *et al.*, 2014). Mol *et al.* (2020) found a relationship between solid waste management and the occurrence of dengue, and pointed out the importance of implementing adequate solid waste management in municipalities as a strategic action to reduce dengue cases. In this sense, "participation, mobilization and social control work as a transversal axis", uniting health education, environmental education and basic sanitation. Faria *et al.* (2023) addressed that, for public policies to reach a level of control of



arboviruses, it is essential "a set of measures and interventions in environmental risks that prevent or minimize the spread of the vector, avoiding or destroying potential breeding sites of *A. aegypti*".

In this sense, good practices in solid waste management can be cited, such as the installation of ecopoints for the correct segregation and packaging of waste. Based on the analysis of the guiding instruments in the context of combating urban arboviruses in Brazil, it is necessary to present clear and specific actions with good health management, which allows decisions between different sectors of the government (intersectoriality) in the implementation of public policies to combat urban arboviruses.

THE GENETICS OF *Aedes* spp.

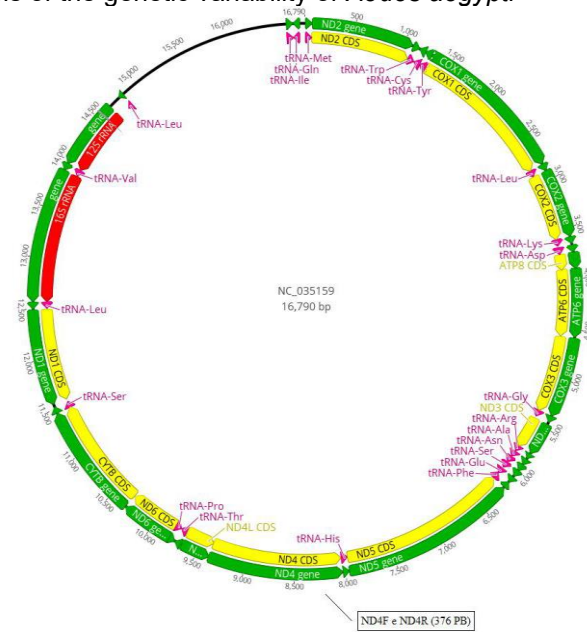
According to Valdivalangan *et al.* (2016), shedding light on genetic deviation in populations of *A. aegypti* is of crucial importance to fully understand its molecular ecology and evolution. It is also important to know the chromosomal structure, diversity, and patterns of population dispersal to understand the genetic mechanisms that have led to this mosquito, which has a relatively small genome (with three chromosomes), adaptive abilities, and resistance to insecticides and environmental changes.

Spadar *et al.* (2024), to support monitoring efforts and strategies for vector control, provided the first global catalog and geographic distribution on mutations and gene duplications in *A. aegypti*. These researchers used whole-genome sequencing samples from this species in 15 countries, including nine in Africa, to investigate genetic diversity in four genes linked to insecticide resistance. The use of genetic sequencing revealed important information on molecular markers involved in insecticide resistance. These researches provide guidance for functional studies and the design of genotyping assays.

The differences and similarities of homologous mtDNA sequences between species have been used extensively to construct evolutionary trees. mtDNA evolves relatively quickly and so this approach has been most useful in tracing recent evolution (Griffiths *et al.*, 2016). To study gene regions and their variations, amplifications from pairs of *primers* (primers) are required, which are easily designed when diversified genomic data are available.

Mitochondrial DNA (Figure 7) has been used as an important tool in the study of population structure, and to understand population dynamics and evolutionary processes.

Figure 7 - Illustrative image of the mNAHD4 gene in which it is possible to observe the site and its number of base pairs used for the analysis of the genetic variability of *Aedes aegypti*



Source: <https://www.ncbi.nlm.nih.gov>

Mitochondrial genetic systems are made up of DNA and the molecular mechanism necessary to replicate and express the genes contained in that DNA. Mitochondrial DNA, abbreviated as mtDNA, was discovered in the 1960s, at first by electron micrographs that showed DNA-like fibers in mitochondria. Later, these fibers were extracted and characterized by physical and chemical procedures. The advent of recombinant DNA techniques has made it possible to analyze mtDNA in great detail. In fact, the complete nucleotide sequences of the mtDNA molecules of many different species have already been determined. Each mitochondria contains multiple copies of DNA, and because each cell usually has many mitochondria, the number of mtDNA molecules per cell can be very large. Animal mtDNA is small and compact... The size of invertebrate mtDNA is approximately equal to that of vertebrates, but their genetic organization is somewhat different (Snustad; Simmons, 2017).

Chu *et al.* (2016) presented the complete sequence of the mitochondrial genome of *Culex tritaeniorhynchus* (Diptera: Culicidae) using the traditional Sanger sequencing method. According to the authors, the sequences of the mitochondrial ND4 and ND5 genes can offer relevant insights into population genetic structure and gene isolation as a function of distance. In addition, the cytochrome c oxidase I (COI) gene, widely used as a marker in the DNA barcode, is highlighted as an essential tool for molecular identification. This method, used for more than two decades in eukaryotes, has contributed significantly to resolving taxonomic conflicts and distinguishing subspecies with recent genetic divergences (Chu *et al.*, 2016). According to Hebert *et al.* (2003) and Hajibabaei *et al.* (2007), precursors



in the use of DNA barcode, the results obtained from this region of mtDNA can facilitate the identification of species, providing subsidies for detailed genetic analyses, especially when genomic data already available delimit target sequences with precision.

Contessoto *et al.* (2023) described the genomic architecture of the *Aedes aegypti* mosquito and discovered a different organization from that found in most living beings, through data-based physical simulations to study the three-dimensional construction of the genome. A computer modeling of the mechanical properties of the genome revealed that the chromosomes of *the Aedes aegypti*, due to their atypical presence of the organism This latest discovery provides a possible physical mechanism that links clues to gene regulation and gene structure analysis between different populations.

For the species *A. aegypti* from Mato Grosso, it would be of great value to do a study based on the example of Kurucz *et al.* (2022), which used *A. koreicus* specimens collected over 12 years from five European countries. With this it would be possible to determine genetic variations and point out the difficulties of control in populations with greater adaptive abilities. The absence of consistent genomic data for different populations of this vector may mean ineffective public investments in the control of the four main viruses transmitted by the species in the state.

The genetic basis for vector control was contemplated in the work of Almeida and Romano (2024), who demonstrated that genetically modified mosquitoes, through lethal genes or with the ability to make insects sterile, transform or replace the next generations in the population, through the introduction of an effector gene, which blocks the transmission of the virus in the wild population.

Research with the complete sequencing of the mitochondrial genome of *Aedes* (Behura *et al.*, 2011; Souza *et al.*, 2023) contributed with sequences and primer designs, which made it possible to analyze genetic variations that can enable the species to increase population and/or resistance to control mechanisms, as well as to discuss the evolution of the genus in tropical regions. The main *primers* currently used in Brazil for the genus *Aedes* and mtDNA and their variations are ND4, ND5, COI. However, each population of the species can have different mutations, in different states of this continental country and in terms of climatic and ecological diversity.

A methodological strategy for teaching in health and environment is to know the biology of an important dengue vector in different populations. In the article published by Mariotto *et al.* (2022) investigated the genetic variability of *A. aegypti* through molecular markers of mitochondrial DNA, COI, ND4, and ND5. The mosquitoes were collected with the aid of ovitraps for capture at points located in four municipalities in the state of Mato



Grosso: Cuiabá (CB), Várzea Grande (VG), Chapada dos Guimarães (CP), and Santo Antônio do Leverger (SA). PCRs (Polymerase Chain Reactions) were tested on 169 samples, with the best quality DNA, using three pairs of *primers* for mitochondrial DNA (mtDNA): Cytochrome Oxidase Subunit I (COI - F and R), Nicotinamide Adenine Dinucleotide Dehydrogenase Subunit 4 (ND4 - F and R) and Nicotinamide Adenine Dehydrogenase Subunit 5 (ND5 - F and R).

An interesting finding of the aforementioned work was the difficulty of amplifying the specimens in the IOC Region. Of the same DNAs that amplified the ND4 and ND5 regions, with more than 95% positive results, less than 20% resulted in useful amplifications for sequencing, when the pairs of primers used were for the IOC region. Most amplifications seemed nonspecific, and after purification they disappeared, clearly showing punctual differences in the COI region with probable mutations, which was not observed for the other two gene regions (ND4 and ND5). The authors suggest that other *primers* need to be established for populations of different Brazilian regions of *A. aegypti*. However, for this, it is necessary that new genomes are available. Attempts at new *primers* with available genomic data, from other Brazilian states and even from other countries, were frustrating, because when tested they did not amplify DNAs from specimens from Mato Grosso.

On the other hand, the study by Naim *et al.* (2020), showed reduced levels of genetic variation in *Aedes* mosquitoes in regions of Malaysia, which suggest they retain less genetic variation when subjected to fewer changes in environmental conditions. The authors also cite studies with similar results and more stable environmental conditions. This could explain, in part, the high variation of the COI gene region for *the A. aegypti* of Mato Grosso, in view of the sudden changes that the state has undergone in recent decades with the advances of agriculture and deforestation. Moore *et al.* (2013), found a wide variation in the ND4 region of *Aedes* among different populations and locations, and suggest that for local haplotypes of the species there may be adaptive abilities, while Fachinelli *et al.* (2023), state that consistent information on the behavior of the vector and human actions is necessary to implement effective actions to control it.

Conforme Bilton *et al.* (2017), divergences in the mitochondrial COI sequence may not corroborate what was previously recognized to delimit taxonomic groups of insects, such as subspecies. The authors also cite several previous studies that show incompatibilities between traditional taxonomic boundaries and phylogenies based on mitochondrial DNA. These discrepancies can be attributed to factors such as introgression, differential selection, and changes in nuclear and mitochondrial genomes. Thus, the expansion of available genomic databases significantly increases the probability of more



robust diagnoses, allowing a more accurate assessment of the conservation or genetic variation of populations.

Castro Júnior *et al.* (2024) found high intrapopulation genetic variation and low interpopulation variability in the semi-arid region of Paraíba and suggested that it may indicate constant vector migrations to locations with a high number of individuals. When there are smaller genetic distances, interindividual, within a population of vectors, it is suggestive that there is the action of factors extrinsic to the individuals of the population, including the removal of the breeding habitat, which can contribute to reduce variability. As a consequence, intercrosses between individuals who are very close genetically. This was found for the analyses in the municipality of Chapada dos Guimarães, as opposed to the data for the municipalities of Cuiabá, Cáceres, and Santo Antônio de Leverger (Mariotto *et al.*, 2022).

Like mtDNA, nuclear DNA also has significant alterations, especially in populations constantly subjected to pesticides used in fumaces or pyrethroids, common in fumigation. The frequency of nuclear mutations, related to resistance, can result from the pressure exerted artificially and induce the genetic variability of mosquito populations.

Mutations in the Kdr (nDNA) gene, for example, show that this incongruence is consistent across different datasets and taxon sampling, and that it can be explained by any peculiar evolutionary event, such as different levels of saturation in certain lineages or a past history of hybridization across the genus. Even more discrepant is the origin of the species of *A. aegypti* and *A. albopictus* that divide mitochondrial and nuclear data ranges, with no overlap in their confidence interval, and statistically this indicates that the two sets of data reject each other, as well as for the mtDNA data, also the nuclear genes are scarcely known, and different authors insist on the need to expand genomic data for the genus *Aedes*. To delve deeper into nDNA and/or compare with mtDNA, we suggest the recent articles by Parimittr *et al.*, (2018); Zadra *et al.* (2021) and Chieli *et al.* (2023).

The genetic-based analysis of vectors such as *A. aegypti* contemplates, in a contextualized way, a formative learning that contributes to a perception of the importance of science in vector research, and leads to a reflection on the constant variability of populations in different environments. Knowledge of the different adaptations and interpopulation gene flow can expand and renew forms of control for this important vector of viruses.



CONCLUSIONS

The *Aedes aegypti* and *Aedes albopictus* mosquitoes are main vectors of highly pathogenic viruses. Its spread is expanding due to social, environmental, and ecological factors. Understanding the biology of the genus *Aedes*, the epidemiology of arboviruses, and developing new intervention strategies are crucial.

Monitoring requires detailed studies of larval resistance and factors that regulate its proliferation, including adaptation to different breeding sites. The control of *A. aegypti* depends on population awareness, elimination of breeding sites and research into control alternatives against insecticide resistance.

Improving vector control in infested areas is essential, especially since *A. aegypti* is associated with dengue, chikungunya, and Zika in Brazil. Poor urbanization increases the risks of reproduction and transmission. Research carried out in Mato Grosso highlights the importance of genomic data and reinforces the need for surveillance and government actions for effective prevention and combat.



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