







**Research Article** 

# Aedes (Stegomyia) albopictus in rural areas in Brazil: First Record in the State of Ceara

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# **Abstract**

**Background:** This work aims to describe the finding of Aedes albopictus in an intra- and peridomiciliary area of the rural area in the south-central region of Ceará and discuss the need for entomological surveillance of this species in the country.

**Method:** Through traps installed in an intra- and peridomiciliary environment and artificial breeding grounds, eggs and larvae of Ae. albopictus were captured in a rural area of the state of Ceará.

Results: This is the first record of Ae. albopictus in the south-central region of the state of Ceará. We collected 418 eggs and 252 larvae from Ae. albopictus, being 85% of eggs in the peridomiciliary area, on average 29.41 (± 37.34) eggs per trap. As for larvae, about 70% of them were caught on tires. Only 42 larvae (14,2%) belonged to species other than the genus Aedes, found in tires and an engine.

**Conclusion:** The presence of Ae. albopictus in rural areas close to urban areas warns of the need to effectively insert this vector as a surveillance target, carry out an active search, monitor its presence in municipalities, and investigate its involvement with outbreaks in or near areas where they are present.

#### Introduction

Aedes albopictus is one of the main vectors of arbovirus that threaten public health in the world, Dengue (DENV), Yellow fever (FA), Zika (ZIKV), and Chikungunya (CHIKV) [1]. In Brazil, several studies have demonstrated their ability to acquire these viruses and transmit them to their offspring [2–4], classifying them as vector potential. Studies conducted with natural populations of Ae. albopictus detected the presence of DENV in pools from different Brazilian regions and states [5–8], natural ZIKV infection in eggs and adult females collected in the states of Bahia and Espírito Santo [8,9]. During an epidemic outbreak in areas near an urban park of the Atlantic Forest, Natal–RN CHIKV was identified in adults [4].

Ae. albopictus was registered in Brazil for the first time in 1986, in the state of Rio de Janeiro [10] and, since then, has been conquering more and more space. It is currently present in all states, more widely distributed in the Southeast and Midwest regions [11].

In the state of Ceará, the vector was first found in 2005 in the city of Fortaleza, the capital of the state [12] and subsequently, other records continued to be made, but always in the same municipality. In the countryside of the state, its state of distribution, density, and involvement in the transmission of arboviruses in these areas is unknown [13–15].

This work aims to describe the finding of Aedes albopictus in an intra- and peridomiciliary area of the rural area in the south-

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central region of Ceará and discuss the need for entomological surveillance of this species in the country.

# **Methods**

#### Characterization of the area

The study area comprises the rural area of the municipality of Lavras da Mangabeira, located in the mesoregion of south-central Ceará, about 434 km from the capital Fortaleza (Figure 1). The total population is 31,090 inhabitants, being in the urban area 18,132 (60%) and 12,958 (40%) in the rural area, with a total area of 947.95 km², corresponding to a demographic density of 32.8 inhabitants/km². The average annual temperature is 27 °C, with the rainy season varying from January to April and the average rainfall is 866.4 mm per year [16].

# Mosquito captures

During a field visit in early April 2022, the presence of mosquitoes with characteristics of the genus *Aedes* was detected when they sought blood repast from visitors. Three larvitraps produced with disposable plastic bottles, containing about 300 mL of water were installed on site, a peridomiciliary area rich in vegetation, and organic matter, with a low incidence of sunlight and presence of porcine.

The traps were collected seven days later and were all positive. The contents were taken to the Cariri Medical Entomology Laboratory for identification. 40 larvae were

counted and identified according to the classification key provided by Consoli and Lourenço-de-Oliveira (1994) [17]. All specimens were identified as *Ae. albopictus*.

After the identification of the species, new visits were made to the same area in the same month, including the selection of new points for collection. This time, ovitraps composed of a black pot containing about 300 mL of water and a eucatex reed measuring 3x12 cm for oviposition were installed. The traps were installed inside two residences (Res), equidistant 1.5 km, in addition to the peridomiciliary area, and collected five days after their installation. In each residence (Res 1: 6.79076589°S; 39.10096783°W) and (Res 2: 6.7793123°S; 39.0929763°W) six traps were distributed, three mounted in the intradomiciliary area and three in the peridomiciliary area. In Res 2, the peridomiciliary points consisted of porcine and goat stables (Figure 2).

These points are at about 277 m in height, around the Mescla stream, which is part of the drainage network of the Salgado River sub-basin, a tributary of the Jaguaribe River Basin (Figure 3). The circled points correspond to the flight radius of *Ae. albopictus* which can reach up to 800 m [15].

During the second field visit for the installation of the ovitraps, two outbreaks of larvae were found in artificial breeding sites closer to Res 1. The first, in a tractor tire with clear water, and the other in the engine of the same equipment, with a layer of oil on the water. The distance from this focus to the residence was about 5 meters.

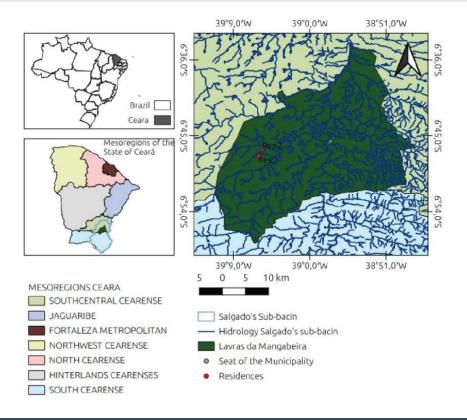


Figure 1: (A) Map representing the location of the state of Ceara in Brazil. The state of Ceara stands out in relation to other Brazilian states (B) Mesoregions of the state of Ceará, with emphasis on the municipality of Lavras da Mangabeira. The municipality is located between the central south and south cearence mesoregions. (C) Expansion of the region of the municipality of Lavras da Mangabeira.



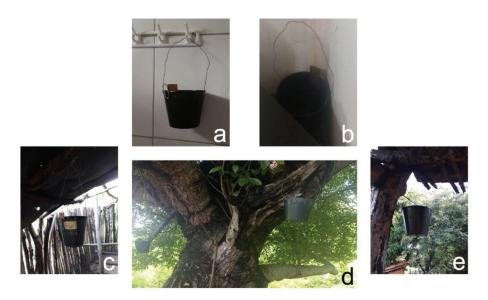


Figure 2: Ovitraps installed in the intradomiciliary (a,b) and peridomiciliary area (c-e).

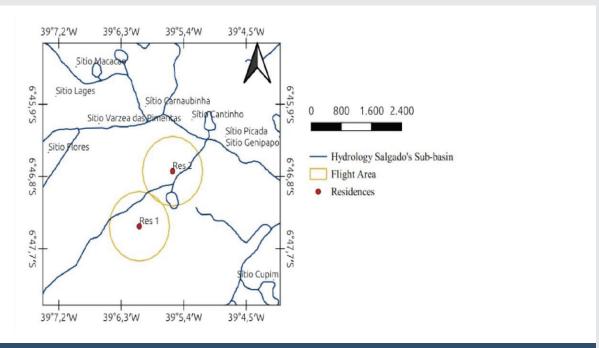


Figure 3: Installation location of traps for collecting eggs of Ae. Albopictus and flight area in relation to points in the rural area of Lavras da Mangabeira – CE, Brazil.

# **Data analysis**

Data were tabulated and analyzed using GraphPad Prism software. The number of eggs collected in the traps (three in the intradomicile) and (three in the peridomicile) installed at each visit was summed and presented in a table. The average of eggs per trap was calculated for each area and presented graphically. To compare the two samples (intradomiciliary and peridomiciliary area), the Mann–Whitney U test was applied at a significance level of 0.05. The number of larvae collected was counted separately and presented graphically, distributed by type of breeding.

## **Results**

The materials collected in the ovitraps and foci were taken to the laboratory, where they were quantified and identified. Figure 4 shows important characteristics for the identification of *Ae. albopictus*.

Figure 5 shows the number of eggs captured during the visits. In all, 418 eggs were collected in intra and peridomiciliary areas, with an average of 16.25 ( $\pm$  19.93); 88.25 ( $\pm$  80.20) eggs per area, respectively, and with an average of 5.50 ( $\pm$  10.14), and 29.41 ( $\pm$  37.34) eggs per trap, respectively. Although the

number of eggs collected in the peridomiciliary area represents approximately 85% of the total, there is no evidence to indicate a statistically significant difference between the two samples (p = 0.0833).

In the artificial breeding sites (trap, tire, and motor) 294 larvae were counted, of which 252 (85.7%) of Ae. albopictus. Among the larvae of another species, only one was Culex and the others were not identified but did not have characteristics of the genus Aedes. The largest number of larvae was collected on tyres (175 u), predominantly Ae. albopictus with 76.5% of

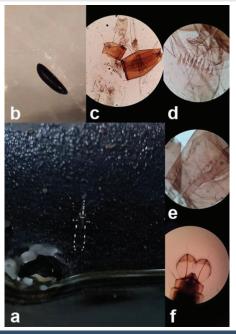


Figure 4: Aedes albopictus in diferent instars. a) Adult mosquito photographed in an artificial breeding site. A longitudinal strip of silvery scales is observed on the cephalothorax. The other images were taken using specialized equipment. b) The egg shows a shiny finish and spherical, evenly spaced horns with regular, homogeneous geometry of surface ridges. c) and d) 4th instar larva: pecten in a single row, in the shape of a long spine and serrated base. e) Larva highlighting the shape of the siphon comb. f) A pupa highlighting the shape of the swimming vanes with a clear fringe and the presence of the single bristle 9 in segment VIII.

them (Figure 6). Considering the number of eggs and larvae collected, 670 specimens were counted.

## **Discussion**

It is believed that Ae. albopictus is spread in at least 59% of Brazilian municipalities [8,18,19] leaving the remaining portion unaware of its local presence. This information is produced from research conducted for the purpose of active search or should be collected during the Rapid Index Survey for Ae. aegypti (LIRAa), carried out by endemic agents. The Ministry of Health advises conducting a bimonthly sample larval survey, or four LIRAa per year in-home visits. This survey is used to monitor and subsidize the control of mosquito infestation Aedes, identifying more infested areas in the territory and thus predicting the occurrence of outbreaks [20].

It occurs that these surveys have been suffering recurrent questions as to their effectiveness in predicting the risk of epidemic transmission and the weaknesses identified during their execution, either due to lack of knowledge or professional commitment [21]. Thus, in order for the presence of Ae. albopictus to be detected during LIRAa, technical training is required and these conditions must be provided by the state. As Figure 4 explains, most of the biological characteristics that differentiate the two vectors are microscopic and require technical knowledge to perform this task. Therefore, if there is no efficient identification of mosquitoes during these surveys, there is no way to estimate the infestation rate of Ae. aegypti or Ae. albopictus.

On the other hand, despite the proven vector transmission competence of Ae. albopictus in Brazil, all the attention of the control program is still focused on Ae. aegypti. There is no mention of another vector in documents, guidelines, plans, and control strategies. This finding can be corroborated by the example of a robust international project carried out in the country, which aims to introduce into the field Ae. aegypti with Wolbachia, a bacterium that prevents Dengue, Zika, Chikungunya, and Urban Yellow Fever viruses from developing within it, contributing to the reduction of these diseases [22] in the population.

Installation					2007
Trap per house	1st	2nd	Number	Mean/trap	<u>∞</u> 150-
(3); total =12			of eggs	$\pm$ SD	s 150- 50 to 100-
Intradomiciliary					nmpe
House 1	1	42	43	5.50	<sup>2</sup> 50-
House 2	22	0	22	$\pm 10.14$	<sub>0</sub>
Peridomiciliary					Intradomiciliary Peridomiciliary
House 1	197	52	249	29.41	Mean ± SD number of eggs (area) Intradomiciliary: 16.25 ± 19.93 Peridomiciliary: 88.25 ± 80.20 p-value: 0.0833
House 2	94	10	104	$\pm 37.34$	
Number of eggs	314	94	418		
a					b

Figure 5: Number and mean of eggs collected in oviposition traps in intra and peridomiciliary areas. a) Number of eggs collected per house and mean of eggs per trap; b) mean of eggs per area and result of Mann-Whitney test (p = 0.0833).

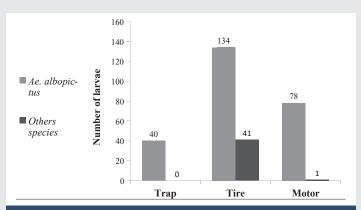


Figure 6: Number of larvae captured in artificial breeding sites.

It is important to note that there is no criticism of an important action against *Ae. aegypti*. It is necessary to alert to the need to implement surveillance measures of *Ae. albopictus* in Brazil, because without them, mosquitoes and arbovirus transmission will continue to expand silently, and may be coresponsible for the high incidences of arboviruses in recent years. It should also be considered that, in addition to the urban environment, this mosquito disperses easily in the rural, semiwild, and wild environments, not depending on the places of great human concentration to reproduce, such as *Ae. aegypti*, and it also withstands lower temperatures [14].

Garcia-Rejon, et al. (2021) [23] refer that *Ae. albopictus* is an invasive mosquito with wide phenotypic plasticity to adapt to new areas and high vector competence to transmit several arboviruses, mainly by transovarial transmission. Thus, it can participate in the endemic spread of diseases, and it serves as a bridge vector for emerging arboviruses between wild, rural, and urban areas. In Brazil, the detection of DENV-3 in males of *Ae. albopictus* was carried out in periods without registration of autochthonous cases with this serotype, suggesting that the silent circulation of the virus may occur by this type of mechanism [3]. In the State of Rio de Janeiro, where the yellow fever virus was isolated in females of *Ae. albopictus*, a study suggested that the mosquito may have acted as an additional vector from wild or rural areas to the urban area [24].

In females and males of *Ae. albopictus*, CHIKV was identified in areas at the epicenter of an epidemic outbreak that occurred in people living near an urban park of the Atlantic Forest, located in Natal-RN[4]. Rezende, et al. (2020) [8] reported the first finding of *Ae. albopictus* specimens infected with DENV-1 and ZIKV during an outbreak of dengue-like disease in a rural area in the state of Espírito Santo.

Based on the information showcased, it is clear that the rural area cannot be neglected. Therefore, it is necessary to have access to the same entomological surveillance services offered in the urban area so that the false idea of the non-existence of *Ae. aegypti* or *Ae. albopictus* in the rural area is not created, since, especially, in those located close to the cities, there are no barriers that prevent the proliferation of vectors, especially because there is a daily flow of people moving between one area and another in busing school, commercial, medical-hospital and leisure services. Residents can become

infected in the city and introduce the virus to the site, where the vector already circulates.

Rural communities close to urban centers often present consumption and packaging disposal behaviors similar to those of the city. However, the aggravating factor is that according to the 2019 National Rural Sanitation Program (PNSR), in these areas, only 55.6% of people have adequate service to water supply; 37.5% to sanitary sewage service, and 44.8% to solid waste management [25]. The inadequate coverage of these essential services can provide several breeding grounds for mosquitoes, as was recorded in this work. The lack of piped water, for example, induces inadequate water storage and, consequently, the supply of artificial breeding sites, favoring the proliferation of vectors.

The presence of Ae. albopictus in the intradomiciliary environment in the rural area suggests its domiciliation and alert to the risks of transmission of the viruses conveyed by it since several studies indicate its vector competence [23]. The predominance observed in the peridomiciliary area is corroborated by the vector's own ecological characteristics in wild environments, which exceed those of Ae. aegypti. Their blood-feeding pattern observed in studies carried out in the United States and Brazil indicated that these mosquitoes are opportunistic, as DNA from humans, wild, and domestic animals was found in the mosquito's blood meal [26,27]. In the present study, the area peridomiciliar where the traps were installed offered varied sources of food, such as mammals (humans, porcine, goats, and dogs) and various birds. Melo, et al. [4] found a predominance of Ae. albopictus over Ae. aegypti in an urban park in Brazil. It is important to investigate whether the same occurs in other Brazilian locations.

#### Conclusion

In the present study, even in small numbers, other insect larvae share the same breeding ground as *Ae. albopictus*, but none of *Ae. aegypti*, were observed perhaps because the rural area presents characteristics that favor the *Ae. Albopictus*. This reinforces the need to effectively consider this vector as a surveillance target, perform an active search, and monitor its presence to know the real situation of vector infestation in the country. In addition, it is recommended to search for arboviruses in *Ae. albopictus* captured in areas with arboviruses and in their vicinity.

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