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Ovitrap to monitor the incidence of *Aedes aegypti*

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Abstract
Dengue is considered an important arbovirus transmitted by female mosquito *Aedes aegypti* that affects humans, and is therefore a public health problem. Aim to monitor the incidence of *A. aegypti* through ovitraps in Ituiutaba municipality, MG and verify the influence of abiotic factors on the oviposition of the vector female. Were used 120 ovitraps in 29 neighborhoods the municipality to monitor the incidence of *A. aegypti*. The ovitraps were collected after 7 days, when egg counting was carried out and the association was made between abiotic factors. 70,821 vector eggs were collected, 22.8% during the month of October. The highest Ovitrap Positivity Index (69.47%) and Egg Density Index (106.17) occurred in October. It is noteworthy that in this period, the highest rainfall recorded in the year (317 mm) was verified. Our data show that ovitraps with beer yeast solution proved to be proved to be effective in monitoring *A. aegypti* and *Culex* incidence.

Keywords: Ovitrap, *Aedes aegypti*, vectors, Dengue

1. Introduction
Dengue is an acute febrile disease and its etiological agent is a virus belonging to the *Flavivirus* genus that infects men through the bite of the *Aedes aegypti* female mosquito. Currently, four serotypes are known: DEN-1, DEN-2, DEN-3 and DEN-4 [1, 2].
The disease has great epidemiological importance with an increase in the cases number over the past decades. In the Americas, the number of infected people comes to 16,679,800 cases [3]. Brazil contributes to these cases due to the climate that favors the permanence of the vector and population cultural habits [4]. The epidemiological bulletin issued by the Brazilian Ministry of Health, 241,664 cases of Dengue fever had been recorded throughout the country in 2018 [4, 5].
In Minas Gerais state, 25,949 cases have been reported in the National Notifiable Diseases Information System (SINAN). A study carried out in Ituiutaba city, Pontal do Triângulo Mineiro during 2012 showed that in the first months of the year there was a greater number of notified dengue cases, February with 162 positive reports, followed by March and January months, with 115 and 88 cases, respectively. These data were correlated with the characteristics of the local summer climate, with a peak in the months where rainfall and average temperature increase [6].
Vector control should be to eliminate mosquito breeding sites, especially to avoid potential reservoirs that can accumulate water in backyards, such as exposed tires, cans, bottles and plastics, in addition to changing water from aquatic plants, treating water in swimming pools, keep water boxes closed, gutter cleaning, among other preventive measures. The use insecticides against the larval and adult forms of the mosquito is a very important method in controlling the vector [7].
The population monitoring of *A. aegypti* is done through special traps for collecting mosquito eggs, known as ovitrap, where the female mosquito lays its eggs. This method has been proving to be very efficient and economical for the monitoring vector, since it allows the counting and identification of eggs. In addition, the use of attractive substances enhances the effect of the trap, attracting pregnant mosquito females [6, 8-11].
Aimed in the present study monitoring the seasonality of *A. aegypti* using ovitrap in different neighborhoods of Ituiutaba, Minas Gerais, Brazil and correlate with a dengue notification registered in the municipality in 2018.

2 Materials and methods

For this study, secondary data obtained by the Zoonosis Control Center (CCZ) of the city of Ituiutaba, Minas Gerais by active search for eggs and larvae of *A. aegypti* in ovitraps.

2.1 Ovitrap

The method used was the ovitrap type trap, which consists of a black plastic container with 16 cm in height and 10 cm in diameter with an average capacity of 800ml and a paddle is of raw MDF (Medium Density Fiberboard) reed with 13.5 cm long and 2.5 cm wide was used as substrate for oviposition of mosquitoes. MDF paddle were prepared two days before the trap is set up, soaked in a basin of water for 24 hours to remove sawdust remnants. After 24 hours, the straws were put to dry on paper towels for another 24 hours. After that, the paddle was ready to be used in the field. The paddle was placed inside the container and fixed with a clip with the smooth part facing out of the trap and the rough part facing inwards, as it facilitates the setting of the eggs at the time of laying (Fig 1). A 0.04% beer yeast solution was prepared to attract the female mosquito, 1.0mL of this solution was added inside the ovitrap in 300 ml of water. The ovitraps were installed in places in the house that were away from animals and children and protected from sunlight and rainwater.

All ovitraps were labeled with the name of the project, the ovitrap number and the

municipality identification, an IBGE (Brazilian Institute of Geography and Statistics) number of the municipality and the date of installation. A form was filled in which contains all the trap information, it contains the residence block, address, name of the resident, trap number, place of installation, date of installation, number of eggs and observations if necessary.

2.2 Study area

The municipality has an area of 2,598,046 km² and an estimated population of 104,067 inhabitants. The study was conducted in Ituiutaba municipality, Minas Gerais State, in 29 neighborhoods (Setor Norte, Centro, Setor Sul, Natal, Jerônimo Mendonça, Residencial Carlos Dias Leite, Novo Tempo, Mirim, Jamila, Pirapitinga, Guimarães, Marta Helena, Helio, Platina, Morada do Sol, Novo Mundo, Central, Alvorada, Satélite Andradina, Distrito Industrial, Residencial Monte Verde, Jardim Europa, Jardim Europa II, Lagoa Azul I, Lagoa Azul II, Residencial Estados Unidos, Sol Nascente, Camilo Chaves e Residencial Dr. Marcondes) from March to October 2018.

2.3 Installation of ovitraps

120 traps were installed and egg collection was carried out throughout the month. The ovitraps were placed in the pre-determined places at the beginning of each month and was maintained for seven days. This period was determined to make adult appearance unfeasible (hatching of larvae) and exclude the possibility of the trap becoming an artificial breeding ground for the mosquito [12] The criterion for choosing the neighborhoods was the *A. aegypti* infestation index (LIRAa). After collecting the paddles, they were sent to the laboratory at the CCZ for counting and identification of eggs and larvae.

2.4 Indexes for measuring egg density

The Ovitrap Positivity Index (OPI) was calculated by counting the number of eggs harvested from the straws, this index allows to observe the spatial distribution of the infestation in a locality. By counting the eggs in each reed, it is also possible to calculate the Egg Density Index (EDI), which is important to identify the periods of higher and lower reproduction of females [13].

Ovitrap Positivity Index (OPI)
OPI = NPT / NTE X 100
NPT = Number of Positive Traps
NTE = Number of Traps Examined.
Egg Density Index (EDI)
EDI = NE / NPT
NE = Number of Eggs
NPT = Number of Positive Traps.

The eggs number was associated with the monthly values of minimum, average and maximum temperature and relative humidity taken from the National Meteorological Institute, and precipitation, collected at the Ituiutaba Water and Sewage Superintendency.

The results obtained were described and evaluated through the correlation analysis using the Spearman test and Kruskal Wallis analysis between groups were performed in the Statistica 8.0 Software. Values with $p < 0.05$ were considered significant.

3. Results

During the months of March to October 2018, eggs from 120 ovitraps were counted in 29 neighborhoods of Ituiutaba-MG. The analysis of these paddle, a total of 70,821 eggs and 109 larvae of *A. aegypti* and 14 larvae of the genus *Culex* were counted. A greater number of eggs was observed during the months of March, April and October with a total of 16,169 (22.8%), 14,048 (19.8%) and 13,594 (19.2%) eggs, respectively (Fig 1). In addition, during the months of March, May and June, the largest number of larvae were collected, with 65 (59.6%), 37 (33.9%) and 14 (12.9%) larvae, respectively.

In relation to the locations for collecting the paddle, the highest amount of eggs was observed in the Centro 16,772 (23.7%), Platina 7,221 (10.2%) and Alvorada 4,765 (6.7%) neighborhoods. However, in Guimarães, Jerônimo Mendonça and Platina neighborhoods, a larger number of larvae were found in ovitrap (22, 20 and 14, respectively) (Fig 2). It should be noted that the Pirapitinga and Marta Helena neighborhoods were found 7 *Culex* larvae, which corresponds to 50% of those observed in ovitraps.

Considering the total of collected paddles, an Egg Positivity Index (EPI) of 69.47% was observed, with the months of April, March and October showing higher values of EPI, 84.1%, 82.5% and 81.6%, respectively (Fig 3).

The municipality's Egg Density Index (EDI) was also analyzed, which reached a value of 106.17 during the year. Considering the EDI obtained in March, April and October, it was observed that it was the months with the highest indexes: 160.08, 143.34, 134.31, respectively. The neighborhoods with the highest EDI were Jardim Jamila (317.3), Centro (220.4) and Camilo Chaves (152.7) (Fig 3).

There was a correlation between the quantity of eggs and the precipitation observed during the collection period ($p = 0.0260$; $r^2 = 0.5902$) and between the relative humidity and the quantity of eggs ($p = 0.0044$; $r^2 = 0.7670$). There was no correlation between the quantity of eggs collected and the

temperature ($p = 0.1418$; $r^2 = 0.3227$).

4. Discussion

According to the Notifiable Diseases Information System (http://sinan.saude.gov.br/sinan), 665 cases of dengue and 1 case of severe dengue were confirmed in the Ituiutaba, until October 2018. Due to these data, there was a greater concern in carrying out the collection of eggs by oviposition traps to verify the municipality vector's incidence.

The results of the present study show that a greater number of eggs was found in the Centro neighborhood, as it is an area of great crowding of people, it favors the availability of food for females of the vector and consequently the laying of eggs in these places. Dos Santos Nunes *et al*. (2011) and Barata *et al*. (2001) found the highest number of eggs in regions with a large number of residences and residents and in places far from that area, the number of eggs decreased [14, 15]. According to Donaliso and Glasser (2002) and Natal (2012), the females of the vector prefer the home environment to perform the repast and egg laying because they are close to the food source [16, 17].

This study showed that the climatological variables are correlated with the quantity of eggs and larvae, since the months where there was greater relative humidity and precipitation were practically the months of greatest egg and larvae obtaining in ovitrap. Although there was no significant correlation between the number of eggs and temperature, the data in this study showed a tendency towards greater oviposition in the months with the highest average temperature. The main abiotic factors such as temperature and rainfall are the most directly affect the survival, reproduction, distribution and density of eggs and therefore the mosquito vector that has been shown to be associated with cases of dengue. The highest incidence of the disease coincides with the summer, which is the season of the year when rainfall and temperature are highest [18-21].

The EPI and EDI described in the present study were considered high [20]. In Ituiutaba, the months in which the highest EPI was observed were October (84.1%) and April (82.5%), and the highest EDI was October (160.08) and March (143.34). Unlike those authors who observed a value of 57.9% EPI in the months of October and December 2004, and approximately 80% and 85% of EDI in the months of August 2004 and June 2005, respectively [20]. Therefore, the results found indicate the need to encourage the use of ovitraps in the neighborhoods, especially those most needy located in the municipality, with the EPI and EDI it is possible to perform in the early detection of new infestations and in the surveillance of vector populations, in addition to showing the spatial distribution of the infestation in any given location that was worked on.

5. Conclusion

Our data show that ovitraps with beer yeast solution proved to be effective in monitoring the incidence not only of *A. aegypti* but also allowed the detection of other species of mosquitoes such as *Culex* which, depending on the region, could prove useful in monitoring of this specie. In addition, the significant amount of eggs and larvae principally of *A. aegypti* detected in ovitraps in the different studied locations in the studied municipality, suggest the need for intense monitoring and the development of preventive actions to combat the vector mosquito, which may reduce the notified cases of the disease thus improving the quality of population life.

Acknowledgment

Zoonoses Control Center, Ituiutaba City Hall, Minas Gerais, Brazil, for providing secondary data for this study.

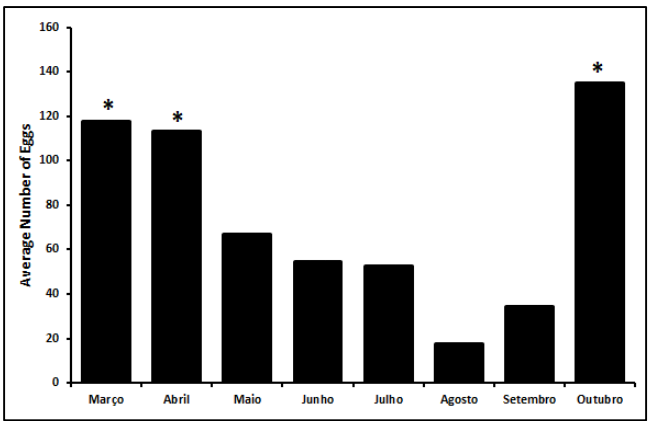


Fig 1: Seasonality of oviposition detected in ovitraps. * p< 0.05

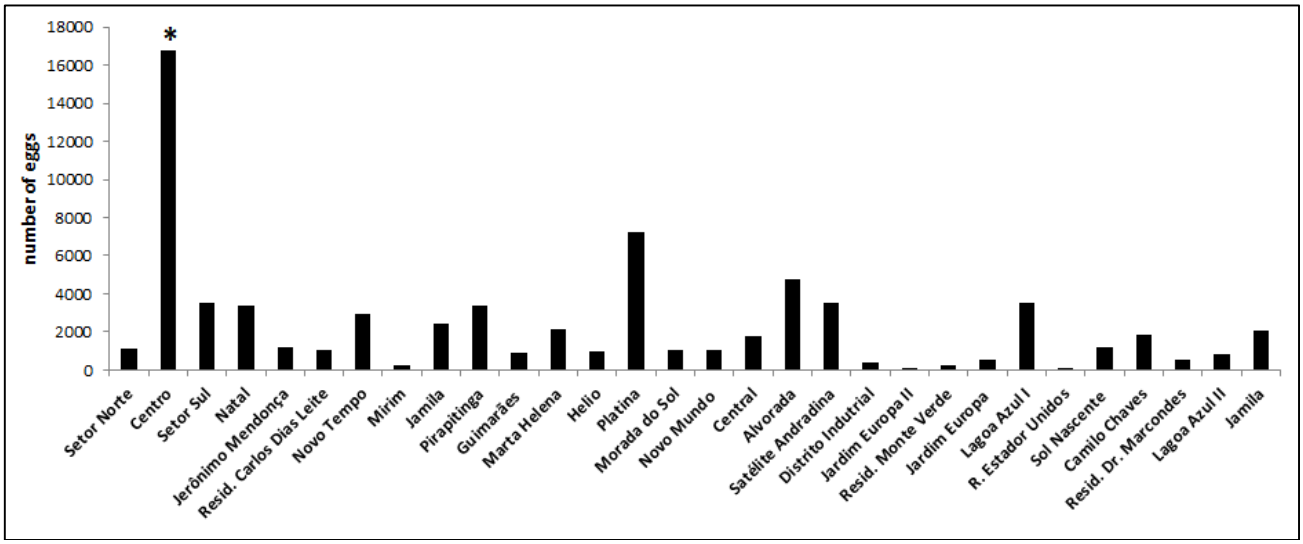
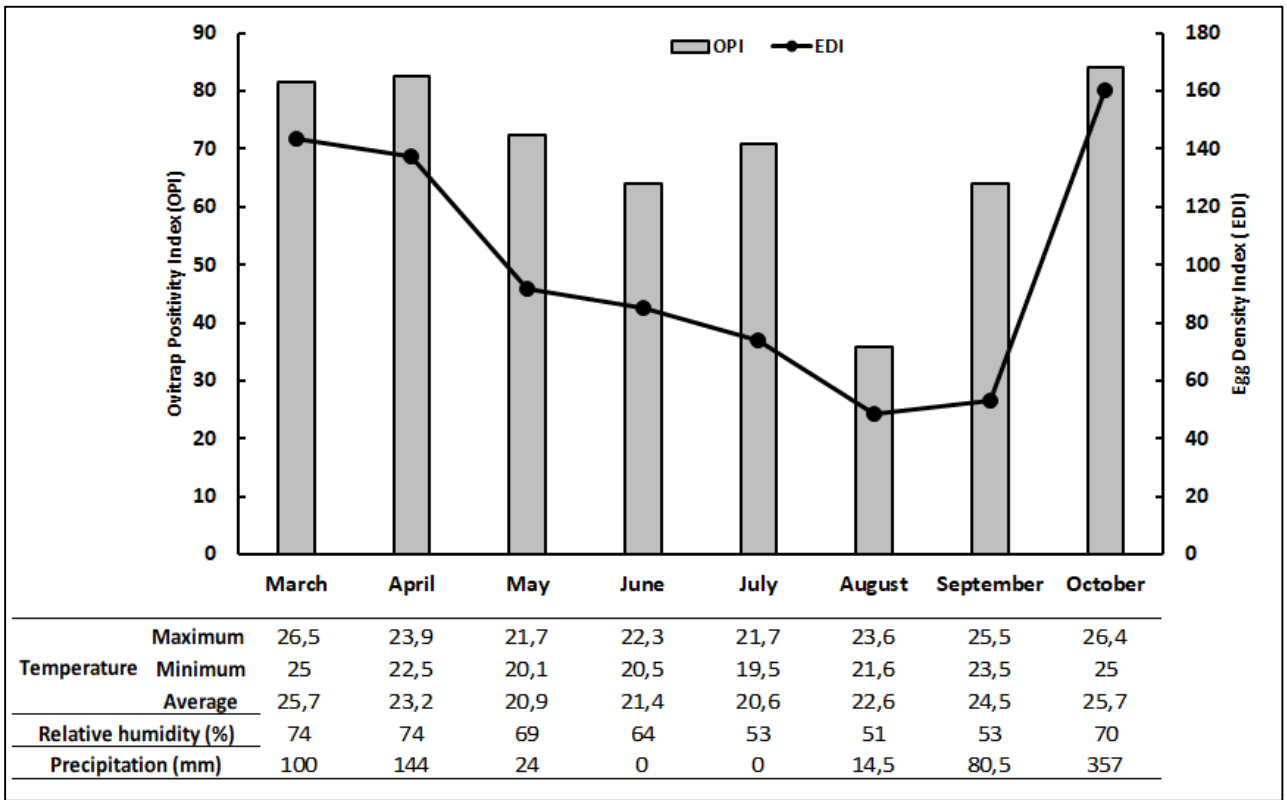


Fig 2: Potential de reproduction in the neighborhoods of *A. aegypti*, number of eggs collected in ovitraps March to October. *p<0.05



		March	April	May	June	July	August	September	October
Temperature	Maximum	26,5	23,9	21,7	22,3	21,7	23,6	25,5	26,4
	Minimum	25	22,5	20,1	20,5	19,5	21,6	23,5	25
	Average	25,7	23,2	20,9	21,4	20,6	22,6	24,5	25,7
Relative humidity (%)		74	74	69	64	53	51	53	70
Precipitation (mm)		100	144	24	0	0	14,5	80,5	357

Fig 3: Oviposition Positive Index (EPI) and Eggs Density Index (EDI) and its relationship to abiotic factors

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