

# Ovitrap surveillance as *dengue* epidemic predictor in Belo Horizonte City, Brazil

## Vigilância com ovitrapas como preditor de epidemia de dengue em Belo Horizonte, Brasil

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### Resumo

**Métodos:** Usamos ovitrapas a cada duas semanas para monitorar o *Aedes spp.* em Belo Horizonte, Capital do Estado de Minas Gerais, Brasil, no período 2002-2013. A proporção média de ovitrapas positivas, o índice de ovitrapas (IPO) apresentou flutuações nesses anos. **Resultados:** O número médio de ovos de *Aedes spp.* (NMO) revelou-se um índice mais sensível para a detecção da presença de *Aedes spp.* que o índice de Breteau. As eclosões dos ovos também mostraram que o *Ae aegypti* era uma espécie predominante durante o período de estudo. A análise dos casos mensais de febre do dengue (FD) e febre hemorrágica da dengue (FHD) correlacionadas com o número de ovos de *Aedes spp.* coletados em todos os anos, mostraram coeficiente de correlação de 0,54 no período 2002-2013. Nossa pesquisa também indicou que dados climáticos, como temperatura e precipitação, estiveram diretamente relacionados ao aumento de ovos de mosquitos em ovitrapas. **Conclusões:** Assim, em circunstâncias em que o índice de Breteau é baixo, a ovitrapa pode ser aplicada como ferramenta de vigilância de boa qualidade e oportuna para avaliar o risco de infecção por dengue humana, e a necessidade de ampliar medidas para o controle do mosquito vetor.

**Palavras-chave:** Dengue. Ovitrapas. Vigilância.

### Abstract

**Methods:** We used ovitraps, set out every two weeks to monitor for *Aedes spp.* in Belo Horizonte City, Brazil, 2002-2013. **Results:** The average proportion of ovitraps with mosquitoes – the ovitraps index (OIP) presented fluctuations in these years. The average egg number of *Aedes sp.* (MNE) revealed that it is a more sensitive index for detecting the presence of *Aedes spp.* than the Breteau index. They also showed that *Ae aegypti* was a dominant species during the period of study. The analysis of monthly cases of *Dengue fever* (DF) and *Dengue hemorrhagic fever* (DHF) revealed that they were correlated with the number of adult female *Aedes spp.* collected through the years with the correlation coefficient of 0.54 in 2002-2013. Our survey also indicated that climate data regarding temperature and precipitation are directly related to the increase of mosquitoes ovules in ovitraps. **Conclusions:** Thus, under circumstances where the Breteau index is low, the ovitrap method should be applied as a good and opportune surveillance tool to evaluate the risk of human *dengue* infection, and the need to scale up for mosquito vector control.

**Keywords:** *Dengue*. Ovitrapas. Surveillance.

### INTRODUCTION

The most important arbovirus disease in humans, *dengue* annually affects 50 million individuals in many countries, and approximately 2.5 billion people live in *dengue* endemic countries<sup>1</sup>. The main vector is the mosquito *Aedes aegypti*, an arthropod with an extremely high capacity of adapting to urban areas.

Since the 1980s, the reemergence of *dengue* has been reported in urban centers in all Brazilian regions. On the following decade, the incidence of *dengue* increased largely as a consequence of the dissemination of *Ae Aegypti*.

Dispersion of the vector was followed by the dissemination of *dengue* virus serotypes 1 and 2 in twenty of the 27 states of the country. Between 1990 and 2000, several epidemics occurred, mainly in the largest urban areas of the Southeast and the Northeast Brazil Regions. The first great *dengue* epidemic occurred in 1998, with approximately 528,000 cases in Brazil<sup>2</sup>.

In Brazil, the increase on the incidence of *dengue* cases in 2002 and the emergence of a third serotype (DENV-3) led to a prediction of an increased risk of *dengue* epidemics

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and an increase of the cases of *Dengue* Hemorrhagic Fever (DHF). In Brazil, cases of the disease that were first observed in large urban centers have been registered in all regions over the next two decades, affecting municipalities of all population sizes<sup>3</sup>. During this period serotypes 1, 2 and 3 have alternated as dominant in different regions, with a major epidemic in 2010, with a circulation of DENV-1 mainly<sup>4</sup>. The DENV-4 serotype reintroduced early in the second half of 2010, after 28 years in the state of Roraima, Northern Brazil, was then identified as indigenous in the state of Amazonas (also in the Northern region) in December 2010, followed by 6 other states in the first half of 2011<sup>5</sup>.

However, the current epidemiological situation shows that these program measures have not achieved the expected results. Epidemiological impact assessments of these interventions have shown that their effectiveness has been extremely limited. In 2013 1.4 million probable cases of *dengue* were reported in the country due to a circulation of DENV-4 serotype, which accounted for 60% of cases<sup>6</sup>.

In Belo Horizonte (BH), the principal city of Brazil's third largest metropolitan area, the first *dengue* epidemic occurred in 1996, and since then, epidemics have occurred every year. The 1996 epidemic (DENV-1), started in the Southern hemisphere's fall, different from the subsequent epidemics, which occurred early in the summer<sup>7</sup>.

By the end of 1997, another epidemic of great intensity (about 86,000 *dengue* cases) started, and lasted until June 1998, characterized by the simultaneous circulation of DENV-1 and DENV-2<sup>7</sup>. The two serotypes continued to produce successive epidemics every year in the city. On February 2002, DENV-3 was identified for the first time in BH, and then the three serotypes co-existed<sup>8</sup>. In 2010, a larger epidemic occurred, DENV-1 predominant, with around 50,000 *dengue* cases<sup>9</sup>. Unfortunately, the DENV-4 was detected in 2011, and in 2013 the biggest epidemic ever reported by the serotypes DENV-1 and DENV-4 (the main serotype) occurred, consisting of around 100,000 *dengue* cases<sup>10</sup>.

The incidence of *Aedes spp.* is related to climatic factors that influence their abundance and distribution. *Ae. aegypti* is widely distributed in tropical and subtropical regions, while *Ae. albopictus* is able to survive at lower temperatures<sup>11</sup>. The survival of *Ae. aegypti* is favored by higher temperatures because in these periods, they have greater longevity, and females are able to increase their blood feeding and egg laying. Another factor that may influence the level of artificial and natural breeding is precipitation due to the accumulation of water in peridomestic sites. The spread of *dengue* vectors is helped by the intensity, frequency and speed at which people and cargo are transported within the country. The reemergence of *dengue* fever in Brazil began in 1986, after the recolonization of the country by its main vector *Ae. aegypti*<sup>12</sup>.

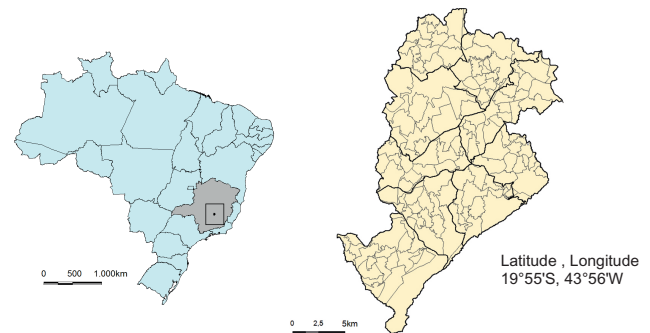
Changes in density are important in disease epidemiology because the vector-to-host ratio is a determinant of the vector capacity of a population<sup>13</sup>. The preventive measures requires efficient vector surveillance tools and methods sensitive enough to predict or detect in real time a sudden mosquito population growth.

Several technological innovations, in different levels of development, evaluated to be incorporated into programs for vector control, as the traps to capture adult mosquitoes, which could provide a better entomological indicator. However, there is a simple and inexpensive tool, the ovitrap survey, which isn't well valued, although the use of ovitraps in the surveillance of *Ae. aegypti* is recommended by the World Health Organization<sup>14</sup>.

## MATERIALS AND METHODS

### Study sites

Belo Horizonte City (19° 55' S, 43° 56' W, mean elevation: 852.28 m) is a very important site for *dengue* cases in Brazil, with 2.4 million inhabitants living in a 331 Km<sup>2</sup> area (figure 1).



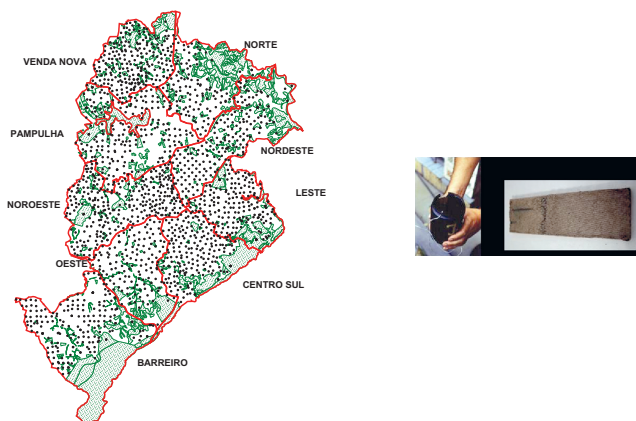
**Figura 1** Location of Belo Horizonte City

There are good sanitation conditions in the formal urban occupation (potable water - 99.7%; garbage collection -100%; Sewer - 93.0%), but slums are located near the formal areas<sup>15</sup>. Throughout the months of October-May it has high temperatures (average temperature ranging from 21.1 °C to 22.3 °C) and relative air humidity (72% – 79%). The city has excellent conditions for breeding mosquitoes at that time of the year, which explains the maintenance of *dengue* transmission. A cold season with sparse rainfall from June-Mid October precedes the heavy rainy season<sup>16</sup>.

Since 1998 the local health service has established a structured mosquito surveillance and control program. The Environmental Health Program currently employs 1200 agents (ACE) visiting about 850.000 premises every two months. Among other health actions, they are responsible for the control of *dengue* vectors, using larvicides in a non-disposable reservoir breeding, as well as for environmental management, information, education and mobilization<sup>10</sup>.

## Ovitrap surveillance

At every two epidemiological weeks, eggs are collected from positive ovitraps (of 1703 placed in 200 meters radii), covering the inhabited areas of Belo Horizonte City (Figure 2).



**Figura 2** Ovitrap Surveillance in Belo Horizonte City

A conventional ovitrap (Fay & Eliason 1966) was used for sampling *Aedes spp.* eggs set out over an eleven-year period from July 2002 through June 2013. It consists of a black plastic cup initially filled with 500ml of grass infusion diluted in water to 30%. A wooden paddle 3 x 12 cm is vertically fixed with clips to the inner wall of the cup as oviposition substrates. Paddles were replaced every two weeks. The trap was hung in the exterior area of a residential premise 1m above ground level in the shade and protected from rainfall. The paddles were collected after seven days and taken to the BH Entomology Laboratory. The paddles collected in these traps were examined and paddles eggs were counted.

The eggs collected are hatched every three months. Then, fourth instar larvae (L4) reared in the laboratory from field-collected eggs were used for species identification.

## Data Analysis

The abundance of *Aedes spp.* in the potential field site was analyzed as follows: Ovitrap index, the percentage of positive ovitrap against the total number of ovitraps recovered for each site (OPI); The number of *Aedes spp.* eggs per recovered ovitrap; Egg density index (EDI) is estimated by dividing the total number of eggs found on the pallets by the number of positive ovitraps (number of eggs / positive ovitraps); Mean Number of Eggs per ovitrap (MNE) is estimated by dividing the total number of *Aedes spp.* eggs found on the pallets by the number of ovitraps (number of eggs / ovitraps).

Comparisons were made between the OPI, EDI and MNE by year (2002-2013).

Comparisons were made between MNE and climatic data.

Data regarding the average temperature and rainfall in BH from 2002 to 2013 were obtained from the National Institute of Meteorology (INMET)<sup>17</sup>.

Comparisons were also made between MNE and cases of DF. Weekly data of DF incidence were recorded according to the time of the onset of symptoms and residence in data base of the Belo Horizonte Municipality Health Secretariat.

In order to analyze the predictive power of a specific egg count we used Simple Linear Regression plotting the MNE from August-September with the occurrence of cases in later years and calculated the Pearson's correlation coefficients with respective scatters plots correlating MNE in previous dry weeks (August-September) compared with the number of DF cases in the subsequent year in the city and the Administrative Regions.

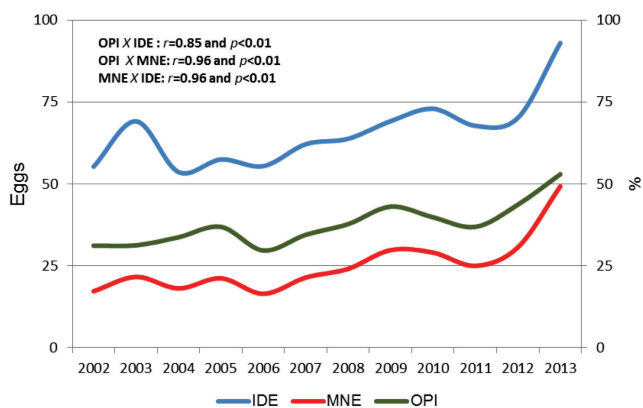
Data was compiled and graphics were made using Excel 2003. Then statistical correlations and significance were done with the R package version 2.40.

## Ethical Considerations

The data of DF incidence were recorded according to the time of the onset of symptoms and residence in secondary data base of the Belo Horizonte Municipality Health Secretariat. The vector presence data source was the Belo Horizonte Municipality Health Secretariat System Zoonosis Control (SCZOO-BH). The use of the database was approved by the Belo Horizonte Municipality Health Secretariat.

## RESULTS

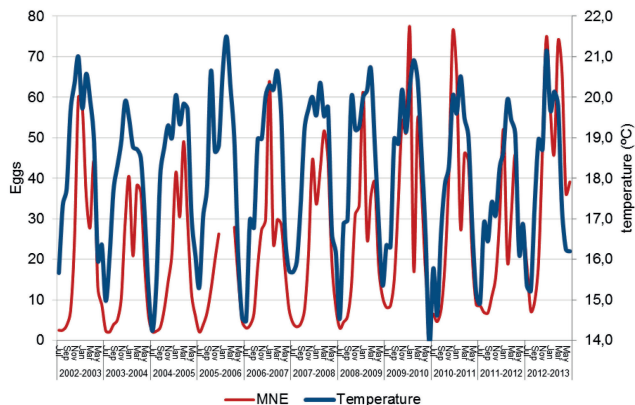
The Pearson's correlation between positivity rate of egg trap (OPI) and MNE  $r=0.96(p<0.01)$ ; OPI versus IDE  $r=0.85(p<0.01)$ ; and MNE versus IDE  $r=0.96$  and ( $p<0.01$ ) (Figure 3).



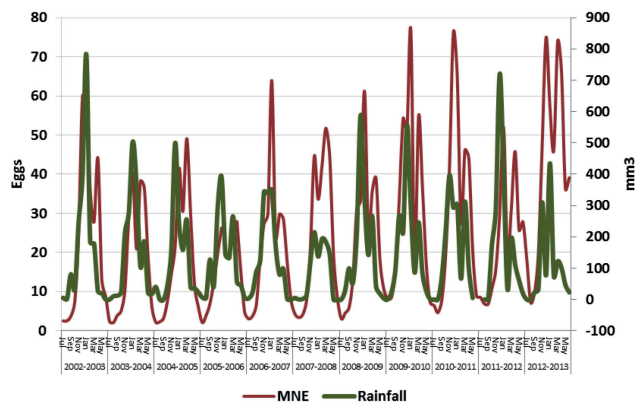
**Figura 3** *Aedes spp.* ovitraps index (MNE, OPI and IDE), Belo Horizonte City /Minas Gerais, 2002-2013

The MNE enabled the detection of temporal and seasonal fluctuations in the population of *Aedes spp.*, with yearly increased trend in the analyzed period (Figure 2). These

*Aedes spp.* presence fluctuations are correlated with the minimum temperature in the installation month  $r=0.65$  and  $p<0.01$ (Figure 4). The monthly rainfall is also correlated with the monthly MNE  $r=0.54$  and  $p<0.01$  (Figure 5).

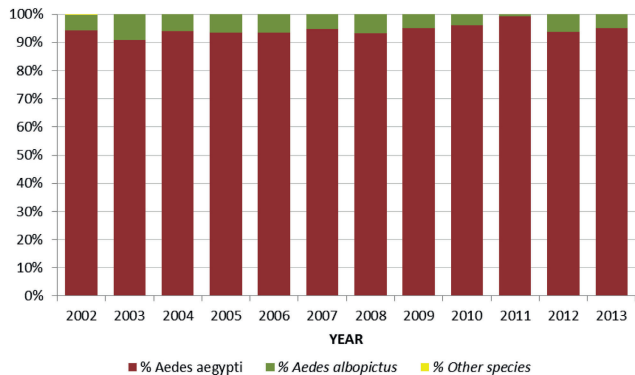


**Figure 4** Values of mean monthly of minima temperature daily and the entomological, indexes mean number of eggs (MNE), July/2002 and June/2013



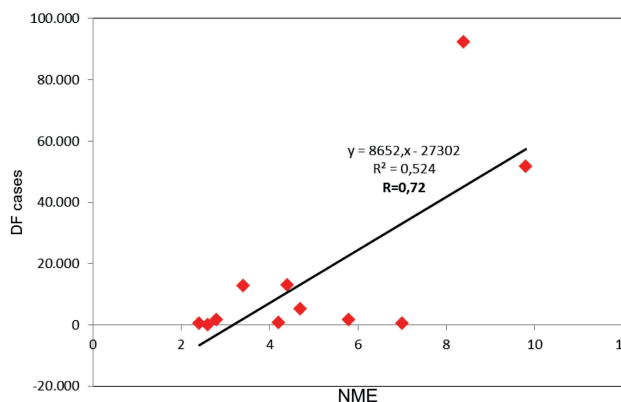
**Figure 5** Values of mean monthly of rainfall daily and the entomological indexes, mean number of eggs (MNE), July/2002 and June/2013

The fourth instar larvae (L4) reared in the laboratory from field-collected eggs used for species identification showed that *Ae. aegypti* was a dominant species (90.8% - 99.2%) in study periods (Figure 6).



**Figure 6** Proportion of larvae species identified in the entomology laboratory from field-collected hatched eggs (2002-2013)

Simple Linear Regression plotting the MNE from August-September with the occurrence of cases in later years showed  $R=0.72$  with  $p<0.01$ (Figure 7)



**Figure 7** Linear regression yearly DF cases compared with the MNE for the months Aug-Sept previous year (2002-2013).

### DISCUSSION

Giovanini et al, 2008, demonstrated that the relation between the Larval Index (LI) and the rate of growth of *dengue* epidemic may be obtained almost two months before the beginning of *dengue* outbreaks and could serve as a guide for early intervention. However, it was concluded that the larval index (LI) is not reliable as a predictor of *dengue* incidence, a fact already known for other LIs<sup>18</sup>. The larval surveys, the most used *dengue* vector surveillance, classically based on the House Index (HI) and the Breteau Index (BI), have limitations because of the larvae’s ability to escape rapidly and their capacity to remain submerged for long periods<sup>19</sup>.

Câmara et al, 2010, speculated that unpredictable fluctuations in the local vector indexes potentiate outbreaks in areas where vector indexes are supposedly under control. Epidemics are initially in places delimited before broaden its geographic boundaries. It does not seem possible to prevent epidemics of *dengue* vector even if the indexes are low and pragmatic steps are taken to eliminate breeding sites are taken<sup>20</sup>. However, they take unpredictable measures of local vector indexes as a premise. The ovetraps surveillance could be an alternative to enhance the knowledge of the vector indexes in smaller areas.

It’s frustrating that timely prediction does not correspond to actions that prevent the occurrence of epidemics. Makay et al, 2013, developed an autocidal gravid ovetrap (AGO) as a simple, low-cost device for surveillance and control of *Ae aegypti* without the use of pesticides that does not require servicing for an extended period of time. They concluded that the AGO can be used to efficiently attract and capture gravid *Ae aegypti* females with low cost<sup>21</sup>. Newer tools are proposed at the surveillance of adult *Ae aegypti* and for controlling this vector using integrated means: adult mortality caused by a number of insecticide-impregnated

tools that are currently undergoing field testing, such as curtains and covers for water-storage vessels<sup>22</sup>, bednets<sup>23</sup>, and “lethal” ovitraps<sup>19</sup>. All these tools need to be integrated with a good surveillance system, and the ovitraps surveillance can be the best solution for this integration.

## CONCLUSION

These results showed that the ovitrap surveillance, a simple and easily adaptable device, is a feasible tool for alert of *dengue* incidence two months earlier and has a good predictive power. It is important to draw attention to the strategy multi-institutional and multi-disciplinary components with integrated use of spatially information technology, entomological and epidemiological knowledge in a common objective<sup>24</sup>. This strategy was designed to help increase the competence of the control of transmissible

diseases, by providing new tools for surveillance and control systems, including environmental aspects, risk factor detection, and automatic warning methods. It should provide more efficient outbreak detection. The special interest would be to get a tool on data referring with a framework so that we would be able to gain a proxy into the predictability of upcoming *dengue* outbreaks. This would help to allow control measures and therefore to guide policies of prevention and control of the *dengue* virus transmission.

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