

Quantitative monitoring of *Aedes albopictus* in Emilia-Romagna, Northern Italy: cluster investigation and geostatistical analysis

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Abstract

The *Aedes albopictus* (Skuse) (Diptera Culicidae) control program currently applied in the Emilia-Romagna region (Northern Italy) is based on the use of ovitraps as a tool for mosquito population density estimation. During the favourable season 2008 (May-October), 2,741 ovitraps were activated in the urban areas of 242 municipalities according to standard criteria and were checked weekly. The universal kriging interpolation was used to estimate the seasonal abundance of the species at unsampled locations, and spatial cluster analysis was used to identify particular areas that had statistically significant high or low mosquito density. The overall data pattern was highly clustered and autocorrelated, and the choropleth and LISA cluster maps showed high egg density in the North, North-East and in the South-West areas of the region. The cross-validation statistics and results showed that the predicted values were reasonable for map production. The characterization of large geographic areas with high or low abundance of *Ae. albopictus* may provide information both on the environmental variables that promote species dispersion, and on the epidemic diseases risk, essential to develop effective disease surveillance programs, particularly for Chikungunya and Dengue.

Key words: *Aedes albopictus*, ovitraps, geostatistics, cluster analysis, monitoring, Chikungunya.

Introduction

An outbreak caused by Chikungunya virus (CHIKV), never previously reported in Europe, occurred in Emilia-Romagna Region, Italy, in December 2007, with 247 identified cases between July and September (Angelini *et al.*, 2007). The CHIKV causes a nonfatal, self-limiting disease characterized by abrupt onset of high fever, severe arthralgia, or arthritis, often associated with skin rash.

Aedes albopictus (Skuse) (Diptera Culicidae) was indicated as the primary vector of CHIKV in Italy (Bonilauri *et al.*, 2008). The first detection of *Ae. albopictus* in Emilia-Romagna Region dates back to 1994 (Carrieri and Bellini, unpublished data).

This species lay eggs in a variety of shaded artificial containers, particularly in the catch basins in urban areas; eggs hatch within one or two days after immersion, during the warm season.

The development time to the adult state may take from 6 to 10 days. Adults disperse at a maximum distance comprised in the range of 600-800 m (Honorio *et al.*, 2003; Liew and Curtis, 2004).

Population density was measured in many mosquito control programs in several Italian cities by means of monitoring systems based on specific ovitraps (Bellini *et al.*, 1996).

Ovitraps are largely used worldwide to monitor containers breeding mosquito species (Thaggard and Eliason, 1969) and their reliability in terms of quantitative estimation of the adult population density is controversial and questioned by several authors (Hawley, 1988; Holck *et al.*, 1988; Zhang and Lei, 2008).

In the last years, the use of Geographic Information System (GIS) has given important practical contributions to the investigation on the spatial component of the epidemiology of infectious diseases (O'Dwyer and Burton, 1998), including vector-borne diseases such as malaria, trypanosomiasis, rickettsiosis, and a range of arboviral diseases (Liebhold *et al.*, 1993; Kitron, 1998; Brooker *et al.*, 2002).

Moreover, the collection of georeferenced epidemiological sensitive data can be useful also for cluster identification and geostatistical analyses. The investigation on possible disease and vector born-disease clustering is fundamental to epidemiology and medical entomology, with one of the aims being to determine whether the clustering is statistically significant and worthy of further investigation, or whether it is likely to be a chance occurrence. Global and local indicators of spatial association like Moran's I (Cliff and Hord, 1981) or Getis-Ord Statistics (Getis and Ord, 1992) are often used to measure the data clustering level. Geostatistical techniques are used to produce prediction surfaces and also an error or uncertainty surfaces, giving an indication of how good the predictions are.

Since 2006, the Emilia-Romagna Region Public Health Department has started a vector surveillance and risk assessment program that includes the implementation and management of a monitoring system based on georeferenced ovitraps and the development of a specific GIS. This study is part of a project that aims to develop a large scale *Ae. albopictus* monitoring network, based on the mean egg density. This monitoring method can be achievable at low cost but need to be well-designed in order to provide reliable information for the

estimate of the infestation level in the large urban areas (exceeding 600 hectares), where the health risk of mosquito vectored diseases is higher.

We explored the spatial structure of *Ae. albopictus* using the data collected from the ovitrap network in the season 2008. The number and arrangement of the ovitraps had been defined on the base of the data of the 2007 season by Carrieri *et al.* (unpublished data). Our specific aim was to detect significant clusters of abundance and to depict, by means of geostatistical analysis, a continuous surface map of *Ae. albopictus* density in the study area.

The definition of large continuous geographic areas with high or low abundance of *Ae. albopictus* may provide information on the environmental variables that promote species dispersion, useful to implement the disease risk surveillance programs to prevent CHIKV and Dengue epidemic.

To achieve this aim, the mean egg density data collected by ovitraps in the season 2007 were used to assess the mosquito population's aggregation degree, through the application of the Taylor's power law, and to define the minimum sample size to set-up a monitoring design for the year 2008 proficient to ensure a high degree of accuracy at the provincial and municipal scale. The reliability of the method and its efficiency were assessed in the season 2008 measuring the relative variation.

Materials and methods

Study area

The Emilia-Romagna Region is situated in the middle North of Italy, lying between 9°11' and 12°45'E longitude and 43°44' to 44°59'N latitude. This region is bounded by the Apennine Mountains to the South and West, by the Adriatic Sea to the East, and by the Po River for most of the northern border (figure 1). Respectively, 47% and 27% of the territory is represented by lowlands and hills, with a warm humid climate, characterized by mean daily summer temperature of 24-30 °C (May-September) and a mean relative humidity of about 60% (ARPA, 2008).

The area has a mean annual rainfall of 600 mm, occurring for 60% between April and October (ARPA, 2008). In 2008, the region included 341 municipalities for a total inhabited area of 22,122 km². Seventy percent of the 4,275,843 (ISTAT, 2009) inhabitants live in the lowlands.

Mosquito's egg sampling

The optimal number of ovitraps to be placed in an urban area varies as a function of the species density and dispersion; these parameters depend on weather trend, stage of colonization and other environmental conditions (vegetation, breeding sites, etc.). At the initial step of colonization the density of the species is patchy and aggregated (low mosquito density and high data aggregation) and more ovitraps are needed than in areas at a mature step of colonization (high mosquito density and more uniform spatial dispersion) in order to achieve the same reliability level.

We calculated the minimum number of ovitraps needed according to the Taylor's equation (Taylor, 1961; Taylor, 1984; Kuno, 1991). This function has been largely used to quantify the aggregation degree and the statistically significant sample size in insects monitoring:

$$(1) \quad S^2 = a * m^b$$

b measures data aggregation (which is a constant of the species), and, when greater than 1, indicates a relationship between mean and variance, i.e. that data are aggregated; a is a constant depending on environmental conditions. Both are necessary to define the minimum sample unit through the following equation:

$$(2) \quad N = [Z_{\alpha/2}/D]^2 * a * m^{b-2}$$

where Z is the Standard Normal Distribution Value for a given probability (Buntin, 1994); D is the precision level, and according to literature, $D = 0.1$ is considered a sufficient value (Southwood, 1978) while a D value between 0.2 and 0.3 is considered optimal for the binomial sampling of *Ae. aegypti* (Mogi *et al.*, 1990); m is the mean eggs density value.

In our study, 242 municipalities participated to the monitoring network, and we referred to regional coordination of information on the environment (CORINE) Land Cover 2003 to individuate the classes (continuous urban fabric, discontinuous urban fabric, industrial or commercial units) which were considered as inhabited areas and covered a total area of 1,050 km². Every inhabited area was divided into a number of quadrants equal to the minimum number of ovitraps to be placed; 2,741 ovitraps were positioned, and the distance between two ovitraps varied from 200 to 800 m, according to the number of quadrants per area.

Ae. albopictus eggs were sampled weekly (from May to October). Each ovitrap was constituted by a black conic plastic cup (400 ml volume, upper diameter 8 cm, lower diameter 6 cm), filled up to 2/3 of its height with 285 ml of de-chlorinated water (Celli *et al.*, 1994). A masonite strip of 12.5 x 2.5 cm was used as egg deposition substrate.

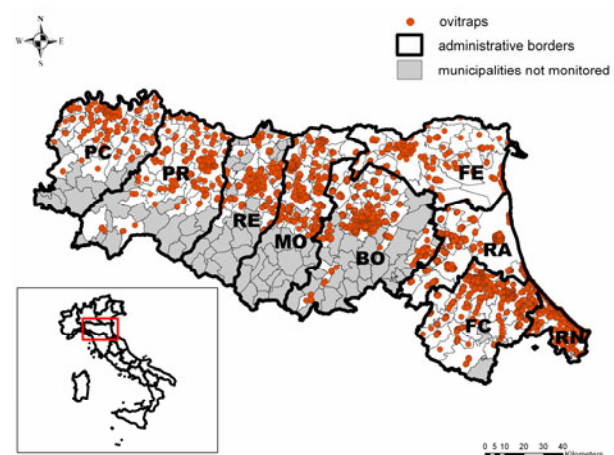


Figure 1. Emilia-Romagna Region map (Northern Italy). Abbreviations of nine provinces: PC = Piacenza, PR = Parma, RE = Reggio-Emilia, MO = Modena, BO = Bologna, FE = Ferrara, RA = Ravenna, FC = Forlì-Cesena, RN = Rimini.

The ovitraps were positioned at the beginning of the season by a staff of skilled technicians, according to precise instructions and rules to assure the maximum homogeneity of the microhabitat conditions (in a green, shaded and easily accessible area, laying on the ground, with a free space above of at least 1 meter) and were maintained unchanged until the end of the season. Ovitrap were georeferenced in the field by using Global Positioning System (GPS) equipped palmtops and entered into GIS with the proper address, in order to be easily found by the technicians. Every week the masonite strips were changed and sent to ARPA (Regional Agency for Environmental Protection in Emilia-Romagna) for eggs counting by stereomicroscope. All data were weekly published on a dedicated web site (www.zanzaratigreonline.it).

Cluster analysis

Monitoring data of each municipality were aggregated into eggs number/ovitraps/week calculating the mean of the 22 sampling weeks from May to October 2008.

We evaluated whether these data were autocorrelated by calculating the Global Moran's I index, while the statistically significant difference against the null hypothesis (the absence of spatial autocorrelation) was tested using the permutation procedure (Anselin, 1996).

In global spatial statistics, local and small areas of spatial heterogeneity are often masked by averaging the spatial pattern over the entire study area into a single value of spatial autocorrelation. In order to avoid this occurrence, Local Indicator of Spatial Association (LISA) (Anselin, 1995) was used as a spatial exploratory tool to detect localized spatial structure that could indicate a lack of stationarity (Fortin and Dale, 2005), i.e. to assess if municipalities formed statistically significant high or low abundance clusters.

GeoDa package (www.geoda.uiuc.edu) (Anselin, 2003; 2005) was used for both the global and local autocorrelation analyses and ESRI ArcGIS for map drawing.

Weight distance matrix, essential for the computation of spatial autocorrelation statistics, was based on queen contiguity and Euclidean distance.

Geostatistical analysis

Geostatistics assumes that at least some of the spatial variations of natural phenomena can be governed by random processes indicating that a certain degree of interdependence is present between the values of a variable at different geographic scales to which we refer to as spatial autocorrelation.

Many methods are available in the geostatistical analysis. The kriging interpolation method can accomplish two distinct tasks: quantifying the spatial structure of the data and producing a prediction. Quantifying the structure, known as variography, consists in a spatial dependence model fitted to the data (variogram). To predict an unknown value for a specific location, the kriging interpolation method uses the fitted model obtained from variography, the spatial data configuration and the values of the measured sample points around the prediction location (Krishna Murthy and Abbaisah, 2007).

According to Service (1993), the efficiency of the sampling design for each municipality was evaluated

using relative variation (RV):

$$(3) \quad RV = \text{standard error}/\text{mean}$$

An RV of 0.3 has been considered the upper limit to define the sampling design as statistically adequate, according to the RV calculated for *Ae. aegypti* binomial sampling by Mogi *et al.* (1990).

For the geostatistical analysis, the centroid was calculated for each of the monitored polygons (that identified the municipalities) with a mean RV < 0.3 calculated over the 22 weeks of sampling.

Exploratory Spatial Data Analysis (ESDA) provided by ArcGIS Geostatistical Analyst extension was used to identify the distribution of the data, the global trends, the global and local outliers and the spatial autocorrelation.

Stationarity and independence of the data were verified by the Kolmogorov-Smirnov test (StatSoft, 2001; Statistica 6). If the data were stationary, we analyzed them geostatistically. If data were not stationary, we proceeded to data transformation and data de-trending through a mathematical formula, and the non-random (deterministic) component of a surface was removed from the data in order to obtain a normal distribution.

The presence of spatial autocorrelation among centroids data was performed calculating the Local Moran's I index on five lag classes of 7.5 km each (one lag was intended as the half of the maximum distance between centroids, as they were not uniformly distributed). The maximum lag distance (37.5 km) measured less than one-half of the smallest dimensions of study area (i.e. the mean North-South dimension, measuring 90 km). Logarithmic ($y + 1$) transformation of the data was used to homogenize variance.

Correlograms were tested for spatial autocorrelation significance using Monte Carlo simulation and progressive Bonferroni correction (Legendre and Legendre, 1998) to adjust for repeated testing; the analysis was performed by ROOKASE Software (Sawada, 1999).

The best experimental variogram model was identified fitting our data by using VARIOWIN (Pannatier, 1996) and the universal kriging interpolation was used to estimate species abundance at unsampled locations throughout the study area, on the base of the mean egg density of each municipality. Local averaging of mean egg density data was based on a search radius of 30 km (maximum positive significant autocorrelation) in order to include a minimum of 20 data, being the nugget more than half of total sill height (Welhan, 2004).

The universal kriging interpolation is an adaptation of the ordinary kriging method that accommodates trend or non-stationarity in the mean, for example when large variation in local means obtained from different geographic areas occur (Ryan *et al.*, 2004).

A leave-one-out cross validation method was used to determine whether the universal kriging interpolation provided reliable estimates of mean egg density at unsampled locations. The criteria used for accurate prediction in the cross-validation were requested to be the followings: slope line near to 1, mean error close to zero, root mean square error average standard error as small as possible and standardized root mean square error close to 1.

Analyses were conducted using ArcGIS Geostatistical Analyst (ESRI Geostatistical Analyst Tutorial).

Results

Cluster analysis

A total of 2,710,668 eggs were collected by the 2,741 ovitraps over 22 weeks of sampling (from week 21 to 42). The mean egg density calculated for each municipality ranged from 0 to 159 eggs/ovitraps/week as reported in choropleth map (figure 2).

The cluster analysis was conducted on all the municipalities' data sets ($N = 242$) and those municipalities with $RV > 0.3$ were evidenced by wired polygons in the maps (figures 3a and 3b). The overall data pattern was highly clustered and autocorrelated (Global Moran's $I = 0.25$; $p < 0.01$).

After the global autocorrelation study, using Univariate LISA analysis by GeoDa package we obtained a cluster map (figure 3a) with the respective significance map (figure 3b). Municipalities areas that formed high density clusters were evidenced in red, while municipalities forming low density clusters were evidenced in blue. The choropleth and LISA cluster maps showed high egg density in the north, north-east and in the south-west areas of the region. In table 1, the number of municipalities forming high and low clusters, divided per province, is reported.

Geostatistical analysis

Those municipalities with $RV > 0.3$ were excluded from the geostatistical analysis because their patterns resulted not adequate to measure mosquito population and to produce a smoothed interpolation map.

The analysis was conducted only on the municipalities' data sets which showed a sufficient degree of monitoring precision ($RV < 0.3$; $N = 160$). The nearest neighbour distance between centroids varied from 2,233 to 14,803 m, with a mean of 6,272 m.

Mean egg density frequencies and log transformation data resulted stationary (Kolmogorov-Smirnov Test: $D = 0.10$; $p < 0.05$).

In table 2, we reported local Moran's I analysis. Spatial autocorrelation was greatest (0.42) at the lag 0-7.5 km, medium at the second lag and low in the last three lags. The correlogram expressed a linear correlation with significant high positive values at the short distances (0-7.5 km) and low positive ones at large distances (22.5-30.0 km) local autocorrelation was statistically significant ($p < 0.05$) for the first 4 lag distances.

Trend analysis results were presented in three-dimension perspective trend plots for the egg number/ovitraps/week calculated for the municipalities with $RV < 0.3$ (figure 4).

The detrending of data was not applied due to the complexity of the polynomial approximating trend surface, and because the polynomial trend surfaces accelerated without limit to higher or lower values in areas where there were no control points, such as along the edges of the maps.

Outliers and directional influence in the spatial data were also examined by using semivariogram cloud/surface in ArcGIS Geostatistical Analyst. No particular outliers and directional influence were found.

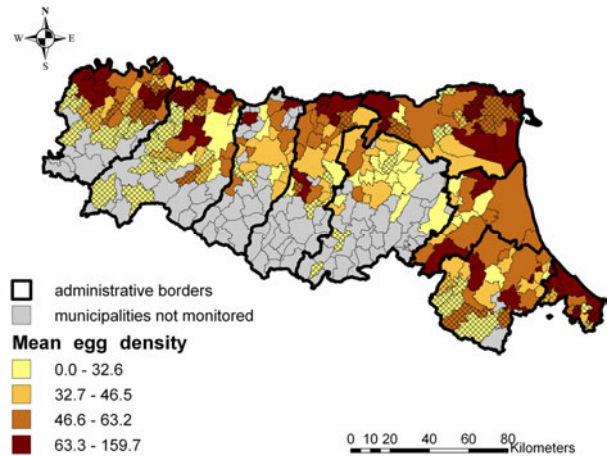


Figure 2. Choropleth map of mean egg density (number eggs/ovitraps/week) calculated for 22 monitoring weeks. (Legend values are subdivided into quartiles; wired polygons represent municipalities with a sampling design not statistically efficient to measure real population densities for $RV > 0.3$).

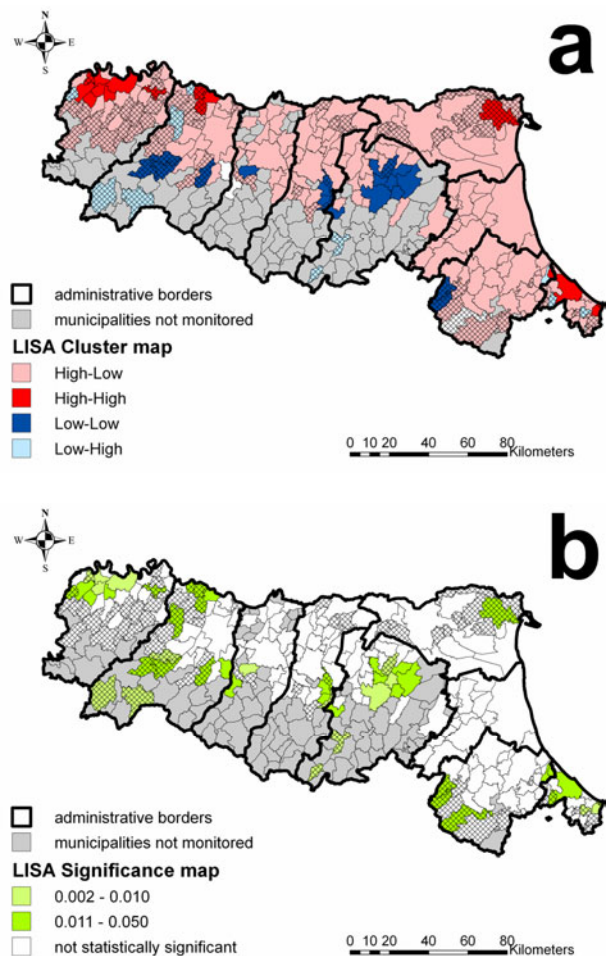


Figure 3. Local Indicator Spatial Association (LISA): cluster (a) and significance (b) maps. (Wired polygons represent municipalities with a sampling design not statistically adequate to measure real population densities for $RV > 0.3$).

Then we performed variogram analysis by using VARIOWIN and the best model obtained from the data was the spherical model with 0.11 as nugget (the variance at zero distance), 60 km as range (beyond which the semivariance is constant), 0.09 as partial sill (the constant semivariance value beyond the range). Model parameters obtained from variogram analysis were used in ArcGIS Geostatistical Analyst for obtaining the interpolation map (figure 5a).

Table 1. Number of municipalities forming high (High-High) and low (Low-Low) *Ae. albopictus* density clusters. In brackets, municipalities with a statistically adequate monitoring design for measuring species population ($RV < 0.3$).

Province	N. municipalities number forming clusters	
	High-High	Low-Low
Bologna	0	9 (7)
Ferrara	1 (0)	0
Forli-Cesena	1 (0)	2 (0)
Modena	0	4 (2)
Parma	3 (1)	4 (0)
Piacenza	8 (6)	0
Ravenna	0	0
Reggio-Emilia	0	1 (1)
Rimini	3 (2)	0

Table 2. Local spatial autocorrelation Moran's I calculated for 5 lags distance of 7.5 km each. Pairs' number and Z-values for each index were calculated.

Lag Increment (LI) (km)	Neighbour pairs (N)	Moran's I	Z-value
0 < LI = 7.5	123	0.42**	4.79
7.5 < LI = 15.0	435	0.34*	7.30
15.0 < LI = 22.5	578	0.18*	4.59
22.5 < LI = 30.0	677	0.17*	4.89
30.0 < LI = 37.5	727	0.17	4.97

* $p < 0.05$, ** $p < 0.01$

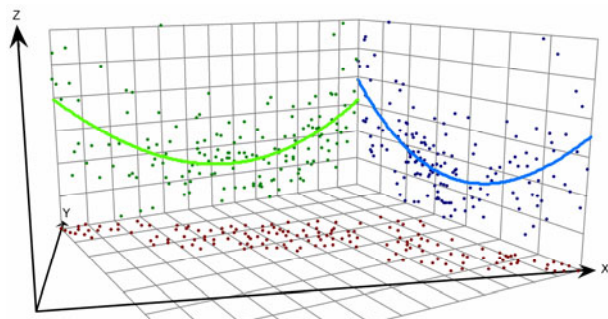


Figure 4. 3D trend plot of mean eggs/ovitraps/week calculated for the period between week 21 and 42, for municipality areas with $RV < 0.3$ ($N = 160$). The X and Y axes represent the coordinates of each municipality centroid, while Z axis represents the mean egg density value.

The quality of the prediction map has been examined by creating the prediction standard error surface (figure 5b). The prediction standard errors quantified the degree of uncertainty for each location in the surface. Standard errors map showed low errors in the province of Bologna, Modena, Reggio-Emilia, Parma and Ravenna, high and medium errors in the province of Ferrara, Rimini and in particular Piacenza.

The Cross-validation statistics (table 3) and results (figure 6) showed that the predicted values were reasonable for map production (linear regression analysis, $R^2 = 0.25$; $p < 0.05$; $y = 0.305x + 36.84$).

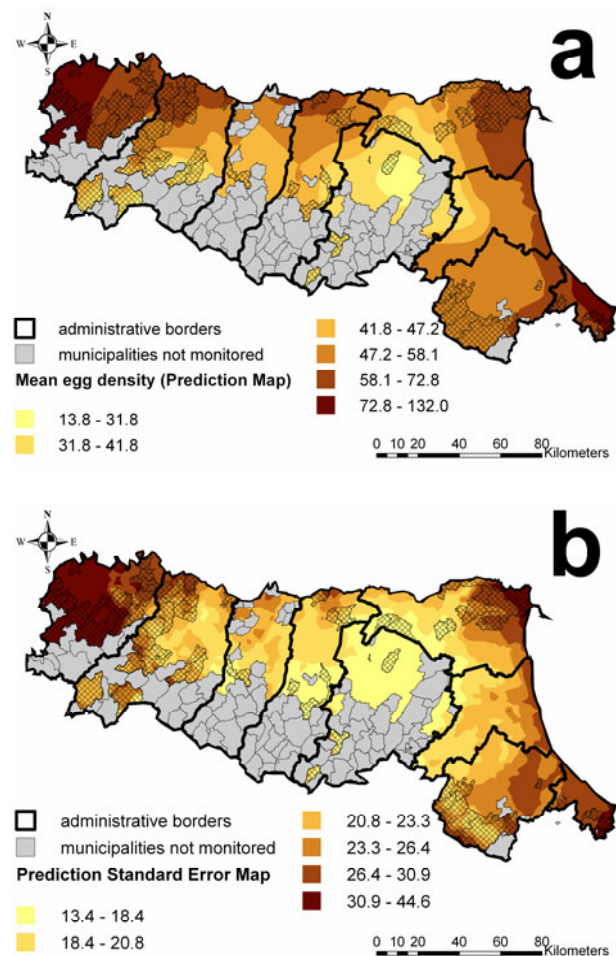


Figure 5. Universal kriging interpolation map (a) and standard error map (b) of mean egg density in Emilia Romagna Region (municipalities with $RV > 0.3$ were not considered in the interpolation calculation, and were indicated in the map with wired areas).

Table 3. Cross-Validation statistics.

Parameter	Value
Mean Error	-0.060
Root-Mean-Square Error	2.400
Average Standard Error	23.400
Mean Standardized	-0.002
Root-Mean-Square Standardized	0.880
Slope Line	0.270

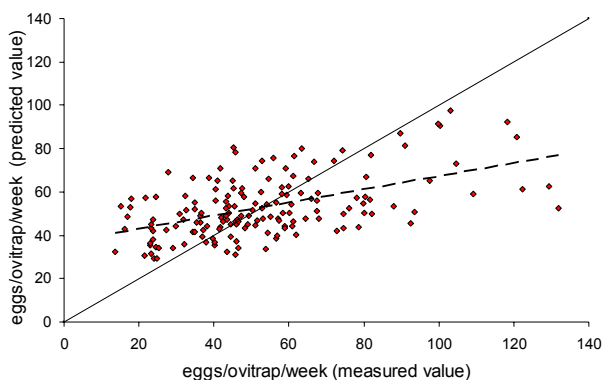


Figure 6. Cross validation results. Scatter plot of the predicted versus measured values (the slope is lower than one; the kriging interpolation tends to underpredict large values and overpredict small values).

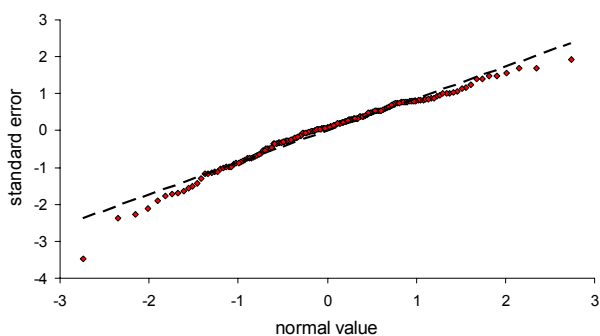


Figure 7. QQPlot regression results. This shows the quantiles of the difference between the predicted and measured values divided by the estimated kriging standard errors and the corresponding quantiles from a standard normal distribution.

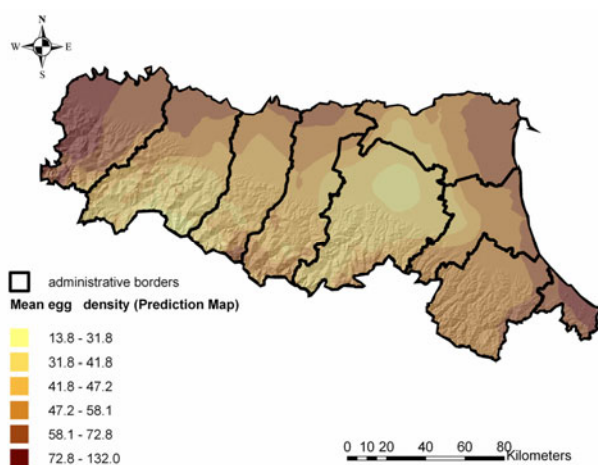


Figure 8. Example of environmental informative layers overlay: shaded elevation map (Void-filled seamless SRTM data V2, 2006, International Centre for Tropical Agriculture (CIAT), available from the CGIAR-CSI SRTM 90m Database: <http://srtm.csi.cgiar.org>) overlaid to interpolated egg density map.

Cross-validation showed low errors near municipalities with about 53 eggs/ovitraps/week (intercept between the 1:1 correlation line and the best fit line; figure 6) and large errors at higher egg density. The QQPlot regression ($R^2 = 0.97$; $p < 0.05$; $y = 0.872x - 0.002$) (figure 7) showed that some values, in particular the high ones, fall slightly far from the straight dashed line, but most points fall very close to it, indicating that prediction errors were close to be normally distributed.

The correlation calculated between mean egg density and elevation classes resulted high ($R^2 = 0.88$; $p < 0.05$; $y = -9.50x + 63.72$). In figure 8, the shaded elevation map, acquired from satellite images, is overlaid to the interpolated egg density map. Both layers show a similar spatial trend indicating the relationship found between ovitraps data and altitude.

This is only one example of the possible comparisons with other informative layers, such as NDVI (Normalized Difference Vegetation Index), temperature and rainfall distributions, land use/land cover maps.

Discussion and conclusions

In this study we evaluated the usefulness of standard ovitraps monitoring methodology, in combination with GIS, geostatistical analysis and computer-based mapping techniques as practical tool for entomological and epidemiological studies and operational use.

Data analyses showed that the regional *Ae. albopictus* monitoring system adopted was sufficiently reliable to determine spatial variation within *Ae. albopictus* data at municipality level. In fact our results indicated that *Ae. albopictus* mean egg density data, aggregated for municipality were spatially correlated and significant at a distance less than 30 km, in particular between 0 and 7.5 km, and cross-validation results indicate that estimated egg density at unsampled locations were reasonably acceptable with some limits due to the not uniform distribution of the data.

Mean egg density data aggregated for municipality were sufficient to produce a spatial interpolation at the municipality level, while it was not yet sufficient to produce spatial interpolation at the locality level because the number of locality data with a sufficiently reliable monitoring pattern ($RV < 0.3$) were very low (only 20 localities), not sufficient for trying an interpolation over the whole region.

Extrapolation and interpolation of data need to be conducted with caution, and the production of computer-generated maps that seem more informative than the data upon which they are based should be avoided. Anyway, contour smoothed maps obtained from geostatistical analysis and cluster maps obtained from cluster detection could be overlaid to other smoothed informative layers to identify environmental variables such as elevation, rainfall distribution, mean temperature, relative humidity that could influence seasonal mosquito population density in the region or could be overlaid to epidemiological data to identify health risk.

Our work was not aimed to find out a risk threshold of *Ae. albopictus* population density for the spread of an

epidemic of mosquito-borne disease, but instead to be the first leap to develop a practical instrument of evaluation of mosquito population distribution over the whole region, aimed to make prediction, verify hypotheses, assess the efficacy of mosquito control programs on large and local scale.

Another field of application for the spatial analyses of *Ae. albopictus* egg density data could be the evaluation of the efficacy of the control programs performed in different municipalities, whose quality significantly affect the mosquito population and the ascription of a municipality to a high or low egg density cluster.

The comparison between the two municipality groups forming clusters, showed that in 2008 *Ae. albopictus* control programs conducted in municipalities with low mosquito density were qualitatively different from those applied in municipalities with high mosquito density, as evidenced by the survey on some key aspects. The former were characterized by more than five years of *Ae. albopictus* survey and control programs, a standardized high quality control on larvicidal treatments that allow to reduce the number of treatments in public catch basins and the number of adulticides treatments, thanks to a more organized entomological inspection service in private properties after citizens' complains.

A quantitative difference in the mosquito control programs between the two cluster municipality areas is documented by comparing the mean budgets invested: 49.1 €/ha by the municipalities of the low mosquitoes density clusters (that adopted lasting and high quality control programs, and 38.4 €/ha, 27% less, by those municipalities of the high density clusters (that have incomplete or newly introduced control programs).

The results of our study encourage the adoption of geostatistics and in general of spatial statistics as efficient tools for *Ae. albopictus* surveillance and monitoring systems. To summarize, some advantages of the use of these spatial techniques are:

- 1) Cluster detection/investigation identifies high and low significant mosquitoes density areas to be considered in mosquito control planning.
- 2) Smoothed contour maps summarize in one frame/layout of simple reading a big amount of monitoring data.
- 3) Large scale interpolation maps of mean egg density are useful for understanding environmental variables, represented by smoothed contour maps, that could promote species development (elevation, rainfall, vegetation covering, etc.).
- 4) Cluster investigation and geostatistics could help to understand, at local scale, where mosquito control programs are achieving satisfactory results and where they must be strengthened to better suppress *Ae. albopictus* populations.
- 5) In our study, the smoothed map obtained through the interpolation of data with an $RV < 0.3$ allowed to make predictions on the egg density in those municipalities where the monitoring design was not statistically efficient, and standard error map evidenced large critical areas in the Ferrara, Rimini and Piacenza provinces, in which the monitoring pattern need to be increased in order to obtain a better and significant interpolation.
- 6) Geostatistics and cluster detection/investigation can provide risk assessment maps useful in epidemiological studies, and could be of crucial importance for defining an epidemiological threshold.

On the other side, critical points in adopting geostatistical analysis of entomological data for creating large scale interpolation maps can be found in the difficulty of assessing a standard procedure to find the best variogram model, and in finding the appropriate dataset (mean eggs, total eggs, rank, etc.) to satisfy the prerequisite of data stationarity, which is necessary for obtaining the best interpolation.

Moreover, while the spatial geostatistical techniques used to determine *Ae. albopictus* population at the large scale are suitable for static data collected in a certain time period, they do not include any reference about the dynamics of population data. To gain this level of information, it requires at least a monthly updating of the local monitoring data. It will also be necessary to pool the data collected over several years, being not possible yet to implement the number of monitoring stations, due to insufficient funding availability.

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