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Source: Journal of Medical Entomology, 49(3):722-730. 2012.

Published By: Entomological Society of America

DOI: <http://dx.doi.org/10.1603/ME11156>

URL: <http://www.bioone.org/doi/full/10.1603/ME11156>

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Processes Affecting *Aedes aegypti* (Diptera: Culicidae) Infestation and Abundance: Inference Through Statistical Modeling and Risk Maps in Northern Argentina

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J. Med. Entomol. 49(3): 722–730 (2012); DOI: <http://dx.doi.org/10.1603/ME11156>

ABSTRACT Understanding the processes that affect *Aedes aegypti* (L.) (Diptera: Culicidae) may serve as a starting point to create and/or improve vector control strategies. For this purpose, we performed statistical modeling of three entomological surveys conducted in Clorinda City, northern Argentina. Previous ‘basic’ models of presence or absence of larvae and/or pupae (infestation) and the number of pupae in infested containers (productivity), mainly based on physical characteristics of containers, were expanded to include variables selected a priori reflecting water use practices, vector-related context factors, the history of chemical control, and climate. Model selection was performed using Akaike’s Information Criterion. In total, 5,431 water-holding containers were inspected and 12,369 *Ae. aegypti* pupae collected from 963 positive containers. Large tanks were the most productive container type. Variables reflecting every putative process considered, except for history of chemical control, were selected in the best models obtained for infestation and productivity. The associations found were very strong, particularly in the case of infestation. Water use practices and vector-related context factors were the most important ones, as evidenced by their impact on Akaike’s Information Criterion scores of the infestation model. Risk maps based on empirical data and model predictions showed a heterogeneous distribution of entomological risk. An integrated vector control strategy is recommended, aiming at community participation for healthier water use practices and targeting large tanks for key elements such as lid status, water addition frequency and water use.

KEY WORDS dengue, *Aedes aegypti*, Akaike’s Information Criterion, vector control

Dengue is currently the most important arboviral disease of humans, with an estimated 50–100 million annual cases of dengue fever and 250,000–500,000 annual cases of its most severe forms, dengue hemorrhagic fever and dengue shock syndrome (Kroeger and Nathan 2006, Kourí et al. 2007). *Aedes aegypti* (L.) (Diptera: Culicidae), its main vector, is a highly anthropophilic and domestic vector that breeds mainly in artificial water-holding containers (Kyle and Harris 2008).

Understanding the processes that determine habitat suitability, distribution and abundance of the mosquito is crucial for evaluating, improving, and/or creating new forms of vector control for the prevention of disease transmission (Morrison et al. 2008). Statistical modeling is often used to test associations between selected explanatory variables and a response variable under study. Causal relationships (processes) may be inferred from the associations to explain the patterns observed. Another use of the outcome of

statistical models is the construction of risk maps that allow spatial stratification of areas with higher transmission risk and gain insight of its determinants (Kitron 2000). In the case of epidemiology and the study of disease vectors, inference is almost always performed based on observational studies (experiments are frequently unethical or impractical), a feature that makes the search for causality a difficult task (Freedman 1999). To draw sound causal inferences from observational data and avoid the perils of data mining, convergent lines of evidence should be developed and modeling should be performed thoughtfully, based on previous knowledge and a priori selection of variables and model structures (Burnham and Anderson 2002).

Processes affecting *Ae. aegypti* range from the scope of ecology, meteorology to social sciences. Ecological factors affecting larvae and pupae are generally related to physical characteristics of containers determining their suitability as habitats. For example, in the Peruvian Amazon (Morrison et al. 2004), containers unlidged, located outdoors, and rain-filled were associated with increased production of *Ae. aegypti* whereas in Puerto Rico, this was the case for containers located in yards, in the shade of trees, unattended, and rain-filled (Barrera et al. 2006). More recently,

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habitat density was also associated to container productivity (Aldstadt et al. 2011). The most common climatic factors generally recognized to influence *Ae. aegypti* are temperature and rain. However, several studies have reported disparate results concerning the importance of weather variables and lagged effects (e.g., Scott et al. 2000, Estallo et al. 2008, Azil et al. 2010). From a more anthropological perspective, water use practices have been found associated with *Ae. aegypti* production (Caprara et al. 2009) probably influencing habitat suitability.

A temephos-based citywide larval control program conducted from 2003 to 2008 in Clorinda, northwestern Argentina, limited dengue transmission but failed to reduce infestation to recommended levels (Gürtler et al. 2009). Large tanks were the most productive containers despite being the main target for control. This probably occurred because the residual effects of temephos in field containers were highly limited by water use practices (Garelli et al. 2011). Statistical modeling showed that containers located in yards, at low sun exposure, unlidded, filled with rain water, and holding polluted water were found to be positively associated with infestation by larvae or pupae (Garelli et al. 2009). However, this analysis did not include other relevant variables likely to affect *Ae. aegypti*, for example, anthropological or meteorological factors.

The objective of this study is to construct improved statistical models of container infestation and productivity by *Ae. aegypti* that build upon our previous analyses of processes operating in Clorinda. The goal of the modeling effort is to infer processes affecting the vector; find the most likely causes of mosquito occurrence and abundance by assessing the importance of different hypothetically relevant variables, and based on model predictions, construct risk maps to identify areas of higher entomological risk. The variables considered in the models were a priori selected to reflect several putative processes and factors affecting the mosquito such as water use practices, vector-related context factors, history of chemical control, container characteristics, and climate. Understanding which processes affect *Ae. aegypti* may serve as a starting point to create and/or improve current vector control strategies.

Materials and Methods

Study Site. The city of Clorinda (latitude 25° 17'S, longitude 57° 43'W) had 47,240 inhabitants in 2001 and is located in the Province of Formosa in northern Argentina on the border with Paraguay. This study was carried out in Primero de Mayo, a large neighborhood with 2,500 houses (20% of the city) and relatively high infestation levels (house and Breteau indices averaging 10.7 and 13.7%, respectively). The piped (tap) water service in the neighborhood is intermittent and insufficient, especially during summer.

Entomological Surveys. Surveys were carried out in the spring of 2007 (between 8 October and 29 November), fall of 2008 (15 April–16 May), and spring of 2008 (5 November–17 December). During the 2008

spring survey the location of the entrance to each lot in the neighborhood was georeferenced with a GPS receiver (Trimble GeoXM or Garmin Legend). All 2,488 lots in the neighborhood were visited in each survey. During the first two surveys, if a house was found closed or refused inspection, it was revisited within the following 2 wk at a different time of the day. The surveys were carried out by experienced personnel of Fundación Mundo Sano (FMS) supervised by the research team. Upon visiting a household, the household head was asked for oral consent to examine the premises. The yard and the interior of each household were thoroughly inspected for containers. All water-holding containers found were examined for mosquito immature stages, taking samples of larvae and collecting every pupa detected with large-mouth pipettes; frequently the operators used small sieves to strain the containers. Collected immatures were placed in test tubes, labeled and transported to the laboratory for processing and counting. Larvae were identified to species using an entomological magnifying glass and an illustrated key (Rossi and Almirón 2004). Pupae were kept in small water-filled plastic vials until emergence for accurate species identification and counted as adults.

Containers were classified according to a new scheme constructed based on field personnel annotations. The previous classification used in Clorinda since 2003 included seven categories: tires; large tanks, drums or barrels; flower pots; construction materials and discarded vehicle parts; bottles, cans and plastic goods; wells, and other types of container. This classification had very coarse categories and classified some of the containers according to its use (e.g., flower pots) and others according to its size and/or shape. The new classification (with vernacular terms for the containers) included: tires; large tanks or barrels; trays; drums; buckets; bottles; ceramic jugs ("cántaros"); small sailcloth-made swimming-pools and sailcloth; pots, and other types (in a frequency lower than 1%, including construction materials, discarded vehicle parts, discarded home appliances, toilettes, boots, toys, etc.). The material of large tanks also was registered (fibrocement, metal, or plastic).

For each container, all variables found associated with infestation and/or productivity in a previous study (Garelli et al. 2009) were registered: location within the lot (inside or outside the house), sun exposure (considered low if any structure such as a ceiling or a tree overshadowed the container, or high otherwise), lid status (only for large tanks, classified as fully lidded or not), water type (rain; piped; pump, or rain and piped water), and water state (considered clean if it contained transparent water, or polluted otherwise).

Householders were also asked for the main purpose of the water held in each container (water use) and the frequency of water addition (coded as a factor with four levels: every one or 2 d; every 3–5 d; every 5 d or more; or rain filled). Water use categories included animal drinking, bathing, human drinking, all-purpose, flower pot, nothing, other (breeding fish,

construction purposes, cooking, ice-making), storage, watering, and washing. Water addition frequency was only registered for large tanks because for other container types, householders were generally uncertain.

After inspection every container found was turned upside down, destroyed, or treated with 1% temephos (Abate, BASF, Ludwigshafen, Germany) at 1 mg/L applied as sand granules using spoons. Animal drinking pots and natural containers were not treated with temephos because it is toxic to some of the local domestic animals. No further vector control was applied between surveys.

Statistical Modeling. The modeling philosophy used aimed at knowledge-based model postulation and selection based on Akaike's Information Criterion (AIC) following Burnham and Anderson (2002). The variables included in the candidate models were a priori selected to reflect different putative processes and factors affecting the occurrence and abundance of *Ae. aegypti* (e.g., the larvae and pupae found during the surveys). Different models reflected different combinations of hypothesized processes and factors. Whether the hypotheses were supported by the data or not was considered at this stage as a problem of selection between different candidate models. Models with lower AIC score are considered better supported relative to the rest of candidate models. AIC values are on a relative scale, and therefore only AIC differences (Δ AIC) between models are informative; the bigger the Δ AIC, the higher the weight of evidence in favor of one of the models. Burnham and Anderson (2002) and several other authors consider that, as a rule of thumb, AIC changes higher than two are sufficiently high to rule out model selection uncertainty.

The construction and selection process was based on models obtained previously ('basic models') (Garelli et al. 2009). The order in which the AIC-based selection process is performed is not relevant for the outcome of the procedure (Burnham and Anderson 2002). Infestation (presence or absence of *Ae. aegypti* larvae and/or pupae among all containers inspected) was studied through multiple logistic regressions, and productivity (the number of pupae in infested containers) through multiple negative binomial regressions. In the basic models, the variables associated with infestation were survey period, container type, location, sun exposure, lid status, water type, and water state; with productivity, the variables were survey period, container type, lid status, water type, and water state.

The additional variables used to construct the candidate models were selected to reflect water use practices, vector-related context factors, history of chemical control, and climate. In addition, the new container classification scheme was compared with the old classification scheme used in Clorinda between 2003 and 2008. As some of the variables and processes identified only affected a subset of the containers found, some of the candidate models were constructed hierarchically (Gelman and Hill 2007). For example, lid status (a variable observed to vary

almost exclusively among large tanks) was modeled hierarchically.

Whether water use practices influence infestation and productivity was assessed by adding the variables water use and water addition frequency to the basic models. The latter variable was modeled hierarchically affecting only large tanks (water use practices was only registered for large tanks). As water state and water type apparently reflected water use practices (Garelli et al. 2009), the candidate models were constructed by combining the presence or absence of these three variables. Water type, also hypothesized to reflect water use practices, was also considered hierarchically as only affecting containers other than large tanks.

Several variables were constructed to model the putative impact of the vector-related context in which containers were found: number of pupae found at the block, number of pupae per container at the block, number of infested containers at the block, container index at block-level, and survey period. The first four variables were calculated after excluding the information obtained from the sampling unit (container). Survey period was also considered a vector-related context factor because the two spring surveys had large differences with regards to infestation and productivity despite being conducted almost during the same months of consecutive years. A comparison of the first spring survey with historical records suggested that infestation levels peaked later than usual during that season.

Two variables were constructed to represent chemical control during the previous survey: the amount of temephos used at block level, and the amount of temephos used at the lot of each container during the last treatment round. Data collected in a survey carried out in the 2007 fall was used to calculate these variables for the spring 2007 survey. Both variables were also considered in quadratic form to account for nonlinear effects.

Meteorological data were collected from 1 January 2003 to 31 December 2007 by a local weather station run by Cooperativa de Provisión de Obras y Servicios Públicos Clorinda Limitada. Four sets of candidate variables were selected: mean daily temperature, minimum daily temperature, maximum daily temperature, and daily rainfall. Each of these variables was considered with several time lags (1, 3, 5, 7, 9 d). This range of lags was selected based on development times for immature stages. A distance of 2 d per lag was selected to exclude highly correlated variables. All variables were considered linearly and adding a quadratic term to account for putative nonlinear effects. The selection procedure was performed by computing all possible models with the restriction of not adding correlated variables in the same candidate model (e.g., variables with the same lag) for temperature and rain variables, respectively. Overall, 17,074 models were tested for infestation and productivity, respectively.

After obtaining the best models, the variables selected were categorized according to the type of processes or factor they reflect. The categories obtained

Table 1. Inspected containers per block, house index, container index, Breteau index, and pupae per container

Survey	Inspected containers per block	House index	Container index	Breteau index	Pupae per inspected container
Spring 2008	12.3	9.7	21.9	11.3	3.94
Fall 2008	16.9	14.3	25.1	20.9	2.81
Spring 2007	15.64	6.3	8.2	7.8	0.96

Clorinda 2007–2008.

were: water use practices; vector-related context factors; climate, and container characteristics affecting mosquito breeding (including container description, location, sun exposure, and lid status). The relative importance of each of these subsets was estimated by computing the Δ AIC between the best models and the best models without the variables included in each category.

The overall quality of the best infestation model was assessed by estimating the area under the receiver operating characteristic (ROC) curve, sensitivity and specificity using as cutoff values the observed prevalence of infestation in containers for the entire data set (18.3%). In the case of the best productivity model, the quality of the fit was assessed by computing the correlation coefficient between model prediction and observed number of pupae per infested container. All calculations were performed using R 2.10.1 (R Development Core Team 2009).

Risk Maps. Based on the best models obtained, risk maps were constructed to identify the areas of higher entomological risk for improved vector surveillance and control. Two types of risk maps were constructed: 1) using all the variables in the models, and 2) using only the variables that change more slowly over time, for example, including water use practices and container characteristics affecting mosquito breeding and excluding vector-related context factors and climate-related variables. The actual distribution of infestation and productivity recorded was also mapped to compare model outcomes with the actual data recorded. As the geographical information was obtained at lot level, each map was constructed by summing the information for each container grouped by lot. The maps reported were obtained by averaging the outcomes for the three surveys. Temporal heterogeneity between surveys was not included in this analysis because of extension constraints. All maps reported were smoothed using a quartic kernel density function (Silverman 1986) with a bandwidth of 50 m. Therefore, two sets of three maps were constructed; one of predicted/observed number of infested containers per meter square and the other of predicted/observed number of pupae per meters squared. All maps were constructed using ArcGIS 9.1 (Environmental Systems Research Institute [ESRI], Inc., Redlands, CA) and its Spatial Analyst extension.

Results

In total, 5,431 water-holding containers were inspected during the three surveys, and 12,369 *Ae. aegypti* pupae collected from 963 positive containers

(Table 1). Data were obtained from 93, 91, and 78% of the lots in the three surveys, respectively. Large tanks were the most productive container type, harboring 71% of the pupae found.

Infestation Models. The best model represented a vast improvement over the basic model (Δ AIC = 431.0) and included variables reflecting water use practices, vector-related context factors, climate, and the new container classification scheme (Table 2; Table S1, available online only).

Adding the variable water use improved the basic model (Δ AIC = 72.2). The best model at this stage (Δ AIC = 120.2 compared with the basic model) also included water addition frequency modeled hierarchically only for large tanks and water type modeled hierarchically for the rest of the containers (Δ AIC = 2.3 compared with the second best model). In addition, the model improved slightly when the variable water state was removed (Δ AIC = 0.6). Containers used for animal drinking were less likely to be infested than the rest, and unused containers had the higher infestation levels. Containers with water addition frequency <5 d had lower infestation levels whereas those with 5 d or more did not differ from those filled with rainwater.

Two variables that reflected vector-context effects were selected in the best model (Δ AIC = 241.19 compared with the basic model); number of pupae per container at block level had a positive effect, including a negative quadratic term, and survey period, which showed the lowest infestation levels in the spring 2007 survey. Variables constructed to reflect history of chemical control did not improve the model.

Several climatic variables were selected and improved the model (Δ AIC = 48.1 compared with the basic model); mean daily temperature with a lag of 3 d (both linear and quadratic terms), maximum daily temperature with a lag of 1 d, daily rainfall with a lag of 6 d, and daily rainfall (both linear and quadratic terms) with a lag of 3 d.

The new container classification scheme improved the basic model despite including more parameters (Δ AIC = 13.5). The model was also improved by modeling container material hierarchically affecting only large tanks (Δ AIC = 9.8). The container types more likely to be infested were buckets, bottles, other types, and pots. Lidded large tanks and plastic-built tanks had lower infestation levels.

Variables reflecting water use practices were the most important among the variable categories constructed because removing them from the best model produced a Δ AIC = -476.7 compared with the best

Table 2. Parameter estimates of the variables in the best model of *Ae. aegypti* infestation

Explanatory variables	Odds ratio	P	N (%)	CI	Explanatory variables	Odds ratio	P	N (%)	CI
Water use practices variables					Container characteristics variables				
Water addition frequency					Container type				
Large tanks					Tires				
Every >5 d	1.00		19	2.7	Buckets	1.93	0.01	4	16.9
Rain	0.94	0.68	28	4.6	Bottles	1.97	0.04	3	28.8
Every 1 or 2 d	0.34	0.00	41	1.8	Ceramic pots	0.02	0.00	4	10.7
Every 3–5 d	0.41	0.00	12	3.2	Large tanks	1.36	0.12	65	16.1
Rest of containers					Container material				
Not rain filled	1.00		45	6.2	Fibrocement	1.00		58	18.5
Rain filled	0.17	0.00	55	33	Metal	0.78	0.38	4	17
Water use					Plastic				
Animal drinking	1.00		7	5.4	Lid status	0.72	0.01	38	11.5
Bathing	2.97	0.00	5	14	Unlidded	1.00		64	20.3
All purpose	1.99	0.03	15	11	Lidded	0.99	0.00	36	9.1
Flower pot	2.86	0.01	3	15	Other	2.10	0.00	4	36.3
Human drinking	0.88	0.01	18	13	Pots	2.66	0.00	8	24.3
Nothing	2.75	0.00	22	32	Sailcloth	1.07	0.84	3	11
Other	3.35	0.00	2	15	Trays	0.76	0.60	1	7.9
Storage	2.39	0.02	4	13	Location				
Washing	2.69	0.00	17	16	Inside	1.00		11	7.9
Watering	3.35	0.00	6	24	Outside	2.03	0.00	89	18.6
Vector-related context factors					Sun exposure				
No. pupae	1.02	0.00			High	1.00		40	15.5
(No. pupae) ²	1.00	0.00			Low	1.55	0.00	60	18.8
Survey period					Climatic variables (lag, in days)				
Fall 2008	1.00		39	24	Daily rainfall (3)	1.00	0.29		
Spring 2008	1.21	0.23	22	22	(Daily rainfall (3)) ²	1.00	0.11		
Spring 2007	0.33	0.00	40	9.3	Daily rainfall (6)	1.01	0.00		
					Mean daily temp (3)	1.20	0.02		
					(Mean daily temp (3)) ²	1.00	0.00		
					Max daily temp (1)	0.96	0.04		

Clorinda 2007–2008.

model. They were followed closely by vector-related context factors ($\Delta AIC = -430.5$). Container characteristics were less important ($\Delta AIC = -108.0$) and climatic variables were much less important ($\Delta AIC = -19.5$). Overall model quality was good; the area under the ROC curve was 0.81; sensitivity, 73%, and specificity 75%, with 75% of observations correctly classified.

Based on the maps constructed, some areas of the neighborhood show greater risk of infestation, especially the center and also some blocks of the southern sector (Fig. 1). A comparison of the maps shows very good correspondence between observed data and model predictions, especially those derived from the full model.

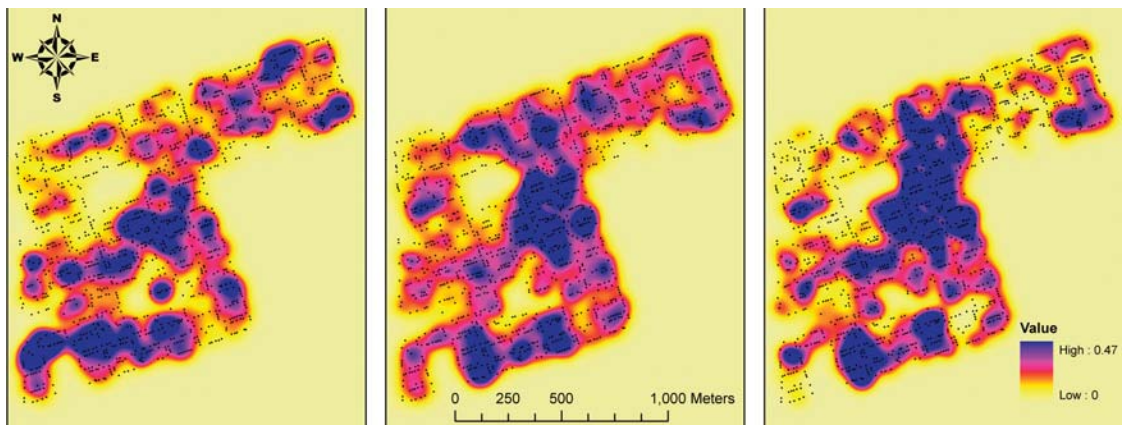


Fig. 1. Maps of infestation, predicted/observed number of infested containers per meters square. Dots represent houses. The maps show the average of the outcomes over the three surveys. (A) Risk map of model including only water use practices and container characteristics affecting mosquito breeding. (B) Risk map of full model. (C) Observed data, Clorinda 2007–2008. (Online figure in color.)

Table 3. Parameter estimates of the variables in the best model of *Ae. aegypti* productivity

Explanatory variables	Incidence rate ratio	P	Pupae per container
Water use practices variables			
Water addition frequency			
Large tanks			
Every >5 d	1.00		2.70
Rain	1.43	0.04	4.60
Every 1 or 2 d	1.16	0.50	1.80
Every 3-5 d	0.68	0.17	3.20
Vector-related context factor			
No. pupae	1.00	0.00	
Survey period			
Fall 2008	1.00		2.80
Spring 2008	2.18	0.00	3.90
Spring 2007	0.66	0.01	1.00
Container characteristics variables			
Container type			
Tires	1.00		1.30
Buckets	0.78	0.38	2.80
Bottles	0.33	0.00	1.30
Ceramic pots	-1.54	0.21	0.20
Large tanks	1.30	0.30	3.00
Lid status			
Unlidded	1.00		4.00
Lidded	1.00	0.04	1.30
Other	0.83	0.49	3.40
Pots	0.62	0.07	1.20
Sailcloth	1.63	0.40	1.20
Climatic variables (lag, in days)			
Min daily temp (9)	1.15	0.14	
(Min daily temp (9)) ²	1.00	0.00	
Max daily Temp (5)	-0.04	0.96	
Mean daily temp (3)	1.75	0.56	
(Mean daily temp (3)) ²	-0.01	0.99	
Daily rainfall (1)	-0.01	0.99	
(Daily rainfall (1)) ²	1.00	0.00	

Clorinda 2007-2008.

Productivity Models. The best model ($\Delta AIC = 53.4$ with respect to the basic model) included variables reflecting water use practices, vector-related context effects, climate, and the new container classification scheme (Table 3; Table S2, available online only).

Water addition frequency modeled hierarchically for large tanks was included in the best model whereas

water type was excluded ($\Delta AIC = 11.5$ compared with the basic model); rain-filled containers harbored more pupae than the rest.

Two variables reflecting vector-related context effects were selected in the best model: number of pupae per container in the block ($\Delta AIC = 4.52$ compared with the basic model) had a positive effect and survey period ($\Delta AIC = 15.2$) had lower productivity levels in the spring 2007 survey. Variables constructed to reflect history of chemical control did not improve the model.

The climatic variables selected ($\Delta AIC = 17.4$ compared with the basic model) included minimum temperature with a lag of 9 d; maximum daily temperature with a lag of 5 d; mean daily temperature with a lag of 3 d (including both linear and quadratic terms), and daily rainfall with a lag of 1 d (including both linear and quadratic terms).

The new container classification scheme was selected in the best model ($\Delta AIC = 3.5$), with large tanks being the most productive container type. Container material was not selected in the best model.

The vector-related context factors were the most important relative to the rest of the variable types ($\Delta AIC = -31.1$), followed by climatic variables ($\Delta AIC = -27.5$), container characteristics ($\Delta AIC = -9.4$) and water use ($\Delta AIC = -2.8$). Model fit in this case was not as good as with infestation models; the correlation between model predictions and observations was significant and positive though small in magnitude ($r = 0.17$; $P < 0.001$).

The correspondence between maps based on model predictions and observed data in this case was weaker (Fig. 2). However, the center of the neighborhood was also identified as having the highest infestation risk based on the full model and observed data.

Discussion

The models obtained showed strong associations between explanatory and response variables (espe-

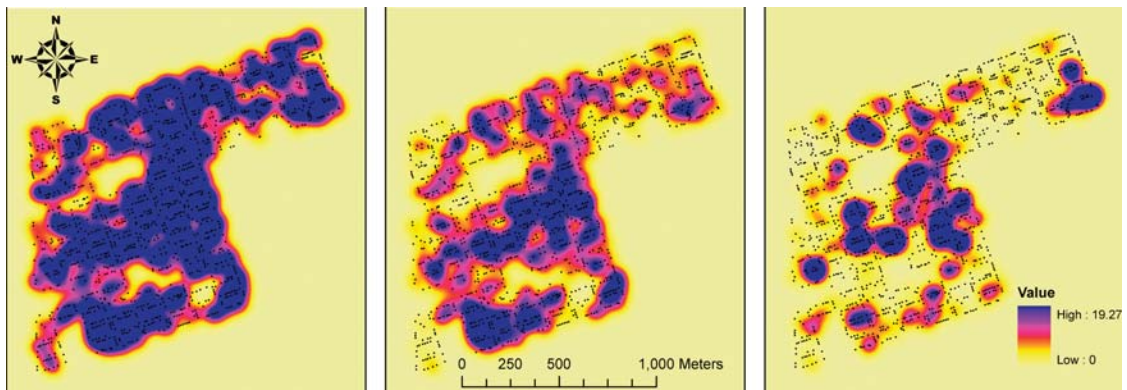


Fig. 2. Maps of productivity in infested containers, predicted/observed number of pupae per meters square. Dots represent houses. The maps show the average of the outcomes over the three surveys. (A) Risk map of model including only water use practices and container characteristics affecting mosquito breeding. (B) Risk map of full model. (C) Observed data, Clorinda 2007-2008. (Online figure in color.)

cially the infestation model), allowing evidence-supported inference of processes affecting *Ae. aegypti* mosquitoes. Variables reflecting water use practices had the highest impact on the AIC score of the infestation model and were the most important explanatory variables. The maps constructed displayed heterogeneous distributions of infestation and productivity across the study area. In addition, the models obtained were a vast improvement over the basic models of our earlier study, allowing for the inference of several putative processes not clearly identified previously.

Almost every putative process and factor considered, including water use practices, vector-related context, characteristics of containers affecting mosquito breeding, and climate, were selected in the best models obtained both for infestation and productivity. The associations found confirm that processes traditionally studied by different disciplines, such as ecology, meteorology, and social sciences, play an important role in determining mosquito occurrence and abundance.

Containers under higher water addition frequency regimes were less infested than those either rain-filled or manually filled every five days or more. The fact that water addition frequency was selected in the best model of infestation, water state was excluded and water type was included only for containers where water addition frequency was not measured, suggests that the latter variables were associated with infestation because they are confounded with water use practices, as hypothesized elsewhere (Garelli et al. 2011). Water use practices in Clorinda are affected by an intermittent piped water service. Water turnover frequency has also been shown to affect temephos residuality strongly; containers with higher water turnover had shorter residual effects (Garelli et al. 2011). These combined effects of water turnover probably have a complex and nonlinear impact on mosquito abundance in the context of repeated chemical treatment of containers. Differences were also found according to water use or function; containers used for animal drinking (generally pots) were less infested, and unused containers were the most infested. These findings suggest that water use practices have a great impact on *Ae. aegypti* dynamics and, therefore, dengue transmission in Clorinda. The importance of water use practices affecting mosquito dynamics has been pointed out in studies during at least the last 40 yr (Pichón et al. 1969, Chadee 1984, Caprara et al. 2009), but still is a subject that should be more and better studied. A recent study performed in Colombia also reported similar results concerning the importance of water use practices and the frequency of water renewal for *Ae. aegypti* production (Padmanabha et al. 2010). These findings also support the recommendation of using water use in containers as an informative characteristic relevant for container classification (Koenraadt et al. 2007).

Vector-related context factors also were associated with infestation and productivity. Part of their importance was probably because of the large differences between the two spring surveys; the first one had

much lower infestation levels. A particularly cold, dry winter occurred in 2007 and the citywide infestation records show that the highest levels of infestation occurred later than usual that year. Considering also the spatial heterogeneity revealed by the maps, further analyses aimed at describing and explaining the spatio-temporal patterns of infestation (e.g., Getis et al. 2003) and habitat quality are warranted but exceed the scope and extension of this paper.

Chemical control variables did not exert effects on infestation or productivity, as expected from the short duration of temephos residual effects in local field containers (Garelli et al. 2009), and that surveys and treatment rounds occurred ≈ 6 mo apart. This does not mean that temephos treatment was ineffective because it reduced infestation levels significantly (Gürtler et al. 2009). Rather, this suggests that the system was resilient, with reinfestation events rising infestation levels between treatment rounds.

Temperature and rain were also important variables, especially in the case of productivity in infested containers. The lags selected were longer for productivity than for infestation models. This is consistent with the fact that productivity was measured only in terms of number of pupae. Lags considered in this study were only up to 9 d because they were selected to represent processes directly affecting the larvae or pupae actually collected. Longer lags up to 21 d were also tested but they were not selected in the best models (data not shown). Lags in the scale of months were not considered because they were not hypothesized to affect our observations; their effects are carried over through generations of mosquitoes. Studying the effects of the climate in more detail may be of great importance, especially considering the estimated impact of climate change on human health and its putative effects on dengue incidence (Patz et al. 1998, 2005; Johansson et al. 2009).

The new container classification scheme constructed based on field personnel annotations was an improvement over the previous scheme. Other container classification schemes, such as the SUM-method (Koenraadt et al. 2007), may allow easier comparisons with other studies and could be a further improvement.

The most important variable types in the models of infestation and productivity were different. Variables constructed to reflect water use practices were much less important in productivity than in infestation models, whereas climatic variables were relatively much more important in productivity models. Perhaps water use practices only determine container suitability (a magnitude that might change more slowly over time) and the more contingent and variable climate variables have a higher impact on productivity, which is probably more affected by cohort effects, stochastic effects and more rapid change over time. The association between the variables included in the best model and infestation was very large and a good fit was obtained ($\Delta AIC = 873.1$ compared with a null model, area under ROC curve 0.81) though in the case of productivity it was weaker ($\Delta AIC = 115.4$; Pearson's

$r = 0.17$; $P < 0.001$). This can be verified qualitatively by comparing the maps constructed. Therefore, our models are better supported to explain and/or predict container infestation than productivity. This may occur because productivity is very likely to be influenced by contingent effects, such as demographic stochasticity, or more fine-grained processes than those that our variables can reflect.

The models reflected many processes but not all putative processes were considered, for example, food availability (Barrera et al. 2006) or water temperature in containers (Tun-Lin et al. 2000). Another limitation of this study is that no direct estimations of adult mosquito abundance were made, especially in view of the importance of vector-related context variables. In addition, the procedure used to identify the most important types of variables may not accurately rank the actual importance of the putative processes because it is highly dependent on how well these are reflected by the variables constructed.

Many of the explanatory variables postulated as causes of infestation and productivity also have causes that are not fully understood. For example, our results show that water use practices largely affect mosquito occurrence. However, the determinants of water use practices, known to be both cultural and structural (determined by access to water sources), are not well understood.

Understanding the determinants of mosquito occurrence or abundance is an important goal for the epidemiology and control of dengue. Statistical modeling is frequently used for this purpose, however it is insufficient (Freedman 1999). Perhaps the ultimate test of a causal hypothesis can be sought when implementing control strategies that would impact the putative causal pathway. To improve or create new control strategies, it is especially important to study processes that may be manipulated by control strategies. In our study, both water use practices and physical characteristics of containers may be modified by putative interventions. In contrast, climatic factors and monitoring measures of infestation (such as adult density) may serve as indicators for early warning systems, but their development needs a different kind of modeling approach (Gubler 2002, Hopp and Foley 2003).

Our results suggest that future interventions should aim at targeting large tanks (the most productive container type) and consider key elements such as lid status, water addition frequency and water use. As water use practices seem to have such strong impact on both infestation and the residuality of temephos, a novel intervention aiming at community participation for healthier water use practices is recommended for mosquito control and dengue prevention in Clorinda and similar cities in the region.

Acknowledgments

For support and advice we thank Evangelina Aldama and the Clorinda team: Marcos Aguirre, Gustavo Caballero, Myriam Cardoso, Oscar Gómez, Gladys López, Alicia Lovera,

Zunilda Lovera, María Lugo, Marisabel Maldonado, Carmen Ovando, Alba Pattenden, Alicia Roa, Gloria Vielma, Susana Zárate, and Cynthia Zárate. For helpful comments and discussion, Juan Manuel Gurevitz. R.E.G. is member of CONICET Researcher's Career, Argentina.

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Received 25 July 2011; accepted 20 October 2011.