

Environmental Predictors of the Occurrence of Ground Water Mosquito Immatures in the Paraná Lower Delta, Argentina

Author(s) :M. V. Cardo, D. Vezzani and A. E. Carbajo

Source: Journal of Medical Entomology, 48(5):991-998. 2011.

Published By: Entomological Society of America

DOI: 10.1603/ME11046

URL: <http://www.bioone.org/doi/full/10.1603/ME11046>

BioOne (www.bioone.org) is a nonprofit, online aggregation of core research in the biological, ecological, and environmental sciences. BioOne provides a sustainable online platform for over 170 journals and books published by nonprofit societies, associations, museums, institutions, and presses.

Your use of this PDF, the BioOne Web site, and all posted and associated content indicates your acceptance of BioOne's Terms of Use, available at www.bioone.org/page/terms_of_use.

Usage of BioOne content is strictly limited to personal, educational, and non-commercial use. Commercial inquiries or rights and permissions requests should be directed to the individual publisher as copyright holder.

Environmental Predictors of the Occurrence of Ground Water Mosquito Immatures in the Paraná Lower Delta, Argentina

M. V. CARDO, D. VEZZANI, AND A. E. CARBAJO¹

Unidad de Ecología de Reservorios y Vectores de Parásitos, Departamento de Ecología, Genética y Evolución, Facultad de Ciencias Exactas y Naturales, Universidad de Buenos Aires, Ciudad Universitaria, Pabellón 2, 4° piso, (C1428EHA) Buenos Aires, Argentina

J. Med. Entomol. 48(5): 991–998 (2011); DOI: <http://dx.doi.org/10.1603/ME11046>

ABSTRACT Characterizing mosquito larval habitats is essential for understanding the complex interactions between immatures and the biotic and abiotic components of their environment. Using generalized linear mixed models, we studied the environmental predictors of the presence of three ubiquitous mosquito species breeding in ground water habitats in the Paraná Lower Delta, Argentina. During a year-round survey, 34.1% of the 419 ground water habitats inspected were positive for either *Culex dolosus* s.l. (Lynch Arribáizaga 1891), *Aedes crinifer* (Theobald 1903), or *Culex intricatus* Brèthes 1916. Univariate analysis showed that the former two occurred throughout the year, whereas the latter during the summer and fall. *Ae. crinifer* and *Cx. intricatus* were more frequently collected in secondary forests, whereas *Cx. dolosus* s.l. was homogeneously distributed among land uses. Best generalized linear mixed models included the sampling period and landscape variables in different combinations for each species. Spatial dependence of the data was evident for *Cx. dolosus* s.l. and *Ae. crinifer*. Our results showed that the most widespread species presented different spatio-temporal distribution patterns, related with land use, anthropic intervention, and seasonality, highlighting the complexity of the wetland under study. This methodological approach could aid in the selection of priority areas for vector control and disease risk management.

KEY WORDS *Culex dolosus* s.l., *Aedes crinifer*, *Culex intricatus*, spatio-temporal distribution, wetland

Mosquitoes are insects of major public health concern for their role as vectors of pathogens to humans and animals (WHO 2002). They also constitute a well-known nuisance and represent an ecologically important component of aquatic ecosystems (Beketov et al. 2010). In this context, the characterization of mosquito larval habitats is essential for understanding the complex interactions between immatures and the biotic and abiotic components of their environment (Laird 1988). Recent studies in different regions of the world have invested effort in identifying which environmental factors are the most important drivers of the occurrence of certain key mosquito species (e.g., Leisnham et al. 2005, Muturi et al. 2008, de Little et al. 2009), but in South American wetlands this remains as an unexplored issue.

The Delta of the Paraná River is one of the main wetland systems in South America, providing different kinds of resources, such as fishing, forestry, transport, and tourism, as well as an important source of freshwater (Kandus et al. 2006). As many other delta areas (see Dale and Knight 2008), it has been traditionally associated with mosquito annoyance. Current knowledge of mosquito fauna breeding in the Lower Delta

is restricted to the pioneering works of Prosen et al. (1960) and García and Casal (1965). In a recent survey, 23 culicid species have been recorded in ground water habitats; *Culex dolosus* s.l. (Lynch Arribáizaga 1891), *Aedes crinifer* (Theobald 1903), and *Culex intricatus* Brèthes 1916 were the most frequently collected species, representing a high percentage of the samples (Cardo et al. 2011). Available literature on these species in the region is mainly restricted to urban areas and their surroundings (Balseiro 1981, 1989; Ronderos et al. 1992; Maciá et al. 1995, 1996; Maciá 1997).

Species distribution models provide detailed predictions of distributions by relating presence or abundance of species to environmental predictors (Elith et al. 2006). In particular, generalized linear models (GLM) (McCullagh and Nelder 1989) are a useful tool for the study of these associations because they develop simple equations relating species presence and absence to environmental variables. In these models, a combination of predictors, the linear predictor (LP), is related to the mean of the response variable through a link function. Using such link function allows the following: 1) transformation to linearity, and 2) the predictions to be maintained within the range of coherent values for the response variable (Guisan and

¹ Corresponding author, e-mail: manimal@ege.fcen.uba.ar.

Zimmermann 2000). Because these models use maximum-likelihood estimators, the fit is measured by the reduction in deviance instead of variance (typical of least-squares estimation). Generalized linear mixed models (GLMM), a particular type of mixed model, are an extension of GLM in which the LP contains random effects in addition to the usual fixed effects. GLMM provide a powerful method to treat data with errors that do not conform to a normal distribution and, at the same time, control for correlations between measures that arise from grouped observations (Patterson and Lello 2003). This correlation may arise because of repeated measures in time, or because of spatially related data. As, in fact, GLM are a special case of GLMM with no random factor, henceforth we will refer to both as GLMM.

In an attempt to identify key factors related with the distribution of three ubiquitous mosquito species breeding in the main wetland of Argentina, we tested by GLMM the association among seasonality, environmental conditions, and the presence of immatures of *Cx. dolosus s.l.*, *Ae. crinifer*, and *Cx. intricatus* in ground water habitats of the Paraná Lower Delta.

Materials and Methods

Study Area and Sampling Design. The Lower Delta of the Paraná River is a wetland macromosaic that extends through the terminal portion of the del Plata Basin (Kandus et al. 2006). It covers $\approx 2,700$ km² and is prograding toward the estuary through the continuous development of new bars and islands. The region is temperate with mean annual temperature of 16.7°C (minimum 6°C, maximum 30°C) and accumulated annual rainfall values of 1,073 mm (Kandus and Malvárez 2004). The hydrologic regime is the product of the combined effects of the Paraná River flow and wind patterns affecting the estuary.

The region presents four main land uses, namely peridomestic areas, Salicaceae plantations, secondary forests, and *Scirpus giganteus* patches locally known as "pajonal." The former two are highly modified environments, whereas the latter two are relatively little impacted. Each land use has a typical location on the islands, determined primarily by water accumulation: peridomestic areas in levees, pajonal in the inner lowlands, and plantations and secondary forests in the areas in between.

Six sampling campaigns were conducted from November 2008 through September 2009, at 2-mo intervals. Site selection was based on a preliminary assessment of 24 sites in an attempt to capture landscape heterogeneity and provide an adequate number of replicates of the main land uses, detected by satellite imagery and ground proofing. Each campaign consisted of 12 consecutive field working days and involved 10 sampling sites situated in the municipalities of Tigre (S 34.4258°, W 58.5967°), San Fernando (S 34.4442°, W 58.5775°), and Campana (S 34.1769°, W 58.9208°). Sites were categorized according to the predominant land use (>50% coverage). Two distinctions were made to embrace environmental hetero-

Table 1. Number of sites, land use composition, and proportions for each of the five site types sampled in the Paraná Lower Delta

Site type	No. sites	Peridomestic areas (%)	Plantations (%)	Secondary forests (%)	Pajonal (%)
1	3	<50	0	>50	0
2	2	50	0	50	0
3	1	>50	0	<50	<50
4	2	<50	>50	<50	0
5	2	<50	>50	0	<50

geneity: when two land uses shared 50% coverage (i.e., no predominant land use was present) and when, given the same predominant land use, the site included pajonal or not (because pajonal was not predominant in any site). The resulting categories were as follows: type 1, majority of secondary forest; type 2, peridomestic and secondary forest in equal proportion; type 3, majority of peridomestic; type 4, majority of plantation with pajonal; type 5, majority of plantation without pajonal (Table 1). Each site was also characterized in terms of its size and distance to the main port in the area (measured in access time by commercial motorboat). A Geographic Information System (GIS) was built with a base map of streams and rivers using ArcView GIS 3.2 software. A land cover types map taken from Kandus et al. (2006) was registered to the database as well as Google Earth images of the sites. At each site, one to three walking transects were randomly distributed in each land use category to ensure that all classes were surveyed; the number of transects varied according to the area occupied by each land use within a site. Each transect was up to 50 m length and subdivided in 10-m-spaced 4 × 4-m quadrats. Geographic coordinates were recorded for each quadrat with a Global Positioning System (Garmin eTrex Legend HCx). A 100-m-radius buffer around each quadrat was built in the GIS to measure the area covered by each land use type.

In the field, any ground water habitat (GWH), as described by Service (1995), present in a quadrat was sampled for immatures of mosquitoes. This included a variety of habitats, such as pools, flooded ground, irrigation ditches, canals, ponds, and streams. Sampling techniques varied according to habitat characteristics in an attempt to maximize mosquito capture. Fine-mesh strainer or tray content was examined, immatures collected with the aid of a pipette, and water content reduced. Sampling effort was standardized according to the size of the water body in terms of minutes of active search for immatures (minimum = 1 min for 1 m²; maximum = 12 min for 16 m², the size of the quadrat). Third- and fourth-instar larvae were preserved in 70% ethanol and morphologically identified to species (Darsie 1985, Forattini 2002, Rossi et al. 2002). Pupae were reared, and the emerged females were also identified with the mentioned keys. Considering that larval specimens of *Cx. dolosus* and *Culex eduardoi* have been largely misidentified and that both taxa may belong to a species complex (Senise and Sallum 2008), we grouped all immatures collected and referred to them collectively as *Cx. dolosus s.l.*

Table 2. Variables included in GLMM for species presence

Variable name	Continuous/Factor (n° of levels)	Scale	Description	Source	Units
Sampling.period	Factor (6)	Global	Each of the six sampling campaigns	Fixed by sampling design	—
Site.type	Factor (5)	Site	Classification of each site in terms of % of coverage of 4 main land uses (see Table 1)	Field data	—
Area	Continuous	Site	Site area	Google Earth	m ²
Time.port	Continuous	Site	Time in motorboat to main port	Field data	Minutes
Forest.buff	Continuous	Quadrat	Secondary forest coverage in a 100-m-radius buffer around each quadrat	Kandus et al. 2006	m ²
Herb.buff	Continuous	Quadrat	Herbaceous coverage in a 100-m-radius buffer around each quadrat	Kandus et al. 2006	m ²
Mosaic.buff	Continuous	Quadrat	Peridomestic coverage in a 100-m-radius buffer around each quadrat	Kandus et al. 2006	m ²
Paj.buff	Continuous	Quadrat	Pajonal coverage in a 100-m-radius buffer around each quadrat	Kandus et al. 2006	m ²
Plant.buff	Continuous	Quadrat	Plantations coverage in a 100-m-radius buffer around each quadrat	Kandus et al. 2006	m ²
Reed.buff	Continuous	Quadrat	Reed coverage in a 100-m-radius buffer around each quadrat	Kandus et al. 2006	m ²
Water.buff	Continuous	Quadrat	Open water area in a 100-m-radius buffer around each quadrat	Kandus et al. 2006	m ²

Data Analysis and Modeling. Joint occurrence between pairs of species was assessed with a χ^2 test by comparing observed and expected frequencies of co-occurrence under the null hypothesis of species independence (Sokal and Rohlf 1969). For descriptive purposes, breeding site index (BSI) for species *i* was defined as the proportion of GWH in which species *i* was present among all GWH surveyed. It was examined among sampling periods and land uses to detect preliminary trends.

Given the fact that mosquito abundance estimations are highly variable and dependent on many factors unrelated to true mosquito density (Rochlin et al. 2009), the analysis of occurrence may be statistically easier and conceptually more appropriate for the scale of the current study. Therefore, the occurrence of each of the three culicids studied was modeled as a function of explanatory variables (x_1, x_2, \dots), which were related to the response variable through a LP. $LP = a + bx_1 + cx_2 \dots$, where a, b, c, \dots are parameters to be estimated. Presence/Absence of each species per quadrat was used as the response variable. We assumed a binomial distribution of errors and applied the logistic function as a link between the response variable and the LP. This link constrains the predicted values to a continuous variable between 0 and 1. The candidate model sets included all possible combinations of the explanatory variables (Table 2) and all two-way interactions between each continuous variable and each factor. We performed a manual forward stepwise procedure that involved testing the models obtained not only from the first most significant variable at each step, but also the alternative models that were obtained when the second or the third most significant variables were included. This branching procedure could eventually produce a set of different models, but in most instances it converged into a single model (Donazar et al. 1993). Term additions were evaluated by Akaike's information criterion (AIC, Akaike 1974): the model that yielded the lowest AIC was selected from all possible models (Zuur et al.

2009). Models with $\Delta AIC \leq 2$ were considered equivalent. Each term addition was further tested with a deviance change test (χ^2 test on the deviance change with df equal to the change in df between both models) using $\alpha = 0.01$ for retention because of the large number of variables considered. To simplify the models, the levels in a factor that were not significantly different were merged together (Nicholls 1989). This procedure was stopped when the merging implied a significant decrease in total explained deviance (χ^2 test for the change in deviance with 1 df). To deal with collinearity between continuous explanatory variables, we computed a pairwise Pearson correlation coefficient; when it surpassed 0.45 the variable responsible for the greater change in deviance was retained, whereas the other was excluded from further analysis. This is another advantage of the manual forward procedure, which allows the exclusion of highly correlated variables in each step of the regression. Given that best models output is a probability, we first considered >0.5 as presence prediction and ≤ 0.5 as absence. Afterward, receiver operating characteristics (ROC) curves (DeLeo 1993) were applied to estimate the cutoff point (from now on referred to as Rcp) that maximized the product between specificity (the proportion of correctly classified absences) and sensibility (the equivalent for presences). To evaluate the final models classification effectiveness, we applied a 10-fold cross-validation using the Kappa index (K) to assess improvement of classification of the model over chance (Fielding and Bell 1997). The Kappa index overcomes the problem of unequal number of presences and absences (Titus and Mosher 1984). We reported mean K values of the 10 cross-validations along with their corresponding standard deviations (SD) and followed Landis and Koch (1977) ranges of agreement: poor $K < 0.4$; good $0.4 < K < 0.75$; excellent $K > 0.75$. We performed this both for cutoff 0.5, labeled K(0.5) and SD(0.5), respectively, and for Rcp, K(Rcp) and SD(Rcp). The area under the ROC curve was also used as a measure of the accuracy of the

Table 3. Breeding site index (proportion of ground water habitats with species *i* present) for each species by sampling period

Sampling period	<i>Aedes crinifer</i>	<i>Culex dolosus s.l.</i>	<i>Culex intricatus</i>
Nov. 08	0.10	0.10	0.01
Jan. 09	0.03	0.03	0.32
Mar. 09	0.13	0.15	0.24
May 09	0.10	0.30	0.21
July 09	0.07	0.20	0.00
Sept. 09	0.28	0.20	0.01

model (the closer to 1, the better, Hangle and McNeil 1982).

To account for potential spatial dependence among samples taken within a given land use or transect, two different groupings were tested as a random factor in GLMM. These groups were each land use in each site ($i = 10$) during each sampling period ($j = 6$) (use_{ij}, 103 groups) and each transect thrown in the field (tra_{ij}, 158 groups). This was run both before and after model simplification. All analyses were performed in R 2.10.1 (R Development Core Team 2009) with boot and lme4 packages. Finally, to check for remnant spatial autocorrelation of the residuals of the models selected, we built variograms (Creese 1993) in S-plus 8.0, taking into account increasing distances up to half of the maximum distance between pairs.

Results

A total of 419 GWH was inspected, of which 34.1% (143) were positive for at least one of the three species under study: 16.7, 12.6, and 11.7% for *Cx. dolosus s.l.*, *Ae. crinifer*, and *Cx. intricatus*, respectively. There was a significant positive association ($P < 0.005$) between *Cx. dolosus s.l.* and *Ae. crinifer*, which were collected together in 3.8% (16/419) of samples, twice as many as expected solely by chance.

Preliminary univariate analysis showed that all species presented some seasonal trend, with BSI for each species different among sampling periods and each peaking in different times of the year (*Ae. crinifer* in September, *Cx. dolosus s.l.* in May, and *Cx. intricatus* from January to May; Table 3). Regarding land use, BSI of *Ae. crinifer* and *Cx. intricatus* in secondary forests were 0.18 and 0.21, respectively. Minimum values (0.03–0.04) were recorded in plantations for *Ae. crinifer* and in plantations, peridomestic, and pajonal for *Cx. intricatus*. *Cx. dolosus s.l.* presented highest values (0.19) in secondary forests and pajonal.

The GLMM model for each species included sampling period (i.e., seasonality) and variables corresponding to two different spatial scales as fixed factors. For *Cx. dolosus s.l.* and *Cx. intricatus* the interaction between sampling period and time.port was significant. Time.port was consistently negatively associated with *Cx. intricatus* occurrence, but positively or negatively related with *Cx. dolosus s.l.* presence depending on the sampling period. Also, site.type was relevant in modeling the presence of *Cx. intricatus* and *Ae. crinifer*. Merging resulted in different groups of sam-

Table 4. Simplified factors of site types and sampling periods for the reduced GLMM for each species

Site type	<i>Aedes crinifer</i>	<i>Culex dolosus s.l.</i>	<i>Culex intricatus</i>
1	A	VNI	A
2	B	VNI	A
3	A	VNI	B
4	B	VNI	B
5	C	VNI	A

Sampling period	<i>Aedes crinifer</i>	<i>Culex dolosus s.l.</i>	<i>Culex intricatus</i>
Nov. 08	A	A	A
Jan. 09	A	A	B
Mar. 09	A	B	C
May 09	A	C	C
July 09	B	D	D
Sept. 09	C	E	D

Equal letters within a column indicate sites or periods grouped together with $P < 0.05$. VNI, variable not included in the model.

pling periods and site types for each species (Table 4). Every species also had one or more variables emerging from the 100-m-radius buffer included in their models: *Cx. dolosus s.l.* and *Cx. intricatus* were negatively associated with paj.buff, and *Cx. intricatus* and *Ae. crinifer* were negatively related with herb.buff. The variables included in best models that presented significant correlations were time.port with water.buff and forest.buff (0.47 and -0.59 , respectively) and paj.buff with plant.buff (-0.57).

Each species responded differently to the addition of random factors. For *Ae. crinifer*, we chose the mixed model, including tra_{ij} (Table 5). For *Cx. dolosus s.l.*, the addition of either random factor, tra_{ij} or use_{ij}, resulted in a significant improvement of the model, equivalent in terms of AIC. We chose tra_{ij} for further analyses to simplify interpretation of the results because of similarities with *Ae. crinifer*. However, the addition of either random factor did not improve the model for *Cx. intricatus*.

In general, predictive accuracy estimators for the selected models were satisfactory (Table 6). The area under the ROC curve yielded high and comparable values; from 0.94 to 0.96. K(0.5) values ranged from 0.54 ± 0.17 to 0.69 ± 0.17 , indicating a good predictive power of the models according to the Landis and Koch criterion. The classification effectiveness of the models was not significantly improved when considering Rep correction, as was evidenced by the similarities between the K(Rep) and K(0.5) values and their low SDs.

Regarding potential remnant spatial autocorrelation, none of the variograms of the residuals for each species showed a spatial pattern of similarity between pairs of observations as a function of separation distance.

Discussion

The distribution of mosquitoes in any given location is influenced by variables operating across spatial and temporal scales (Alfonzo et al. 2005). Thus, effective mosquito control requires a detailed understanding of

Table 5. Model selection for each of the three culicids studied

		<i>Aedes crinifer</i>				<i>Culex dolosus s.l.</i>				<i>Culex intrincatus</i>			
		Complete model		Reduced model		Complete model		Reduced model		Complete model		Reduced model	
		AIC	df	AIC	df	AIC	df	AIC	df	AIC	df	AIC	df
GLM	Null	320.2	1	—	—	380.1	1	—	—	304.4	1	—	—
GLM	Best model	247.7	11	244.6	5	340.4	13	339.3	11	183.2	18	177.9 ^a	11
GLMM	Best model + use _{ij}	247.5	12	241.3	6	327.9	14	326.0	12	185.2	19	179.8	12
GLMM	Best model + tra _{ij}	242.1	12	234.9 ^a	6	327.3	14	325.8 ^a	12	185.2	19	177.6	12

Use_{ij} (103 groups), each land use of each site (i = 10) during each sampling period (j = 6); tra_{ij} (158 groups), each transect thrown in each site (i = 10) during each sampling period (j = 6).

^a Indicates the selected model for each species.

the temporal and spatial variation in immature presence among habitats (de Little et al. 2009). In the Paraná Lower Delta, the three most widespread species that breed in ground water habitats showed temporal variation in their presence. The sharp seasonality exhibited by *Cx. intrincatus* had been previously observed in urban and suburban areas in Buenos Aires Province (Maciá et al. 1996, Maciá 1997). Also, *Ae. crinifer* population outbreaks at the beginning of the spring had been previously reported near the study area (Maciá et al. 1995). Notwithstanding this, the interaction between sampling period and spatial variables showed that seasonality needs to be addressed site by site; that is, no global conclusion can be made without taking into account other factors included in the models. In this regard, GLMM are superior to traditional univariate analyses in the sense that they allow a holistic treatment of the data. Rather than focusing only on one environmental factor at a time, they provide a better opportunity to describe habitats in greater detail and at different spatial scales. In addition, GLMM accounted for grouped observations; the significance of transect as a random factor was expected as a result of sampling design, and was in fact verified for the three species studied (including *Cx. intrincatus*, for which it was equivalent to the selected model in terms of AIC).

The distribution of mosquitoes is largely dependent upon their flight range and the spatial distribution of larval breeding sites and preferred hosts, all of which are heterogeneous in space and time (Gimnig et al. 2005). In relation to this, the classification effectiveness of the models may reflect the extent to which each species is associated with environmental features, provided that the different environmental ef-

fects on distribution surpass baseline variability. In our study, the three species showed a moderate level of association with the environment. In each case, the fraction of variability not explained by the modeling did not present a spatial pattern, validating the regressions used. Certain clues on the biology of the studied species arise from the models: the merging of two or more levels in the factors (sampling periods and site types) may imply either that there is no biological difference among the levels that are merged together or that differences are subtle and thus undetected. However, all levels or groups of levels that did not merge are significantly different in biological terms. Regarding the inclusion of a random term in the models, it allows for the discrimination of spatial dependence. If we had not taken it into consideration, we would have been making a mistake by assuming independence among correlated observations or by considering significant spurious differences. The use of the random term makes *P* values to be higher than in the fixed model; therefore, it is more conservative in the selection of significant explanatory variables. On a biological ground, the random component gives an idea of dependence scale; a significant transect random term means that if a certain species is breeding in a given GWH, another GWH located in the same transect will have a higher probability of harboring the same species. In other words, two GWH located <50 m apart will have a probability higher than random of both harboring/not harboring a given species.

The Lower Delta has long been subjected to anthropogenic impacts consisting mainly of changes in land use and plant cover; about half of its extension is affected by human activities, mainly by Salicaceae plantations and recreation areas (Kandus et al. 1999).

Table 6. Classification effectiveness for the best model selected for each species (indicated with footnote a in Table 5; see Table 2 for variables definition)

Species	Fixed factors	Random factor	Rcp	AURC	K(0.5)	SD(0.5)	K(Rcp)	SD(Rcp)
<i>Aedes crinifer</i>	Site.type + sampling.period + herb.buff	tra _{ij}	0.11	0.96	0.69	0.17	0.58	0.07
<i>Culex dolosus s.l.</i>	Paj.buff + sampling.period × time.port	tra _{ij}	0.17	0.96	0.64	0.16	0.64	0.17
<i>Culex intrincatus</i>	Sampling.period × time.port + site.type + herb.buff + paj.buff	—	0.19	0.94	0.54	0.17	0.57	0.13

Rcp, adjusted cutoff point obtained by ROC curve; AURC, area under ROC curve; K(0.5), Kappa index value considering predicted values &γ; 0.5 as species presence; SD(0.5), its standard deviation; K(Rcp), Kappa index value considering predicted values &γ; Rcp as species presence; SD(Rcp), its standard deviation; tra_{ij} (158 groups), each transect thrown in each site (i = 10) during each sampling period (j = 6).

Significant positive or negative associations among species and variables indicating anthropic processes (e.g., forest fragmentation, agricultural land extension, eutrophication) have been reported elsewhere (Reiter and LaPointe 2007, Vanwambeke et al. 2007a, Johnson et al. 2008, Rochlin et al. 2008). In our study, the time to the nearest port emerged as an important variable for species modeling. This variable may be interpreted as inversely correlated to anthropic intervention, as urbanization decreases away from the mainland ports. Interestingly, its effect was unequal for *Cx. dolosus s.l.* and *Cx. intrincatus*. For the former, it was hard to predict an outcome of the anthropic process because of the change in the sign of the relationship between species' presence and time to port with sampling periods. For the latter, however, a high degree of anthropic intervention would favor its occurrence. This would indicate that some environmental features related with anthropogenic impacts produce differential responses among the members of the studied mosquito community.

Regarding landscape composition, the presence and relative proportions of the different land uses were relevant for *Cx. intrincatus* and *Ae. crinifer* occurrence. Both species were positively associated with secondary forests and negatively related with plantations, refuting the idea that forest management resembles natural vegetation growth, at least for these species requirements. Land use has been suggested to indirectly drive the mosquito community structure in the study area through differential GWH availability (Cardo et al. 2011). Hence, the response toward land use of *Cx. intrincatus* and *Ae. crinifer* may in fact reflect differences in composition and abundance of GWH types with variable breeding suitability. Although not all GWH types were present in every land use (which makes a statistical comparison to disentangle both effects impossible), our conclusions are still valid provided that differential GWH availability is a fixed characteristic of each land use. Regarding vegetation structure, all three species were negatively associated to physiognomies without canopy, suggesting certain dependence on shade, water temperature, or other associated factors that remain to be tested.

Certain issues should be taken into account while interpreting our results. First, presence/absence of immatures was considered separately by species and did not take into account potential competition effects (Vanwambeke et al. 2007b). A significant positive association between immatures of *Cx. dolosus* and *Ae. crinifer* as the one we described was recently reported by Senise and Sallum (2008) in Brazil. Second, although the methodology applied chooses some variables and excludes others because of collinearity and lower explanatory power, we cannot reject a possible secondary relation of the excluded variables with the presence of each species. A possible relation of selected variables with an unmeasured characteristic of the environment that may be the true driving variable determining species' presence cannot even be discarded. However, this limitation is not exclusive of

GLMM, but common to all linear regression methods, which are designed to get the optimal model for predictive relationships, rather than causal ones. At last, the large number of zeros in the data was reflected in the low values of cutoff points estimated by ROC curves.

Several arboviruses (e.g., Venezuelan equine encephalitis virus) have been isolated from *Culex (Melanoconion)* spp. in Argentina. Further investigations are needed to determine the role of the members of this subgenus in the transmission cycles of these viruses (Sabattini et al. 1998, Pires et al. 2009). Even though current vector status of *Ae. crinifer* is unknown, it was previously recorded as the most anthropophilic mosquito in the study area (Loetti et al. 2007). Therefore, it constitutes a serious nuisance for islanders as well as for tourists, and could pose a real danger if it were involved in the transmission of any disease.

This study was focused on the three most widespread species breeding in the Paraná Lower Delta, but several other species were also registered in the same ground water habitats (Cardo et al. 2011). Two of these, *Aedes albifasciatus* and *Culex pipiens*, are well recognized as mosquitoes of medical or veterinary concern (Mitchell et al. 1987, Avilés et al. 1992, Hamer et al. 2008, Vezzani et al. 2011). In addition, several arboviruses have been isolated from *Aedes scapularis*, *Aedes serratus*, and *Aedeomyia squamipennis* (Sabattini et al. 1998, Travassos Da Rosa et al. 2001). The methodological approach in this study, given the appropriate sampling size, could be applied to these species of proven medical concern.

In summary, the methodological approach hereby described attempts to pinpoint the environmental predictors of spatio-temporal mosquito distributions. Our study identified variables related with land use, anthropic intervention, and seasonality as environmental predictors of the distribution patterns of ubiquitous species breeding in ground water habitats. By identifying which times of the year and landscape features are more suitable for species' occurrence, this procedure could aid in the selection of priority areas for vector control and disease risk management. The effect of microenvironmental conditions, through physical and chemical water analyses, should be incorporated in future studies to a more comprehensive approach.

Acknowledgments

We thank Gabriel, Liliana, Ernesto, and Omar, the authorities of Estación Experimental Agropecuaria Delta del Paraná-INTA, Las Carabelas-Papel Prensa S.A., Automovil Club Argentino, Felicaria camp of San Fernando Municipality, Luz y Fuerza camp, and Parque Lyfe camp for allowing us to work at the study sites. Special thanks to Interisla S.A. for transportation discounts. This work was supported by Consejo Nacional de Investigaciones Científicas y Técnicas (PIP 112-200801-00743) and Universidad de Buenos Aires (UBACYT X608).

References Cited

- Akaike, H. 1974. A new look at the statistical model identification. *IEEE Trans. Automatic Control* 19: 716–723.
- Alfonzo, D., M. E. Grillet, J. Liria, J. C. Navarro, S. C. Weaver, and R. Barrera. 2005. Ecological characterization of the aquatic habitats of mosquitoes (Diptera: Culicidae) in enzootic foci of Venezuelan equine encephalitis virus in western Venezuela. *J. Med. Entomol.* 42: 278–284.
- Avilés, G., M. S. Sabattini, and C. J. Mitchell. 1992. Transmission of western equine encephalomyelitis virus by Argentine *Aedes albifasciatus* (Diptera: Culicidae). *J. Med. Entomol.* 29: 850–853.
- Balseiro, E. 1981. Análisis de la actividad nocturna de *Culex* (*C. eduardoi* García y Casal y *Aedes* (*O. crinitifer* (Theobald)) (Diptera, Culicidae), mediante el empleo de un atractivo lumínico. *Rev. Soc. Entomol. Argent.* 40: 211–219.
- Balseiro, E. 1989. Análisis de la actividad diaria de dípteros nematóceros en Punta Lara (Pdo. de Ensenada, Prov. de Buenos Aires). I. Culicidae. *Rev. Soc. Entomol. Argent.* 47: 157–161.
- Beketov, M. A., Y. A. Yurchenko, O. E. Belevich, and M. Liess. 2010. What environmental factors are important determinants of structure, species richness, and abundance of mosquito assemblages? *J. Med. Entomol.* 47: 129–139.
- Cardo, M. V., D. Vezzani, and A. E. Carbajo. 2011. Community structure of ground-water breeding mosquitoes driven by land use in a temperate wetland of Argentina. *Acta Trop.* (in press).
- Creessie, N.A.C. 1993. *Statistics for Spatial Data*. Wiley, New York, NY.
- Dale, P.E.R., and J. M. Knight. 2008. Wetlands and mosquitoes: a review. *Wetl. Ecol. Manag.* 16: 255–276.
- Darsie, R. F., Jr. 1985. Mosquitoes of Argentina. I. Keys for identification of adult females and fourth stage larvae in English and Spanish (Diptera, Culicidae). *Mosq. Syst.* 17: 153–253.
- DeLeo, J. M. 1993. Receiver operating characteristic laboratory (ROCLAB): software for developing decision strategies that account for uncertainty. In *Proceedings of the Second International Symposium on Uncertainty Modelling and Analysis*, 25–28 April 1993, College Park, MD. IEEE Computer Society Press, College Park, MD.
- de Little, S. C., D.M.J.S. Bowman, P. I. Whelan, B. W. Brook, and C.J.A. Bradshaw. 2009. Quantifying the drivers of larval density patterns in two tropical mosquito species to maximize control efficiency. *Environ. Entomol.* 38: 1013–1021.
- Donazar, J. A., F. Hiraldo, and J. Bustamante. 1993. Factors influencing nest site selection, breeding density and breeding success in the bearded vulture (*Gypaetus barbatus*). *J. Appl. Ecol.* 30: 504–514.
- Elith, J., C. H. Graham, R. P. Anderson, M. Dudík, S. Ferrier, A. Guisan, R. J. Hijmans, F. Huettmann, J. R. Leathwick, A. Lehmann, et al. 2006. Novel methods improve prediction of species' distributions from occurrence data. *Ecography* 29: 129–151.
- Fielding, A. H., and J. F. Bell. 1997. A review of the methods for the assessment of prediction errors in conservation presence/absence models. *Environ. Conserv.* 24: 38–49.
- Forattini, O. 2002. *Culicidologia Medica*, vol. 2. Editora da Universidade de São Paulo, São Paulo, Brazil.
- García, M. O., and H. Casal. 1965. Culicidae (Diptera) del Delta del Paraná. II. Apuntes sistemáticos y biológicos. *Delta del Paraná* 5: 5–16.
- Ginnig, J. E., A. W. Hightower, and W. A. Hawley. 2005. Application of geographic information systems to the study of the ecology of mosquitoes and mosquito-borne diseases. In W. Takken, P. Martens, and R. J. Bogers (eds.), *Environmental Change and Malaria Risk: Global and Local Implications*. Springer, Wageningen, The Netherlands.
- Guisan, A., and N. E. Zimmermann. 2000. Predictive habitat distribution models in ecology. *Ecol. Modell.* 135: 147–186.
- Hamer, G. L., U. D. Kitron, J. D. Brawn, S. R. Loss, M. O. Ruiz, T. L. Goldberg, and E. D. Walker. 2008. *Culex pipiens* (Diptera: Culicidae): a bridge vector of West Nile virus to humans. *J. Med. Entomol.* 45: 125–128.
- Hangley, J. A., and B. J. McNeil. 1982. The meaning and use of the area under a receiver operating characteristic (ROC) curve. *Radiology* 143: 29–36.
- Johnson, M. F., A. Gómez, and M. Pinedo-Vasquez. 2008. Land use and mosquito diversity in the Peruvian Amazon. *J. Med. Entomol.* 45: 1023–1030.
- Kandus, P., and A. I. Malvárez. 2004. Vegetation patterns and change analysis in the lower Delta Island of the Paraná River (Argentina). *Wetlands* 24: 620–632.
- Kandus, P., H. Karszenbaum, and L. Frulla. 1999. Land cover classification system of the Lower Delta of the Parana River (Argentina): its relationship with Landsat Thematic Mapper Spectral Classes. *J. Coastal Res.* 15: 909–926.
- Kandus, P., R. D. Quintana, and R. F. Bó. 2006. Patrones de paisaje y biodiversidad del bajo Delta del Río Paraná. Mapa de ambientes. Pablo Casamajor, Buenos Aires, Argentina.
- Laird, M. 1988. *The Natural History of Larval Mosquito Habitats*. Academic, London, United Kingdom.
- Landis, J. R., and G. C. Koch. 1977. The measurement of observer agreement for categorical data. *Biometrics* 33: 159–174.
- Leisnham, P. T., D. P. Slaney, P. J. Lester, and P. Weinstein. 2005. Increased mosquito densities from larval habitats in modified landuses in the Kapiti region, New Zealand: vegetation, water quality and predators as associated environmental factors. *EcoHealth* 2: 1–10.
- Loetti, V., N. Burroni, and D. Vezzani. 2007. Seasonal and daily activity patterns of human-biting mosquitoes in a wetland system in Argentina. *J. Vector Ecol.* 32: 358–365.
- Maciá, A. 1997. Age structure of adult mosquito (Diptera: Culicidae) populations from Buenos Aires Province, Argentina. *Mem. Inst. Oswaldo Cruz* 92: 143–149.
- Maciá, A., J. J. García, and R. E. Campos. 1995. Bionomía de *Aedes albifasciatus* y *Ae. crinitifer* (Diptera: Culicidae) y sus enemigos naturales en Punta Lara, Buenos Aires. *Neotropica* 41: 43–50.
- Maciá, A., J. J. García, and R. E. Campos. 1996. Seasonal variation of three *Culex* species (Diptera: Culicidae) and its parasites and pathogens in Punta Lara, Buenos Aires, Argentina. *Rev. Biol. Trop.* 44–45: 267–275.
- McCullagh, P., and J. A. Nelder. 1989. *Generalized Linear Models*. Chapman & Hall, London, United Kingdom.
- Mitchell, C. J., T. P. Monath, M. S. Sabattini, H. A. Christensen, R. F. Darsie, Jr., W. L. Jakob, and J. F. Daffner. 1987. Host-feeding patterns of Argentine mosquitoes (Diptera: Culicidae) collected during and after an epizootic of western equine encephalitis. *J. Med. Entomol.* 24: 260–267.
- Muturi, E. J., J. Mwangangi, J. Shililu, B. Jacob, C. Mbogo, J. Githure, and R. Novak. 2008. Environmental factors associated with the distribution of *Anopheles arabiensis* and *Culex quinquefasciatus* in a rice agroecosystem in Mwea, Kenya. *J. Vector Ecol.* 33: 56–63.

- Nicholls, A. O. 1989. How to make biological surveys go further with generalised linear models. *Biol. Conserv.* 50: 51–75.
- Patterson, S., and J. Lello. 2003. Mixed models: getting the best use of parasitological data. *Trends Parasitol.* 19: 370–375.
- Pires, D. A., M. Laurito, W. R. Almirón, and R. M. Gleiser. 2009. First record of *Culex (Melanoconion) pilosus* from Córdoba City, Argentina. *J. Am. Mosq. Control Assoc.* 25: 206–207.
- Prosen, A. F., A. Martínez, and R. U. Carcavallo. 1960. La familia Culicidae (Diptera) en la ribera fluvial de la Provincia de Buenos Aires. *Anal. Inst. Med. Reg.* 5: 101–113.
- R Development Core Team. 2009. R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna, Austria.
- Reiter, M. E., and D. A. LaPointe. 2007. Landscape factors influencing the spatial distribution and abundance of mosquito vector *Culex quinquefasciatus* (Diptera: Culicidae) in a mixed residential–agricultural community in Hawaii. *J. Med. Entomol.* 44: 861–868.
- Rochlin, I., K. Harding, H. S. Ginsberg, and S. R. Campbell. 2008. Comparative analysis of distribution and abundance of West Nile and eastern equine encephalomyelitis virus vectors in Suffolk County, New York, using human population density and land use/cover data. *J. Med. Entomol.* 45: 563–571.
- Rochlin, I., T. Iwanejko, M. E. Dempsey, and D. V. Ninivaggi. 2009. Geostatistical evaluation of integrated marsh management impact on mosquito vectors using before–after–control–impact (BACI) design. *Int. J. Health Geogr.* 8: 35.
- Ronderos, R., J. Schnack, and A. Maciá. 1992. Composición y variación estacional de una taxocenosis de Culicidae del ecotono subtropical pampásico (Insecta, Diptera). *Graellsia* 48: 3–8.
- Rossi, G. C., J. C. Mariluis, J. A. Schnack, and G. R. Spinelli. 2002. Dipteros Vectores (Culicidae y Calliphoridae) de la Provincia de Buenos Aires. Universidad de La Plata, Buenos Aires, Argentina.
- Sabattini, M. S., G. Avilés, and T. Monath. 1998. Historical, epidemiological and ecological aspects of arboviruses in Argentina: Flaviviridae, Bunyaviridae and Rhabdoviridae, pp. 113–134. In A.P.A. Travassos da Rosa, P.F.C. Vasconcelos, and J.F.S. Travassos da Rosa (eds.), *An Overview of Arbovirology in Brazil and Neighbouring Countries*. Instituto Evandro Chagas, Belem, Brazil.
- Senise, L. V., and M.A.M. Sallum. 2008. Redescription of *Culex (Culex) dolosus* (Lynch Arribálzaga) (Diptera: Culicidae), based on specimens from Pico do Itapeva, Serra da Mantiqueira, São Paulo, Brazil. *Zootaxa* 1683: 51–62.
- Service, M. W. 1995. Mosquitoes (Culicidae). In R. P. Lane and R. W. Crosskey (eds.), *Medical Insects and Arachnids*. Chapman & Hall, London, United Kingdom.
- Sokal, R. R., and F. J. Rohlf. 1969. *Biometry: the Principles and Practice of Statistics in Biological Research*. W. H. Freeman, San Francisco, CA.
- Titus, K., and J. A. Mosher. 1984. Chance-corrected classification for use in discriminant analysis: ecological applications. *Am. Midl. Nat.* 111: 1–7.
- Travassos Da Rosa, A.P.A., M. J. Turell, D. M. Watts, A. M. Powers, P.F.C. Vasconcelos, J. W. Jones, T. A. Klein, D. J. Dohm, R. E. Shope, N. Degallier, et al. 2001. Trocara virus: a newly recognized Alphavirus (Togaviridae) isolated from mosquitoes in the Amazon Basin. *Am. J. Trop. Med. Hyg.* 64: 93–97.
- Vanwambeke, S. O., E. F. Lambin, M. P. Eichhorn, S. P. Flasse, R. E. Harbach, L. Oskam, P. Somboon, S. van Beers, B.H.B. van Benthem, C. Walton, et al. 2007a. Impact of land-use change on dengue and malaria in Northern Thailand. *EcoHealth* 4: 37–51.
- Vanwambeke, S. O., P. Somboon, R. E. Harbach, M. Isenstadt, E. F. Lambin, C. Walton, and R. K. Butlin. 2007b. Landscape and land cover factors influence the presence of *Aedes* and *Anopheles* larvae. *J. Med. Entomol.* 44: 133–144.
- Vezzani, D., M. Mesplet, D. F. Eiras, M. F. Fontanarrosa, and L. Schnittger. 2011. PCR detection of *Dirofilaria immitis* in *Aedes aegypti* and *Culex pipiens* from urban temperate Argentina. *Parasitol. Res.* 108: 985–989.
- [WHO] World Health Organization. 2002. *World Health report: reducing risks, promoting healthy life*. World Health Organization, Geneva, Switzerland.
- Zuur, A. F., E. N. Ieno, N. J. Walker, A. A. Saveliev, and G. M. Smith. 2009. *Mixed Effects Models and Extensions in Ecology with R*. Springer, New York, NY.

Received 4 March 2011; accepted 10 June 2011.