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Spatial pattern of abundance of the mosquito, *Ochlerotatus albifasciatus*, in relation to habitat characteristics

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Abstract. *Ochlerotatus albifasciatus* (Macquart) (Diptera: Culicidae) is the main vector of the western equine encephalomyelitis (WEE) virus and potentially of other arboviruses in Argentina. Surges of adult population abundance during the rainy season are a nuisance, affecting milk and beef production. Larvae develop in short periods in shallow temporary ground pools on fresh or brackish water. Although adults seem to disperse long distances from larval habitats, little is known about their habitat preferences. This work studied factors affecting the spatial pattern of adult *Oc. albifasciatus* abundance.

Adult mosquitoes were collected using CDC miniature light traps baited with CO_2 at 28 sites located to the south of Mar Chiquita Lagoon, from November 1997 to April 1998. Each site was typified according to its predominating vegetation cover, potential breeding site occurrence, land slope and cattle density. The spatial and temporal patterns of abundance suggested that *Oc. albifasciatus* prefers prairies and natural grasslands subject to periodic flooding vs. woodland and farm land. A discriminant function based on the proximity to potential larval habitats, distance to woodland and land slope accurately classified 95% of the data categorized as having an average high (>500 mosquitoes) or low (<500 mosquitoes) abundance, and was validated using six sites located away from the study area. An analysis of the temporal variation of mosquito abundance highlighted the influence of the dynamics of the larval habitats on adult mosquito abundance.

Key words. *Ochlerotatus albifasciatus*, Culicidae, Diptera, GIS, habitat preferences, spatial pattern, Mar Chiquita, Argentina.

Introduction

Ochlerotatus (*Ochlerotatus*) *albifasciatus* (Macquart), previously included in the genus *Aedes* until Reinert's (2000) proposal to promote the subgenus *Ochlerotatus* to genus rank, is a very common mosquito in southern South America. This species may be found in hilly habitats and as high as 2300 m above sea level (Forattini, 1965), but it is on plains where it can become a nuisance, where breeding sites in small ground depressions are most frequently found after rain.

Ochlerotatus albifasciatus is the main vector of western equine encephalomyelitis (WEE) virus and potentially of other arboviruses in Argentina (Mitchell *et al.*, 1987; Avilés *et al.*, 1992). Additionally, outbreaks of this species irritate livestock and thus affect milk and beef production (Raña *et al.*, 1971).

Extended wetlands associated with the Mar Chiquita basin of central Argentina, and numerous temporary ground pools and ponds that form during the rainy season, provide breeding sites for mosquitoes (Bachman & Casal, 1962; Ludueña Almeida & Gorla, 1995). Increases in the

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adult mosquito population during the rainy season, when *Oc. albifasciatus* is the most abundant species, may result in annual losses for the Mar Chiquita region of around US$5.28 million (Hall & Hillyard, 1993; Ludueña Almeida, 1994). Fluctuations of the flooding levels are determinants of the presence of *Oc. albifasciatus* larvae, and peaks of adult populations usually occur after periods of drought (Ludueña Almeida & Gorla, 1995; Fontanarrosa *et al.*, 2000; Gleiser *et al.*, 2000). The time of synchronous adult emergence from a breeding site may be predicted by monitoring rainfall and average temperature (Almirón *et al.*, 2000; Fontanarrosa *et al.*, 2000; Gleiser *et al.*, 2000), and also from meteorological satellite data (Gleiser *et al.*, 1997).

Previous regional studies in Mar Chiquita have indicated that *Oc. albifasciatus* abundance is heterogeneously distributed and influenced not only by weather events but also by habitat characteristics (Gleiser *et al.*, 2000; Gleiser & Gorla, 2001), as a negative relationship between adult abundance and distance from larval habitats has been reported (Gleiser *et al.*, 2000). Larvae have been collected mainly in natural habitats, in temporary shallow stagnant ground pools, where aquatic vegetation can either be present or absent, either in sunlight or shade sites (Almirón & Brewer, 1996; Bruzzone & Schweigmann, 1998), either in fresh or hypersaline water, with a pH range of 6.4–8.0 (Bachman & Casal, 1962; Almirón & Brewer, 1996).

Only a few vegetation types can develop in areas subject to periodic flooding (Davy *et al.*, 1990), where *Oc. albifasciatus* is most likely to breed, so there is probably a relationship between habitat type and abundance of the mosquito. Floristic and physiognomic variations were reported in relation to the hydrological and geo-morphological features of the Mar Chiquita Depression (Menghi & Herrera, 1995), where some plant species are good indicators of salinity levels and/or flood occurrence, according to their presence, frequency and association. In the Saladillo region, to the West of Mar Chiquita Lagoon, larvae have been collected in ground pools between *Atriplex* sp. ('jumiales') (Bachman & Casal, 1962).

The aim of this work was to analyse the spatial pattern of adult female *Oc. albifasciatus* abundance in relation to local factors that may influence important components of its life cycle, such as occurrence of potential breeding sites, suitable resting sites and possible sources of bloodmeals. The identification of environmental factors more suitable for *Oc. albifasciatus* development or activities could aid in the risk assessment of WEE transmission and to plan more efficient mosquito control programmes.

Materials and methods

Study site

The study site was a 14×14 km area located to the south-east of Mar Chiquita Lagoon (coordinates of upper right and lower left corners: 30°50′ S 62°27′ W–31°2′ S 62°13′ W), in Cordoba province, Argentina (Fig. 1). The dominant physiognomic groups are related to topographic variations and to the influence of surface and underground water (Menghi & Herrera, 1995, 1998). The terrain is lower to the west and north-west (70–72.5 m a.s.l.), where halophyllous scrubland and xero-halophyllous woodland patches grow on brackish wet soils, whereas grasslands are related to areas that flood periodically (Del Sueldo, 1995). These grass communities have been described in detail for the wetlands associated with the Dulce River delta, north of Mar Chiquita (Menghi & Herrera, 1995) as *Spartina argentinensis* grassland ('espartal'), *Distichlis* grasslands, *Allenrolfea vaginata*, *Salicornia ambigua* and *Sesuvium portulacastrum* sp. prairies. There are a few patches of the original sclerophyllous thorny forest (*Prosopis* sp., *Aspidosperma quebracho-blanco*, *Celtis tala*), mainly in field borders, and *Geoffroea decorticans* woods grow in deforested areas. The terrain is higher to the East (approximately 110 m a.s.l.) where the main land use is cattle grazing and pasture cropping. A branch of the Xanaes River crosses the study site developing into interconnected brackish ponds and lagoons that drain in the Mar Chiquita.

Mosquito abundance

Mosquitoes were collected at 28 sampling sites (Fig. 1) with CDC miniature light traps baited with CO_2 (500 g of dry ice wrapped with cardboard paper). One trap per site was operated overnight on 11 and 24 November, 16 and 27 December 1997, 20 January, 2 and 27 February and 1 April 1998. All traps were suspended at a height of 1.5 m, from either trees or man-made structures. On some occasions a trap malfunctioned or could not be activated because of severe climatic conditions and collections from those traps are missing. From a total of 28 sites, data from six sites were discarded as traps were operated on less than four dates. Data from 11 November and 2 February were also excluded from the analysis, as less than 50% of the traps were operated on those dates. Only data on female *Oc. albifasciatus* are reported here.

Environmental variables

Habitats characterized by similar surface water dynamics and predominant plant coverage were grouped into habitat types, as described in Table 1. A habitat type map was digitized (Idrisi32 and Cartalinx) based on field surveys and interpretation of a Landsat image for January 1997. To compare the average *Oc. albifasciatus* abundance among habitat types, sites were assigned to the habitat type most represented at the site. For multiple regression and discriminant analysis, each site was described as covered by fractions of the habitat types present within a 500 m radius (e.g. a site was 50% type 1, 30% type 2 and 20% type 3). The percentage coverage was estimated as the proportion of pixels belonging to a particular habitat type within a 500 m radius around each site. Also, because mosquito

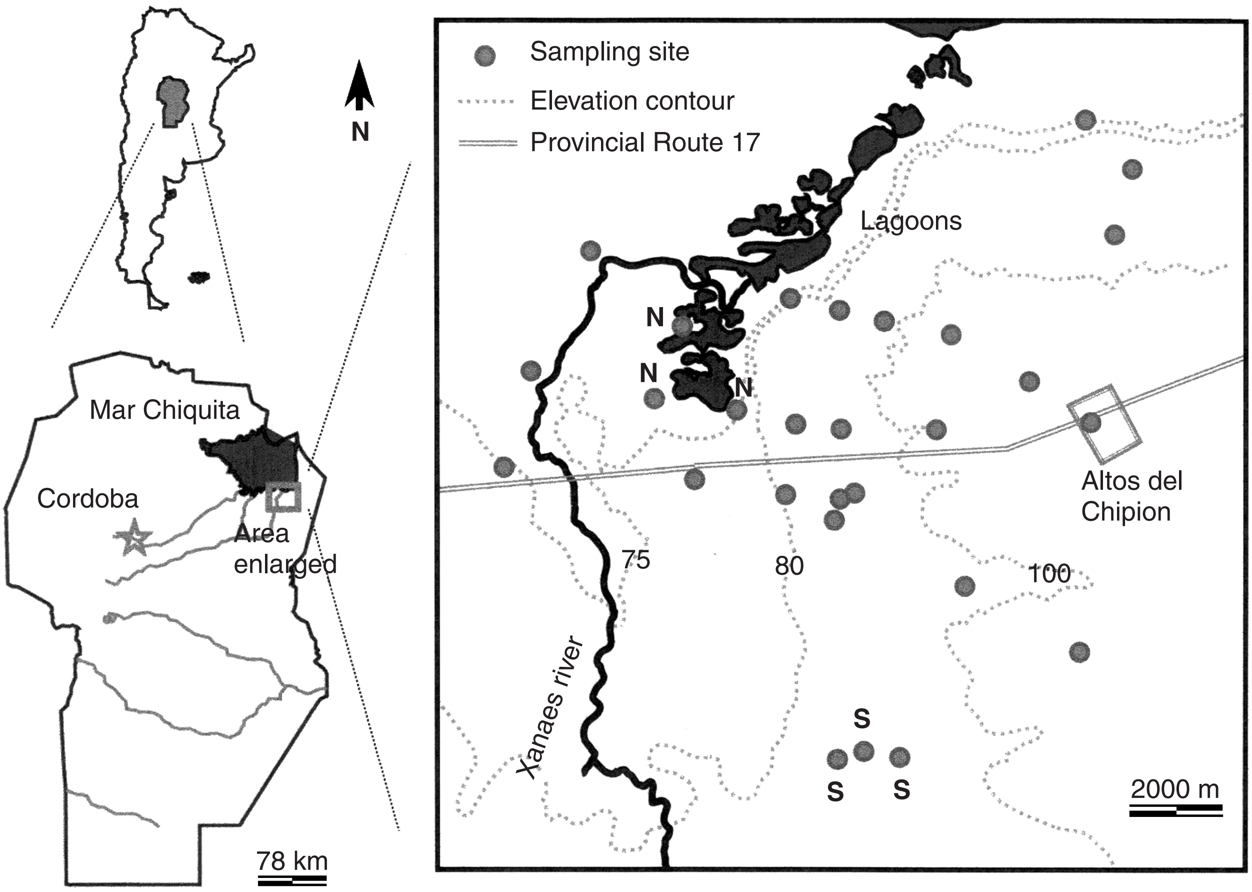

Fig. 1. Location of the study area and sampling sites. S and N indicate South and North sector sites, respectively, referred to in the text and in Fig. 3.

abundance at a site may be influenced by proximity to favorable habitats, the Euclidean distance (m) from a sampling site to a particular habitat type was estimated (Idrisi32) based on the habitat type map.

A cattle density (cows/ha) map was created based on the number of cows in each grazing parcel, where each pixel was assigned a cattle density value. Then, the average cattle density within a 500 m radius around a site was estimated. The number of cows in each parcel was kindly provided by Veterinarian F. Casale, responsible for vaccination campaigns in the Altos del Chipión area.

A digital elevation model (DEM) was developed based on contours of a topographic map (n°3163-17-3) produced by the Instituto Geográfico Militar of Argentina, where each

Table 1. Habitat types based on surface water dynamics and land cover at a regional scale.

Water bodies	Habitat type	Characteristics
Permanent	Permanent water body	Lagoons and Xanaes River, holding water year round.
Temporary		
Stagnant	Prairies	Low areas with subcircular depressions holding water, covered by *Salicornia ambigua* and *Sesuvium portulacastrum* communities.
	Ditches	Man made ditches, usually covered with grasses such as *Cynodon dactylon*, where water accumulates.
Running	Grasslands	Flat low areas covered mainly by *Spartina argentinensis* and *Distichlis* sp., where water runs in sheets.
	Shrubs	Woody scrubs including *Allenrolfea* spp., *Maytenus* spp., *Geoffroea decorticans*. Ephemeral surface water.
	Woods	Woodland remnants or groups of trees on pasture edges (*Prosopis* spp., *Aspidosperma quebracho-blanco*), elevated terrains that do not flood.
	Farmlands	Generic name for pasture fields and crops (alfalfa, sorghum, maize), where surface water is ephemeral.

pixel was assigned a height (in m a.s.l.). The average slope (expressed as a percentage) in each site was estimated based on the DEM using interpolation algorithms.

Rainfall was recorded with a rain gauge in Altos del Chipión, a town within the study area.

Statistical analysis

Differences among habitat types were assessed by a one-way ANOVA with LSD as post-hoc test. Abundance data were log transformed ($\ln(x+1)$) to fit the ANOVA assumptions.

Multiple regression and discriminant analysis were estimated to determine which factors better explained the spatial distribution of *Oc. albifasciatus* abundance. Data on *Oc. albifasciatus* abundance were log-transformed ($\ln(x+1)$) for multiple regression analysis, in order to fit the assumptions of the analysis (Statistica 4.3). The dependent variable was the average mosquito abundance in each site during the study period. For the discriminant analysis, average *Oc. albifasciatus* abundance in each site was classified as low (average abundance <500 mosquitoes) or high (average abundance >500 mosquitoes). The independent variables were percentage covered by each habitat type within 500 m of each sampling site, distance to each habitat type, land slope and cattle density.

Results

The average adult female *Oc. albifasciatus* abundance was significantly higher ($P < 0.05$) in Salicornia and Sesuvium prairies as compared to shrubs, woods, farmland and ditches (Fig. 2). No significant differences were observed between *Distichlis* grasslands and the other habitats.

A significant model explaining 49% of the spatial pattern of *Oc. albifasciatus* abundance was fitted by a forward stepwise multiple regression. The variables included in the model were the percentage covered with Salicornia and Sesuvium prairies, distance to woodland, and land slope (Table 2). This model indicates that mosquito density is higher in sites covered by prairies, and lower near woods and terrain with steeper slope (although land slope was marginally not significant, $P < 0.07$).

A forward stepwise discriminant analysis of the variables listed in Table 3 was carried out to determine which variables contributed most to the discrimination between high and low abundance, giving a significant fit ($P < 0.015$). The standardized coefficients show distance to ditches as the variable with highest relative weight (Table 3). Then a significant ($P < 0.004$) discriminant function was fitted for the five variables with the highest standardized coefficients ($P < 0.08$) (Table 4). This function correctly classified a posteriori 100% of the sites with low abundance (<500 mosquitoes) and 92.3% of the sites with high abundance ($\chi^2 = 18.28$, $P < 0.001$; Table 5).

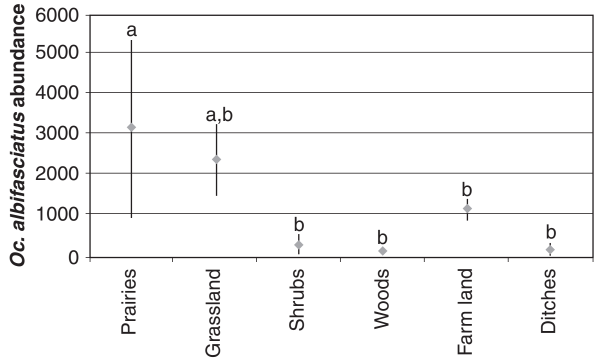

Fig. 2. Adult female *Oc. albifasciatus* abundance in each habitat type (mean abundance ± standard error). a, b Means with no common superscript differ significantly ($P < 0.05$) by LSD test.

To validate the discriminant function another six sites located outside the study area were tested (three sites whose average abundance were >500 mosquitoes and three sites <500 mosquitoes, respectively). Mosquitoes in those sites were trapped between October and March from 1994 to 1997 (Gleiser *et al.*, 2000). Table 6 shows the location of each site and the values for each independent variable. All six sites were correctly classified.

Discriminant analysis as well as multiple regression showed a significant relationship between mosquito abundance and environmental factors (habitat type, land slope) that may be related to potential breeding sites, although the significant variables for each analysis differed.

The temporal pattern of mosquito abundance in the study area was analysed in order to better interpret the results. The average *Oc. albifasciatus* abundance on each date was significantly correlated to rainfall accumulated 7–15 days before each sampling ($r = 0.88$; $n = 28$; $P < 0.005$; abundance $= -191.371 + 23.213 *$ accumulated rainfall), although abundance variation differed from site to site throughout the study period, ranging from an average of four females (standard deviation of 5) to 3880 females (standard deviation 7071). Figure 3 shows a temporal profile

Table 2. Multiple forward stepwise regression of adult female *Ochlerotatus albifasciatus* abundance in relation to site characteristics ($R^2 = 0.49$; $n = 22$; $P < 0.01$).

	Coefficient	Standard error	Standard coefficient	Standard error	*P*
Intercept	1.907	0.165			<0.001
Distance to woods	0.0006	0.0002	0.495	0.17	<0.001
Distance to prairies	2.655	1.105	0.423	0.176	<0.03
Land slope	−0.148	0.122	−0.214	0.175	<0.07

Table 3. Forward stepwise discriminant analysis based on a classification factor represented by two categories of adult female *Ochlerotatus albifasciatus* mean abundance (low abundance <500 and high abundance >500 mosquitoes).

Variables	*F*	*P*	Coefficient	Standardized coefficient
Included in the model:	*F* to remove:			
Distance to ditches	7.709	0.015	−0.005	−2.673
Distance to grasslands	6.831	0.02	−0.001	−1.777
Distance to woods	3.621	0.08	0.004	1.638
Distance to permanent water	4.202	0.06	0.001	1.4001
% land slope	5.155	0.04	−1.042	−0.831
% covered with ditches	1.487	0.24	−31.235	−0.645
% covered with prairie	1.05	0.32	4.429	0.401
% covered with woods	0.041	0.84	−0.959	−0.097
Not included in the model:	*F* to include:			
Distance to prairies	0.691	0.42		
Distance to shrubs	0.118	0.74		
Distance to crops	0.436	0.52		
Cattle density	0.303	0.59		
% covered with permanent water	0.411	0.53		
% covered with grassland	0.052	0.82		
% covered with shrubs	0.131	0.72		
% covered with crops	0.156	0.7		

Table 4. Discriminant analysis function for two adult female *Ochlerotatus albifasciatus* abundance categories: category 0 < 500 mosquitoes and category 1 > 500 mosquitoes.

Variable	Coefficient	Standardized coefficient	*P*
Distance to ditches	−0.00445	−2.11483	0.003
Distance to Spartina-Distichlis grassland	−0.00157	−1.98918	0.004
Distance to woods	0.00345	1.41493	<0.02
Distance to permanent water	0.00076	1.39298	0.03
% slope	−1.13482	−0.90590	0.01

of rainfall accumulated 7–15 days before each collection and of mosquito abundance in two sectors, denominated North and South, where higher abundances were recorded. Each sector represents the average abundance per date at three sites (indicated in Fig. 1). Mosquito abundance increased in both areas on 16 December, after 244 mm of rainfall accumulated during the previous 2 weeks. Even though the amount of rainfall was high in December, mosquito abundance gradually decreased in the North, whereas it continued increasing in the South, showing a peak of almost 12 000 mosquitoes on 27 December.

Table 5. A posteriori classification matrix of *Ochlerotatus albifasciatus* average density categories (low < 500 mosquitoes; high = 500 mosquitoes) in relation to distance from site to permanent water, *Spartina* and *Distichlis* grassland, woods and ditches, and land slope (%).

	Predicted		
Recorded	Low	High	Correct (%)
Low	9	0	100
High	1	12	92.308
Total	10	12	95.454

Discussion

Associations between mosquitoes and plant communities have been reported for several mosquito species (Horsfall, 1963; Dale *et al*., 1986; Rejmankova *et al*., 1998), as not all vegetation is equally attractive to mosquitoes. In malaria endemic areas in Papua New Guinea, variations in the spatial distribution of anopheline mosquito species were found related with the distribution of each species' larval habitats and vegetation, which accounted for the high variability in the transmission of the sporozoite between sites located a few hundred metres apart (Hii *et al*., 1997). In North America, *Culiseta melanura* utilizes unique breeding and resting habitats such as wooded swamps and adjacent hardwood forests (Pierson & Morris, 1982; Guimaraes *et al*., 2000; Moncayo *et al*., 2000).

Gabinaud (1975) reported that floristic composition and vegetation structure may be used to map the distribution of eggs of *Aedes caspius* and *Ae. detritus* in space and in time, and also the resting sites of adults. In plains of the pacific coast of Chiapas, Mexico, a strong relationship between the larval density of *Anopheles albimanus*, the main vector of malaria in Mexico and Central America, and the vegetation type predominating in the larval habitats was reported

Table 6. Location and values for each independent variable of the six sites tested to validate a discriminant function between mean abundance categories (low < 500 mosquitoes and high > 500 mosquitoes) of adult *Ochlerotatus albifasciatus*. *Ochlerotatus albifasciatus* (*Oc. albif.*) abundances represent average abundance at each site and their respective standard deviation in parenthesis.

Site	Location	Distance to water (m)	Distance to grassland (m)	Distance to woods (m)	Distance to ditches (m)	Slope (%)	*Oc. albif.* abundance
1	30°45′ S 63°12′ W	5000	200	30	50	<0.01	521 (1825)
2	30°44′ S 63°06′ W	5000	50	1000	10	<0.01	1013 (1948)
3	30°43′ S 63°06 W	5000	150	1000	100	<0.01	537 (1299)
4	31°02′ S 62°53′ W	6000	5000	10	6000	0.05	0.7 (2.3)
5	30°56′ S 62°40′ W	20	2500	0	100	0.2	3.9 (11.6)
6	30°54′ S 62°38′ W	200	200	2	1500	0.2	14.7 (40.7)

(Rodríguez *et al.*, 1993). There and elsewhere, the discrimination of the land cover units related to larval habitats, based on Landsat TM imagery, was useful to predict anopheline production (Pope *et al.*, 1992; Beck *et al.*, 1997).

A priori it was expected that *Oc. albifasciatus* abundance in the study area would be higher in *Salicornia ambigua* and *Sesuvium portulacastrum* prairies, where subcircular depressions on the ground can provide temporary breeding sites for mosquitoes, and in the lagoon areas. In fact, Fig. 2 shows that those habitats showed the highest average mosquito abundances.

The relationship between adult female *Oc. albifasciatus* abundance and several independent variables was analysed by multiple regression and discriminant analysis. A multiple regression explained 49% of the spatial variability of adult *Oc. albifasciatus* based on percentage covered by *Salicornia ambigua* and *Sesuvium portulacastrum* prairies, land slope and distance to woods, suggesting that abundance is higher in areas that flood periodically, with low slope and away from woodlands.

A discriminant function was fitted for two categories of *Oc. albifasciatus* average abundance (high >500 mosquitoes; low <500 mosquitoes) based on proximity to ditches and *Spartina argentinensis–Distichlis* spp. grasslands, distance to woods, permanent water, and land slope. This function correctly classified a posteriori 95.45% of the field data (Table 5), and was validated for another six sites, all of them correctly classified, located outside the study area (Table 6) (the most distant site was approximately 80 km away).

Even though during December 1997 mosquito abundance was high in the southern pastures where cattle grazed (up to

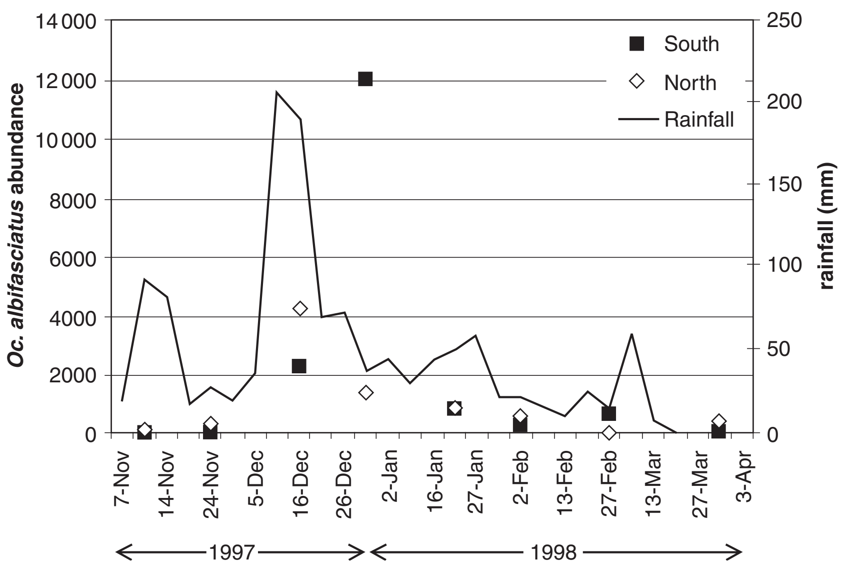

Fig. 3. Biweekly rainfall (mm) and adult female *Oc. albifasciatus* abundance in the two sectors (shown in Fig. 1) where the highest mean abundances were recorded.

19.567 mosquitoes at one site), no significant relation was observed between mosquito and cattle density. As *Oc. albifasciatus* can feed on a number of hosts besides cows, such as birds and other mammals (Almirón & Brewer, 1995), it is possible that for the mosquito abundances recorded in this study, there were enough hosts near the larval habitats so the females did not have to disperse long distances to reach the blood source; unfortunately, no attempt was made at the time to survey other potential hosts.

The spatial distribution of breeding sites provably changes throughout the rainy season. In the north-west of the study area, at the beginning of the rainy season, small temporary pools form providing breeding habitat but later become connected to the permanent nearby lagoons and are colonized by fish and other predators. De Souza *et al.* (1997) reported passive transportation of larvae by water streams and river freshnets produced by floods in their study in the Mar Chiquita area. On the other hand, no permanent water bodies form in the southern part of the study area as a result of drainage programs (L. Riera, personal communication), and water collections after rains last relatively short periods of time, not long enough to allow colonization by predators, but allowing *Oc. albifasciatus* to develop to adulthood. The results presented here support this hypothesis, as mosquito abundance increases with the first rains in both regions (Fig. 3). Then, in mid-December, the flooded surface increases in the south, resulting in a higher availability of breeding sites (resulting in higher adult abundance), but lowering the suitable breeding sites in the northern region, as rain pools would become connected to the permanent ponds. Towards the end of the summer, together with lower rainfall volume the flooded surface would decrease, and thus there would only be pools near the lagoons.

Although the spatial pattern of *Oc. albifasciatus* abundance is associated with environmental variables (such as vegetation characteristics and land slope) that can be related to potential breeding habitats, the significant variables in the multiple regression analysis and in the discriminant analysis differed (Tables 2–4). An analysis of the spatial pattern of abundance throughout the study period suggests that a lack of simple and direct relation between the variables may result because the spatial distribution of suitable larval habitats changes during the rainy season. Consistently, during the study period, mosquito density in the north region (permanent lagoons) increased at the onset of the rainy season, but did not reach levels of economic importance. By contrast, mosquito density reached high levels in the southern area, where pools last long enough for mosquitoes to develop and emerge as adults, but not enough for predators to colonize those habitats. More detailed field studies of the temporal dynamics of *Oc. albifasciatus* larval habitats are necessary to confirm this hypothesis.

Drainage of wetlands south of Mar Chiquita lagoon has been proposed as a way to reduce mosquito populations. Our results suggest that this would only be effective if no temporary pools formed after a rain, because temporary pools would produce more *Oc. albifasciatus* than more permanent ponds. The draining of wetlands would also have negative effects on bird fauna and habitat dynamics, of particular importance in the Mar Chiquita wetlands system. The area was declared a Provincial Reserve (Natural Areas Law 6964/83, Decree 3215/94) and Western Hemisphere Shorebird Reserve Network Hemispheric site (http://www.manomet.org/WHSRN/index.html) in 1993 because of its rich bird fauna and a status as a main wintering and/or breeding site for flamingos and several migratory shorebirds.

Although epidemics of western equine encephalitis and the presence of other potentially important arboviruses have been reported in Argentina (Mitchell *et al.*, 1987; Avilés *et al.*, 1992), the ecology of these mosquito-borne diseases is still very poorly understood. The identification of factors that affect the abundance and spatial distribution of the mosquito vectors is needed to develop models that predict risk of exposure of humans or cattle to WEE and to plan mosquito control campaigns. This study shows that the spatial pattern of *Oc. albifasciatus* abundance within the Mar Chiquita area is influenced by land cover, and a model has been proposed and evaluated that predicts areas more suitable for *Oc. albifasciatus*.

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