

## Seasonal and daily activity patterns of human-biting mosquitoes in a wetland system in Argentina

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**ABSTRACT:** Seasonal and daily activity patterns of human-biting mosquitoes were studied in the lower delta of the Paraná River from March 2003 to February 2004. Monthly captures at four daytime intervals using human volunteers collected 1,289 mosquitoes belonging to 14 species and six genera, with the most frequently captured being *Ochlerotatus crinifer* (49%), *Psorophora ferox* (36%), *Ochlerotatus serratus* (5%), and *Isotomyia paranensis* (3%). *Oc. crinifer* was collected during the four seasons and showed higher values in Summer and Autumn. *Ps. ferox* and *Oc. serratus* were not present in Winter and the highest values were recorded in Summer. Monthly captures of *Ps. ferox* and *Oc. serratus* were positively associated with temperature. With regard to daily activity patterns, *Oc. crinifer*, *Oc. serratus*, and *Ps. ferox* were captured during the four collection intervals with nocturnal captures concentrated during the warm months. The daily activity patterns of these species changed throughout the seasons. *Journal of Vector Ecology* 32 (2): 358-365. 2007.

**Keyword Index:** *Ochlerotatus*, *Psorophora*, *Isotomyia*, mosquitoes, vector ecology, wetland system.

### INTRODUCTION

Seasonality and circadian rhythm of mosquito populations, as well as other ecological and behavioral features, are strongly influenced by climatic factors such as temperature, rainfall, humidity, wind, and duration of daylight (Reiter 2001). Both seasonal and daily activity patterns of mosquito vectors are required as baseline knowledge to understand the transmission dynamics of vector-borne pathogens (Reiter 2001, Lord 2004), and have been widely studied for many mosquito species throughout the world (e.g., Guimarães et al. 2000a,b, Jones et al. 2004).

In Argentina, seasonal abundance and activity patterns were previously reported for several mosquito species. Seasonal abundance patterns were evaluated by monitoring populations of immatures and seasonal activity patterns (i.e., the variation of the host-seeking activity throughout the seasons) by CDC light traps (Ronderos et al. 1992, Campos et al. 1993, 1995, García et al. 1995, Almirón and Brewer 1995, Maciá et al. 1995, 1997). However, only a few studies assessed seasonal biting activity of mosquitoes attracted to humans (García and Casal 1965, Hack et al. 1978, Ludueña Almeida and Gorla 1995). Similarly, there is little information on daily activity patterns of mosquitoes from Argentina (García and Casal 1965, Hack et al. 1978, Balseiro 1981, 1989, Ludueña Almeida and Gorla 1995).

To our knowledge, there is no current information on the mosquito fauna from the delta of the Paraná River (Buenos Aires Province). With the aim of contributing to the knowledge of human-biting mosquitoes from Argentina, we evaluated the seasonal and daily activity patterns of mosquitoes captured by using human volunteers in the lower delta of the Paraná River.

### MATERIALS AND METHODS

#### Study area

The delta of the Paraná River is one of the most important wetland systems in South America. The study site (Santa Clara Island; 34°11'S, 58°29'W) is located in the downstream portion of the lower delta, which is characterized by newly-formed islands directly connected to De la Plata River. This zone is mainly influenced by lunar and wind tides and not by seasonal floods. Lunar tides have 1 m of normal amplitude and southeast winds can raise the water up to 2.5 m over the mean value. The islands are characterized by a pan-shaped form with a surrounding levee as high as 3 m above water level, a central depression temporarily or permanently flooded, and in some cases, channels that increase water outflow to enhance soil drainage. Levees typically occupy 20% of the islands' surface and its original vegetation (complex forest) has been replaced almost totally by human plantations mainly represented by willow (*Salix* spp.) and poplar (*Populus* spp.). The only natural forest that remains in this region is dominated by seibo (*Erythrina cristagalli* L.). Lowlands are characterized by bulrush plant communities dominated by *Scirpus giganteus* Kunth and *Schoenoplectus californicus* (CA Mey) Soják. The climate is moderate with mean annual temperature of 16.7° C (minimum: 6° C; maximum: 30° C) and annual rainfall of 1,073 mm, lacking a definite rainy season (Kandus and Malvárez 2004). Although population density is less than one person per hectare, the delta is a recreational area highly visited during weekends.

#### Mosquito captures

Adult mosquito surveys were conducted monthly with

human volunteers as bait and oral aspirator collections during a one-year period, from March 2003 to February 2004. On each occasion, mosquito collections occurred during the 5-30 min on each of four collection intervals as follows: 8-10:00 h, 12-14:00 h, 16-19:00 h, and 20-23:00 h. The captures during the 20-23:00 h interval were always at night; i.e., during the Summer the collections were performed after 22:00 h. Two of the authors performed all the captures, one to attract and the other to collect mosquitoes as they landed in an attempt to feed. During each collecting period, temperature was recorded with a digital thermometer. Captured mosquitoes were killed by freezing in the field and transported to the laboratory. The specimens were identified using the keys to Argentine mosquitoes (Darsie 1985) and Buenos Aires mosquitoes (Rossi et al. 2002).

#### Data analysis

In order to characterize the mosquito community that was attracted to humans, we calculated richness (S) (number of species) and Shannon diversity index (H) (richness related to the abundance for each species) for each month and season (Zar 1999). The Hutcheson t-test was used to test for Shannon index differences among seasons (Hutcheson 1970). The biting activity of each mosquito species was estimated as the number of specimens captured per hour in each collecting period and month. Williams means were calculated (Haddow 1960, Forattini et al. 1981) with the aim to describe seasonal and daily activity patterns of the most frequently collected species. Biting activity was compared statistically among seasons and hour intervals using the nonparametric Kruskal-Wallis test. For *post hoc* comparisons, the Dunn test was applied (Zar 1999).

The relationships between monthly biting activity (expressed as Williams mean) and environmental variables were evaluated by multiple linear regression analysis.

Environmental variables considered were as follows: monthly mean temperature, photoperiod, and the accumulated precipitation of the previous 15 and 30 days to the mosquito collections. Daily temperature and precipitation data were provided by National Meteorological Service and the photoperiod was calculated considering the latitude, longitude, and sample dates (Strahler and Strahler 1997). The relationships between biting activity in each collection interval and the recorded temperatures were evaluated by simple linear regression analysis.

#### RESULTS

Fourteen mosquito species assigned to six genera were collected (Table 1). The highest richness and diversity were observed in December and the lowest in August and September (Figure 1). Seasonal richness was as follows: Summer  $S = 10$ , Autumn  $S = 9$ , Spring  $S = 5$ , and Winter  $S = 4$ . The diversity in Summer ( $H = 0.54$ ) and Spring ( $H = 0.51$ ) did not differ significantly ( $p > 0.05$ ), and the values of both seasons were significantly higher ( $p < 0.01$ ) than those of Autumn ( $H = 0.37$ ) and Winter ( $H = 0.17$ ). Diversity in Autumn was also higher ( $p < 0.01$ ) than in Winter. Out of 1,289 mosquitoes captured, 95.2% of the specimens belong to genera *Ochlerotatus* and *Psorophora*. Two mosquito species (*Oc. crinifer* and *Ps. ferox*) accounted for 84.7% of the captures.

All mosquito populations showed a clear seasonal activity pattern, with greater numbers in Summer and Autumn than in Winter and Spring (Table 2a). The biting activity of the mosquito species most frequently captured (i.e., *Oc. crinifer*, *Ps. ferox*, *Oc. serratus*, and *Is. paranensis*) also differed significantly among seasons, but with different patterns (Figure 2; Table 2a). *Oc. crinifer* was captured during the four seasons and showed higher values in Summer and Autumn. *Ps. ferox* and *Oc. serratus* were not present in

Table 1. Mosquito species collected by using human volunteers in the lower delta of the Paraná River, Buenos Aires Province, from March 2003 to February 2004.

	No. of mosquitoes collected	Percentage
<i>Ochlerotatus crinifer</i>	630	48.9
<i>Psorophora ferox</i>	462	35.8
<i>Ochlerotatus serratus</i>	61	4.7
<i>Isostomyia paranensis</i>	42	3.3
<i>Psorophora cyanescens</i>	28	2.2
<i>Psorophora albipes</i>	22	1.7
<i>Psorophora albigena</i>	13	1.0
<i>Ochlerotatus scapularis</i>	11	0.8
<i>Mansonia indubitans</i>	6	0.5
<i>Mansonia titillans</i>	5	0.4
<i>Culex eduardoi</i>	4	0.3
<i>Wyeomyia leucostigma</i>	2	0.2
<i>Culex bidens/interfor</i>	2	0.2
<i>Culex maxi</i>	1	0.1
Total	1,289	100

Table 2. Statistical comparisons of biting activity of each species among seasons (a) and among species within each season (b). The numbers represent the estimated mosquitoes per hour of collection.

(a)	Median (first quartile - third quartile)				Kruskal-Wallis statistic (p)
	Autumn	Winter	Spring	Summer	
Overall	234 (93-315) <sup>a</sup>	0 (0-15) <sup>b</sup>	8 (0-38) <sup>b</sup>	384 (245-519) <sup>a</sup>	32.02 (0.0001)
<i>Oc. crinifer</i>	180 (72-268) <sup>a</sup>	0 (0-15) <sup>b</sup>	2 (0-7) <sup>b</sup>	144 (46-175) <sup>a</sup>	30.78 (0.0001)
<i>Ps. ferox</i>	0 (0-15) <sup>a</sup>	0 (0-0) <sup>a</sup>	0 (0-0) <sup>a</sup>	198 (86-307) <sup>b</sup>	31.05 (0.0001)
<i>Oc. serratus</i>	0 (0-5) <sup>a</sup>	0 (0-0) <sup>a</sup>	0 (0-0) <sup>a</sup>	24 (4-45) <sup>b</sup>	23.81 (0.0001)
<i>Is. paranensis</i>	0 (0-0) <sup>a</sup>	0 (0-0) <sup>a</sup>	0 (0-17) <sup>a</sup>	0 (0-4) <sup>a</sup>	8.02 (0.045)
(b)	<i>Oc. crinifer</i>	<i>Ps. ferox</i>	<i>Oc. serratus</i>	<i>Is. paranensis</i>	
Autumn	180 (72-268) <sup>a</sup>	0 (0-15) <sup>b</sup>	0 (0-5) <sup>b</sup>	0 (0-0) <sup>b</sup>	27.33 (0.0001)
Winter	0 (0-15) <sup>a</sup>	0 (0-0) <sup>b</sup>	0 (0-0) <sup>b</sup>	0 (0-0) <sup>b</sup>	16.33 (0.001)
Spring	2 (0-7) <sup>a</sup>	0 (0-0) <sup>a</sup>	0 (0-0) <sup>a</sup>	0 (0-17) <sup>a</sup>	7.29 (0.05 ns)
Summer	144 (46-175) <sup>ab</sup>	198 (86-307) <sup>a</sup>	24 (4-45) <sup>bc</sup>	0 (0-4) <sup>c</sup>	26.61 (0.0001)

Same letters in rows indicate no significant differences ( $p > 0.05$ ) according to Dunn's Multiple Comparison Test.

Winter and the highest values were recorded in Summer. *Is. paranensis* was the only species with higher values in Spring, although these did not differ significantly. When biting activities were compared among species within each season (Table 2b), *Oc. crinifer* showed the highest values in Autumn and Winter, and there was no significant difference among the four species in Spring. In Summer, *Ps. ferox* and *Oc. crinifer* recorded the highest biting activities.

The annual patterns of temperature, photoperiod, and precipitation are shown in Figure 3. Monthly mean temperatures were significantly associated with seasonal activity patterns of *Ps. ferox* ( $R^2 = 0.62$ ,  $F_{(1,10)} = 16.3$ ,  $p < 0.01$ ) and *Oc. serratus* populations ( $R^2 = 0.5$ ,  $F_{(1,10)} = 10$ ,  $p < 0.05$ ). *Oc. crinifer* and *Is. paranensis* were not associated with any of the variables considered.

Specific biting activities did not differ significantly among the four collection intervals when all seasons were considered together (Figure 4; Table 3a); there was not a clear daily activity pattern of the mosquito species captured. *Oc. crinifer*, *Oc. serratus*, and *Ps. ferox* were collected during the four collection intervals, but with lower values at night. *Ps. ferox* and *Oc. serratus* were recorded in the morning (8-10:00 h) only in Autumn and Summer and at night (20-23:00 h) only in Summer. No species were collected at night during Winter. Within each collection interval some statistical differences were observed among mosquito species (Table 3b). Daily biting activity of *Ps. ferox* was significantly associated with the temperatures in the morning ( $R^2 = 0.41$ ,  $F_{(1,8)} = 5.67$ ,  $p < 0.05$ ) and mid-day ( $R^2 = 0.47$ ,  $F_{(1,9)} = 7.99$ ,  $p < 0.05$ ) and also for *Oc. crinifer* during the morning ( $R^2 = 0.43$ ,  $F_{(1,8)} = 5.96$ ,  $p < 0.05$ ).

Table 3. Statistical comparisons of daily biting activity of each species among collection times (a) and among species within each collection time (b). The numbers represent the estimated mosquitoes per hour of collection.

(a)	Median (first quartile - third quartile)				Kruskal-Wallis statistic (p)
	8-10:00 h	12-14:00 h	16-19:00 h	20-23:00 h	
<i>Oc. crinifer</i>	26 (0-90) <sup>a</sup>	57 (6-197) <sup>a</sup>	51 (8-159) <sup>a</sup>	2 (0-31) <sup>a</sup>	3.84 (0.05 ns)
<i>Ps. ferox</i>	0 (0-54) <sup>a</sup>	3 (0-125) <sup>a</sup>	3 (0-75) <sup>a</sup>	0 (0-0) <sup>a</sup>	3.59 (0.05 ns)
<i>Oc. serratus</i>	0 (0-1) <sup>a</sup>	2 (0-12) <sup>a</sup>	0 (0-11) <sup>a</sup>	0 (0-0) <sup>a</sup>	4.67 (0.05 ns)
<i>Is. paranensis</i>	0 (0-2) <sup>a</sup>	0 (0-2) <sup>a</sup>	0 (0-3) <sup>a</sup>	0 (0-0) <sup>a</sup>	3.03 (0.05 ns)
(b)	<i>Oc. crinifer</i>	<i>Ps. ferox</i>	<i>Oc. serratus</i>	<i>Is. paranensis</i>	
8-10:00 h	26 (0-90) <sup>a</sup>	0 (0-54) <sup>a</sup>	0 (0-1) <sup>a</sup>	0 (0-2) <sup>a</sup>	8.06 (0.045)
12-14:00 h	57 (6-197) <sup>a</sup>	3 (0-125) <sup>ab</sup>	2 (0-12) <sup>ab</sup>	0 (0-2) <sup>b</sup>	10.11 (0.018)
16-19:00 h	51 (8-159) <sup>a</sup>	3 (0-75) <sup>ab</sup>	0 (0-11) <sup>b</sup>	0 (0-3) <sup>b</sup>	10.91 (0.012)
20-23:00 h	2 (0-31) <sup>a</sup>	0 (0-0) <sup>ab</sup>	0 (0-0) <sup>ab</sup>	0 (0-0) <sup>b</sup>	9.177 (0.027)

Same letters in rows indicate no significant differences ( $p > 0.05$ ) according to Dunn's Multiple Comparison Test.

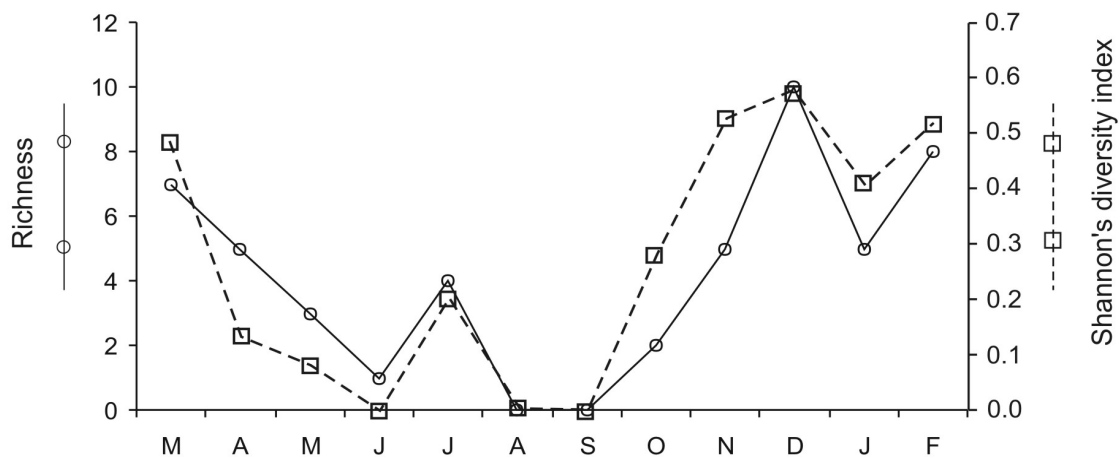


Figure 1. Monthly richness and diversities (Shannon Index) of mosquito fauna collected in the lower delta of the Paraná River, Buenos Aires Province, from March 2003 to February 2004.

## DISCUSSION

Among 74 mosquito species registered in Buenos Aires Province (Rossi et al. 2006), we collected 14 species in the lower delta of the Paraná River. Near our study area, García and Casal (1965) found the same number of human-attracted species, although only six matched with our records (*Oc. crinifer*, *Oc. scapularis*, *Ps. cyanescens*, *Ps. ferox*, *Mansonia indubitans*, and *Ma. titillans*). This richness was lower than those recorded in human-landing collections in tropical areas of South America; e.g., 57 species in Peru (Jones et al. 2004) and 28–57 species in Brazil (Guimarães et al. 2000b, 2001). This difference could probably be due to climatic and environmental differences between tropical and temperate areas, but may also be due to the low collection effort involved in our study. Several of the species found in our survey were frequently captured in human-landing collections in South America (e.g., Hack et al. 1978, Guimarães et al. 2000a,b, Jones et al. 2004) and are recognized vectors of arboviruses (Souza Lopes et al. 1981, Mitchell et al. 1986, Kulasekera et al. 2001, Forattini 2002, Turell et al. 2005).

Seasonal biting activity probably reflects seasonal abundance, but more importantly, it provides an accurate estimation of the timing of risk for human populations. Monthly mosquito captures showed a clear seasonal pattern with higher values in Summer and Autumn than in Winter and Spring. Only *Oc. crinifer* was collected during the four seasons, from October to July, and with higher values in Summer and Autumn. Previous surveys performed in Buenos Aires Province using human bait (García and Casal 1965) or CDC light traps (Ronderos et al. 1992, Maciá et al. 1995, Maciá 1996, 1997) also found host-seeking adults of this species during the four seasons. *Ps. ferox* and *Oc. serratus* were not collected during the cold months, consistently with previous findings in the area (Ronderos et al. 1992, Campos

et al. 1995, Maciá 1996, 1997). Finally, *Is. paranensis* was recorded from October to April and was the only species with higher captures in Spring. Ronderos et al. (1992) collected adults of this species from December to June in Buenos Aires Province. This discrepancy could be due to temperature differences during the years involved in both surveys or due to the differences in the availability of the phytotelmata habitats where they breed, mainly represented by *Eryngium* sp. and *Typha* sp. in the study area.

Among the environmental variables considered, only temperature was associated with seasonal activity patterns of *Ps. ferox* and *Oc. serratus*. We did not find an association between temperature and the seasonal activity of *Oc. crinifer* and *Is. paranensis*, but that might be due to these species being more active at moderate temperatures rather than at either warm or cool ones; i.e., mosquito activity may be strongly related to temperature, but not linearly. In Buenos Aires Province, the host-seeking activities of *Ps. ferox* and *Oc. crinifer* populations were previously associated with temperature and not with precipitation (Balseiro 1981, 1989). On the other hand, Campos et al. (1995) observed a decrease of *Ps. ferox* activity when temperatures reached the highest values. *Oc. crinifer*, *Ps. ferox*, and *Oc. serratus* are flood water mosquitoes, therefore, their abundance depends on the water dynamics of temporary pools. However, we did not find an association between mosquito activity and precipitation. Lunar and wind tides may regulate temporary ponds in the study area, but this variable was not evaluated in the present survey.

The daily biting activities of the main species involved in our study are poorly known in Argentina. Previous papers suggest that *Ps. ferox* females are active during the whole day but with diurnal preference (Hack et al. 1978, Balseiro 1989). With regard to *Oc. crinifer*, García and Casal (1965) and Balseiro (1981, 1989) found a nocturnal preference, whereas Hack et al. (1978) collected females

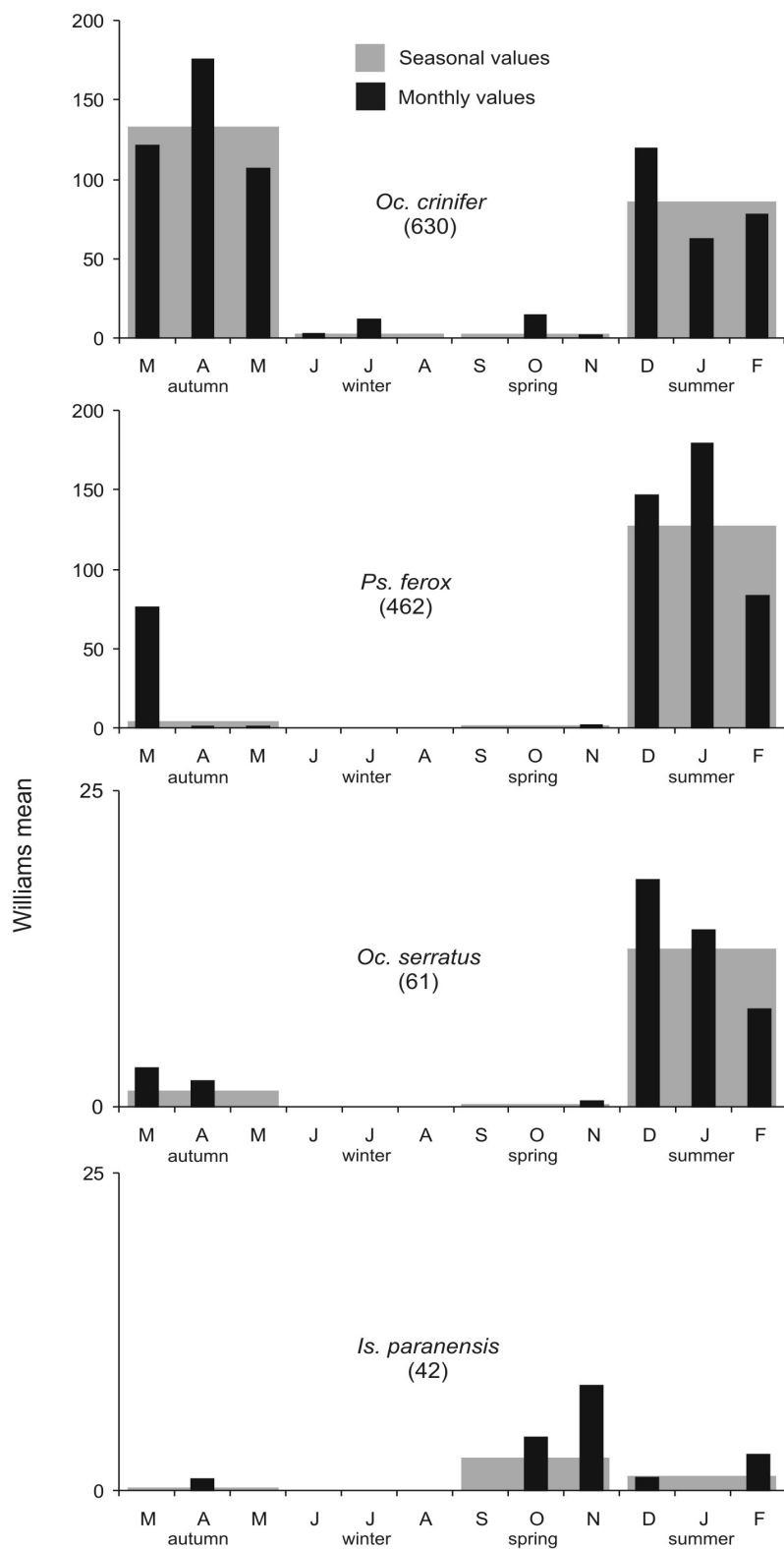


Figure 2. Monthly and seasonal biting activity of mosquito species most frequently captured in the lower delta of the Paraná River, Buenos Aires Province, from March 2003 to February 2004. Total numbers of mosquitoes collected per species within parentheses.

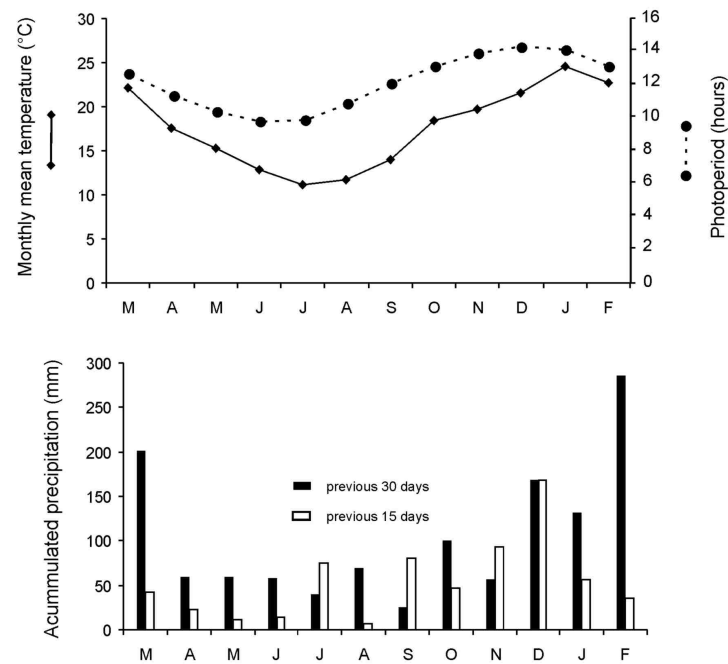


Figure 3. Monthly values of the environmental variables evaluated.

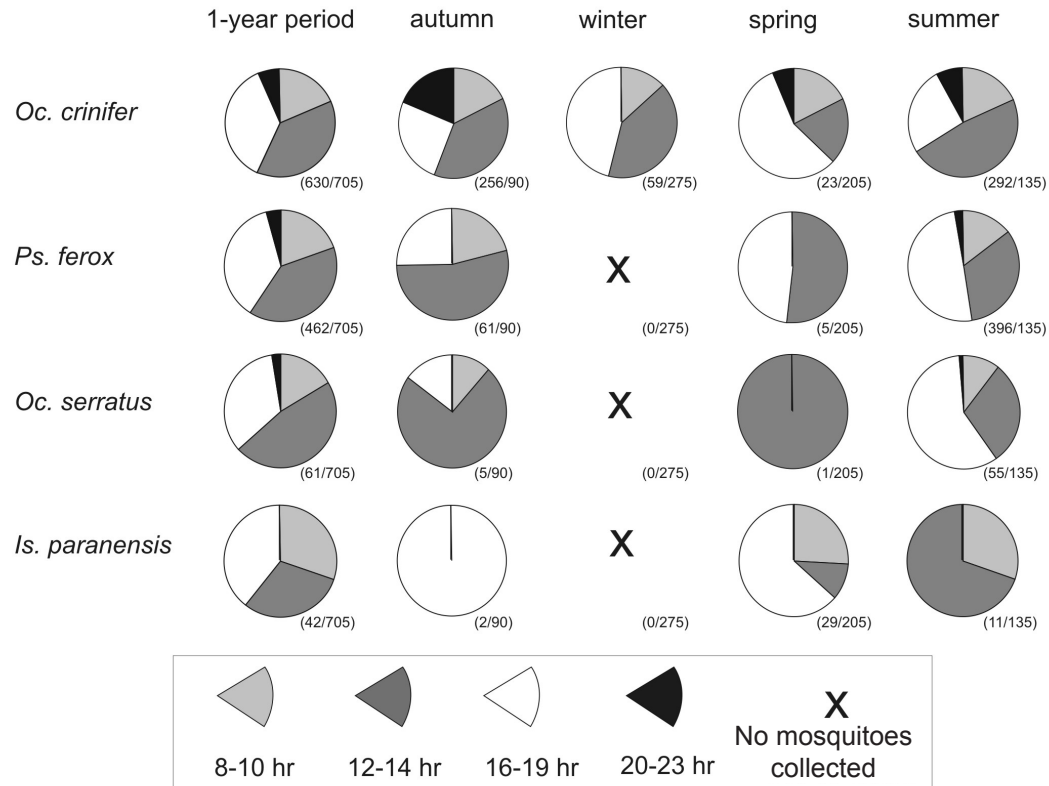


Figure 4. Daily biting activity (expressed as Williams means) of mosquito species most frequently captured in the lower delta of the Paraná River, Buenos Aires Province, from March 2003 to February 2004. Total number of mosquitoes collected / minutes of collection within parentheses.



exclusively during the day, although only a few specimens were captured in the latter. We captured both species in the four collection intervals with a diurnal preference, although it was not statistically significant. Our main conclusion on this issue is that daily activity patterns change throughout the seasons and, in general, the nocturnal captures were during the warm months.

Our study involved a relatively low number of mosquitoes collected. This could bias some results, as for example, the absence of some species in Winter and of any species during winter nights. Also, considering that some mosquito species could take several minutes to orient and approach humans, the observed values could be underestimated in collections of short duration (e.g., 5 min). However, our study determined which mosquito species actually come to humans in the study area, in contrast to studies that use carbon dioxide-baited light traps. This information could be useful for risk assessment in exposed human populations.

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