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DAYTIME RESTING ACTIVITY OF *AEDES AEGYPTI* AND *CULEX QUINQUEFASCIATUS* POPULATIONS IN NORTHERN MEXICO

LIHUA WEI,[1] NADIA A. FERNÁNDEZ-SANTOS,[1,2] GABRIEL L. HAMER,[2] EDGAR E. LARA-RAMÍREZ[1] AND MARIO A. RODRÍGUEZ-PÉREZ[1,3]

ABSTRACT. *Aedes aegypti* and *Culex quinquefasciatus* are disease vectors distributed throughout much of the world and are responsible for a high burden of vector-borne disease, which has increased during the last 2 decades. Most pathogens vectored by these mosquitoes do not have therapeutic remedies; thus, combating these diseases is dependent upon vector control. Improvements in vector control strategies are urgently needed, but these hinge on understanding the biology and ecology of *Ae. aegypti* and *Cx. quinquefasciatus*. Both species have been extensively investigated, but further knowledge on diel resting activity of these vectors can improve vector surveillance and control tools for targeting resting vector populations. From April to December 2021, we determined outdoor daytime resting habits of *Ae. aegypti* and *Cx. quinquefasciatus* male, female, and blood-fed female populations in Reynosa, Mexico, using large red odor-baited wooden box traps. The daytime resting activity for *Ae. aegypti* males, females, and blood-fed females was restricted to a period between 0900 h and 1300 h, with a peak at 0900 h, while the resting activity of *Cx. quinquefasciatus* male, female, and blood-fed females was between 0700 h and 1100 h, with a peak at 0700 h. A generalized additive model was developed to relate relative humidity and temperature to resting *Cx. quinquefasciatus* and *Ae. aegypti* male, female, and blood-fed populations caught in traps. This study advances the understanding of outdoor resting behavior for 2 important vector mosquito species and discusses future studies to fill additional knowledge gaps.

KEY WORDS *Aedes aegypti*, *Culex quinquefasciatus*, daytime behavior, red box traps, resting behavior

INTRODUCTION

Mosquito-borne diseases are a major public health concern causing millions of deaths annually (Chaiphongpachara et al. 2018). *Aedes aegypti* (L.) is the most important global vector of arboviruses, such as dengue, chikungunya, Zika, and yellow fever (Campos et al. 2020). Although *Ae. albopictus* (Skuse) has been reported in Mexico (Dávalos-Becerril et al. 2019), its role as an arbovirus vector is limited (Lambrechts et al. 2010, Vasilakis et al. 2011, Mousson et al. 2012). Members of the *Culex quinquefasciatus* Say complex are the primary vectors of West Nile virus (WNV) throughout much of the tropical and temperate regions of the world (del Carpio-Orantes et al. 2018). In addition, they are vectors of St Louis encephalitis virus (SLEV) (Diaz et al. 2013), Usutu virus (Clé et al. 2019), and the filarial nematode *Wuchereria bancrofti* (Cobbold) (Ant et al. 2020).

The control of mosquito-borne viruses in Mexico and other countries receives considerable resources but returning limited success as severe outbreaks persist in tropical endemic countries worldwide including nonendemic countries such as chikungunya in Italy during 2007 and dengue in France and Croatia during 2010 (Gould et al. 2010, Gjenero-Margan et al. 2011, Poletti et al. 2011, Lwande et al. 2020). In 2020 the Mexican Ministry of Health reported 24,313 cases of dengue, 2,281 of which occurred in Tamaulipas. From 2015 to November 2022, 13,003 confirmed cases of Zika were reported in Mexico, of which 802 cases occurred in Tamaulipas (Mexico Ministry of Health 2022). The persistent transmission is in part, due to a lack of effective vaccines and development of insecticide resistance in vectors populations. Thus, effective vector control needs to be built on detailed knowledge of mosquito population ecology and behavior that has spatio-temporal heterogeneity (Wilke et al. 2019).

Many studies have addressed the resting behavior of *Ae. aegypti* and *Cx. quinquefasciatus*. However, most of them were focused on the indoor or outdoor resting sites (Hecht and Hernandez-Corzo 1963, Schoof 1967, Kay 1983, Kittayapong et al. 1997, Perich et al. 2000, Chadee 2013, Dzul-Manzanilla et al. 2017, Diallo and Diallo 2020, Machani et al. 2020, Diouf et al. 2021, Dalpadado et al. 2022, Janaki et al. 2022), resting behavior associated with residual insecticides (Bøgh et al. 1998, Cooperband and Allan 2009, Manda et al. 2011), and insecticide treatment (Pates and Curtis 2005, Tainchum et al. 2013). Very few studies have looked at the peak daytime resting activity of *Ae. aegypti* and *Cx. quinquefasciatus* male and female populations. To our knowledge, the only study about the diel resting activity of mosquitoes was by Gjullin et al. (1963) in California. There mosquitoes moved out of resting boxes around sunset and returned around sunrise.

[1] Instituto Politécnico Nacional, Centro de Biotecnología Genómica, Cd. Reynosa 88710, Tamaulipas, Mexico.
[2] Texas A&M University, College Station, Texas 77843-2475.
[3] To whom correspondence should be addressed.

Mosquito traps baited with color and odor are widely used to collect or control mosquito populations. The red box trap for resting mosquitoes has been used for *Ae. aegypti* (Edman et al. 1997, Kittayapong et al. 1997), which are known to select resting sites based on visual cues such as color (Manda et al. 2011). *Aedes aegypti* is primarily anthropophilic in host utilization, while *Cx. quinquefasciatus* is primarily ornithophilic, respectively; however, in Reynosa, Mexico, and the Rio Grande valley of Texas, both species were reported to feed more frequently on dogs than on humans (Olson et al. 2020). This is probably due to a high availability of domestic dogs in many low- to medium-income communities in this region.

Here odor-baited wooden box traps painted red were used to study the daytime resting activity of outdoor of *Ae. aegypti* and *Cx. quinquefasciatus* male and female populations. A model was also developed to relate relative humidity (RH) and temperature to each vector resting population. This protocol can help guide the monitoring of *Ae. aegypti* and *Cx. quinquefasciatus* resting populations for further studies such as those reported elsewhere (Estrada-Franco et al. 2020) and can greatly improve the effectiveness of mosquito control programs.

MATERIALS AND METHODS

Study site: This study was carried out from April to December 2021 on the Instituto Politecnico Nacional–Centro de Biotecnologia Genomica campus (IPN-CBG) located in Reynosa, Tamaulipas, northern Mexico (26°4′10.28″N, 98°18′48.53″W; Fig. 1A–C). Reynosa is an industrialized city, which is endemic to dengue in a semiarid climate with 2 marked annual seasons. Winters are short, mild (temperature between 11 and 22°C), and dry, lasting 3 months, from December to February. Summers extend from May to August and very hot with temperatures often exceeding 40°C. Reynosa is 33 m above sea level and has an annual rainfall of 452 mm. Resting traps were placed under trees where many birds roosted and people rarely traversed. The east and south of the campus are adjacent to residential areas, and most of them have dogs or cats at home (Fig. 1D).

Mosquito collection: One wooden box trap painted red on the interior walls (Fig. 2) with a size of 120 cm × 90 cm × 90 cm was placed in 3 collection points around the IPN-CBG campus (Fig. 1D). Three traps were placed in the shade of trees on the institute campus. One Centers for Disease Control and Prevention autocidal gravid ovitrap (AGO) with hay infusion made by steeping 30 g of hay packet in 10 l of tap water for 7 days (Barrera et al. 2014) was placed inside the red wooden box to use as bait. On the top, AGO traps have a screen barrier to prevent any mosquitoes from emerging inside (Barrera et al. 2014). Mosquitoes were collected from April 21 to December 15, 2021, 11 days per month on average, every 2 hours from 0700 h to 1900 h, using Improved Prockopack Aspirators (John W. Hock Co., Gainesville, Florida). Time intervals are chosen to allow for collection and identification. Upon collection, mosquitoes were anesthetized in a –20°C freezer for 5 min, then transferred to a Petri dish and put on a chill table (1429 Chill Table, BioQuip, Rancho Dominguez, CA, USA). Identification was carried out using morphological characters as described in the taxonomic keys by Burkett-Cadena (2013). Upon identification, the number of males, females, and blood-fed females of each species was recorded. After recording, mosquitoes were recovered at room temperature for 30 min before releasing them in a central location on the campus, at an average of 88 mosquitoes from 3 resting boxes (Fig. 1D). Temperature and RH reading were recorded, using an Xiaomi sensor (Mi temperature and humidity monitor 2, Xiaomi, Beijing, China) placed at one of the 3 red wooden box traps.

Statistical analysis: Descriptive statistics based on raw counts for the 107 days of data collection of *Ae. aegypti* and *Cx. quinquefasciatus* males, females, and blood-fed females were carried out using the R with ggplot2 package. Data were inspected graphically through boxplots that revealed dispersion by means of interquartile range (IQR).

A generalized additive model (GAM) was used to relate temperature and RH to ln(total number + 1) of *Ae. aegypti* and *Cx. quinquefasciatus* males, females, and blood-fed females caught hourly at each sampling point using an identity link function with Gaussian distribution. Generalized additive models have been successfully used to explore nonlinear relationship between environmental variables and mosquito populations (Xu et al. 2017). The model equation used was as follows:

$$\ln(Y_t) = \beta + s(C_t) + \varepsilon_t,$$

where (Y_t) is the number of mosquitoes collected each hour t, C is the environmental variable, β is the intercept, s(·) denotes a spline function to avoid overfitting, and ε are the residuals (Liu et al. 2020).

RESULTS

The total counts of *Ae. aegypti* and *Cx. quinquefasciatus* using 3 red wooden box traps at 7 daytime intervals (0700, 0900, 1100, 1300, 1500, 1700, and 1900) are shown in Table 1. A total of 5,327 *Ae. aegypti*, 6859 *Cx. quinquefasciatus*, 108 *Ae. albopictus*, and fewer than 10 unidentified mosquito specimens were collected. Of all the *Ae. aegypti* sampled, 84.3% were males; of all the female *Ae. aegypti* sampled, 12.3% were blood-fed females. Of the 6859 *Cx. quinquefasciatus* sampled, 50.0% were males; and of all the female *Cx. quinquefasciatus* sampled, 49.2% were blood-fed.

The daytime resting activity of *Ae. aegypti* and *Cx. quinquefasciatus* males, females, and blood-fed

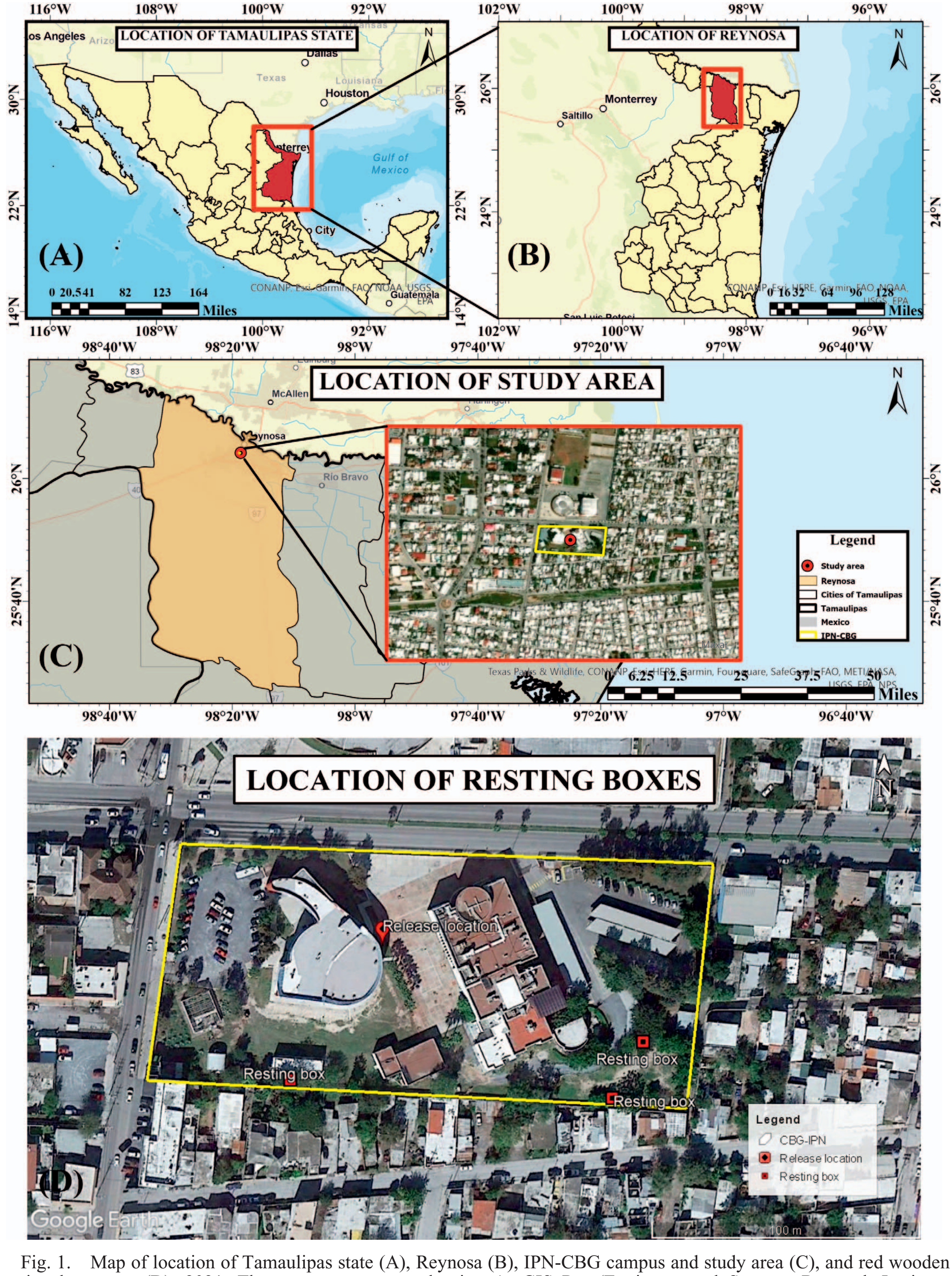

Fig. 1. Map of location of Tamaulipas state (A), Reynosa (B), IPN-CBG campus and study area (C), and red wooden resting box traps (D), 2021. The map was generated using ArcGIS Pro (Environmental Systems Research Institute, Redlands, CA, USA) and Google Earth Pro (Google, USA). Free geographic (GIS) data of administrative areas of Mexico was downloaded from DIVA-GIS (https://www.diva-gis.org/gdata).

Fig. 2. Wooden resting box painted red on the inside used for collecting mosquitoes in Reynosa, Mexico, from April to December 2021.

females are shown in Fig. 3. *Aedes aegypti* males' resting activity peaked at 0900 h, with a secondary peak at 1100 h; the lowest collection hour was at 1900 (Fig. 3A). For *Ae. aegypti* females and blood-fed females, almost equal numbers of mosquitoes were obtained between 0900 h and 1100 h. The lowest collection hour of *Ae. aegypti* females was at 1900 h, and the lowest collection hour of *Ae. aegypti* blood-fed females was at 1500 h.

For *Cx. quinquefasciatus* (Fig. 3B), the highest peak collection hour for males, females, and blood-fed females was 0700 h, and the lowest peak collection hour was at 1700 h. At peak resting (0900 h) for *Ae. aegypti* males, females, and blood-fed females, the average temperature and RH were 25.4 ± 0.34°C and 84.1 ± 0.61%, respectively. At peak resting (0700 h) for *Cx. quinquefasciatus* males, females, and blood-fed females, the average temperature and RH were 24.2 ± 0.31°C and 83.3 ± 0.63%, respectively.

The association between temperature and RH and *Ae. aegypti* male, female, and blood-fed female mosquitoes analyzed with GAM is shown in Table 2 and Fig. 4. *Aedes aegypti* male, female, and blood-fed female mosquitoes had weak positive correlation with humidity (adjusted $R^2 = 0.0712$, $P < 0.001$; adjusted $R^2 = 0.0301$, $P < 0.001$; adjusted $R^2 = 0.0162$, $P < 0.001$, respectively) and temperature (adjusted $R^2 = 0.122$, $P < 0.001$; adjusted $R^2 = 0.0389$, $P < 0.001$; adjusted $R^2 = 0.0186$, $P < 0.05$, respectively).

The association between temperature and RH and *Cx. quinquefasciatus* male, female, and blood-fed female mosquitoes analyzed with GAM is depicted in Table 2 and Fig. 5. *Culex quinquefasciatus* males, female, and blood-fed females had a positive association with RH (adjusted $R^2 = 0.406$, $P < 0.001$; adjusted $R^2 = 0.441$, $P < 0.001$; adjusted $R^2 = 0.347$, $P < 0.001$, respectively), and a negative association with temperature (adjusted $R^2 = 0.363$, $P < 0.001$; adjusted $R^2 = 0.325$, $P < 0.001$; adjusted $R^2 = 0.247$, $P < 0.001$, respectively). Increasing number of *Cx. quinquefasciatus* were observed when RH ranged from 25% to 84%; however, the mosquito numbers started to decrease as RH exceeded 84%. Therefore, RH recorded during our mosquito sampling had an effect on the number of *Cx. quinquefasciatus* caught daily in the red wooden box traps in Reynosa, Mexico. We also observed increasing numbers of *Cx. quinquefasciatus* when the temperature ranged from 22 to 26°C; however, mosquito numbers dropped sharply when the temperature went over 26°C.

DISCUSSION

We present the daytime resting activity of *Ae. aegypti* and *Cx. quinquefasciatus* males, females, and blood-fed females and perform a GAM to relate RH and temperature to these 2 species of mosquitoes in Reynosa, Mexico. Temporally, males, females, and blood-fed females of both species, *Ae. agypti* and *Cx. quinquefasciatus*, preferred resting in the red wooden box traps at 0900 h and 0700 h, respectively. However, no large numbers of blood-fed *Ae. aegypti* females were collected at any hour point of the day. We suspect 1) the odor in traps was not attractive for *Ae. aegypti* females, 2) the vertebrate host utilization and flight range of *Ae. aegypti* were different as compared with *Cx. quinquefasciatus*, and 3) the location of traps was farther from *Ae. aegypti* hosts compared to *Cx. quinquefasciatus* hosts. In this study

Table 1. Total collection by hour for *Aedes aegypti* and *Culex quinquefasciatus* from wooden resting boxes in Reynosa, Mexico from April to December 2021.[1]

Time (h)	*Ae. aegypti*				*Cx. quinquefasciatus*				Mean temp. °C ± SE	Mean RH % ± SE
	Total	Males	Females	Blood-fed	Total	Males	Females	Blood-fed		
0700	414	327	87	18	4,757	2,536	2,221	1,113	24.2 ± 0.31	83.3 ± 0.63
0900	1,303	1,134	169	22	1,185	523	662	291	25.4 ± 0.34	84.1 ± 0.61
1100	1,545	1,343	202	26	338	148	190	94	29.0 ± 0.34	72.7 ± 0.67
1300	1,012	871	140	15	184	64	120	65	30.9 ± 0.34	64.5 ± 0.78
1500	493	384	109	8	127	54	73	39	32.1 ± 0.35	60.0 ± 0.92
1700	351	278	74	6	92	41	51	16	31.4 ± 0.36	61.4 ± 0.11
1900	209	152	57	8	176	62	114	70	29.5 ± 0.40	67.2 ± 0.11

[1] Temp.: temperature, RH: relative humidity, SE: standard error.

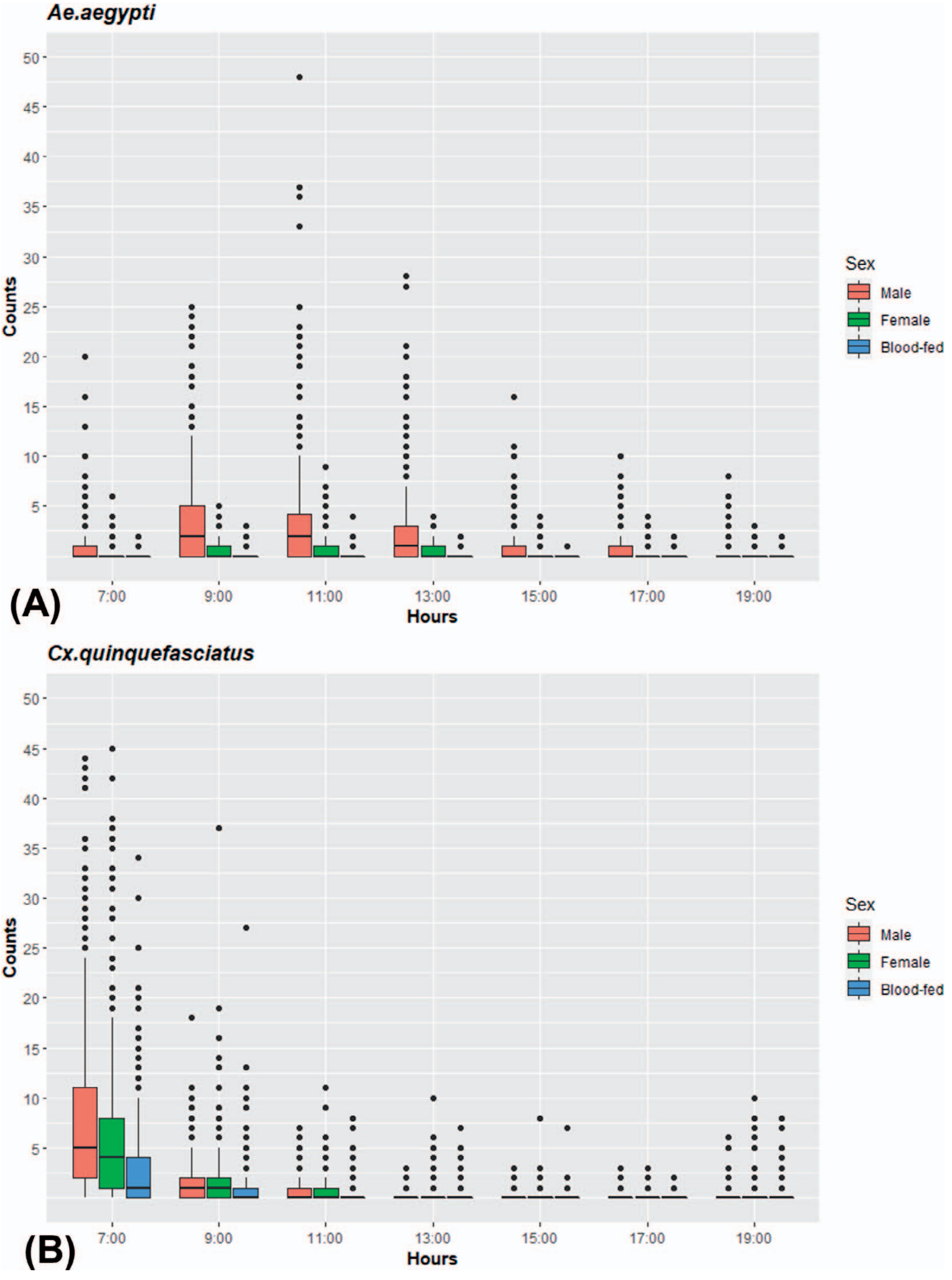

Fig. 3. Daytime resting pattern of male, female, and blood-fed female *Aedes aegypti* (A) and *Culex quinquefasciatus* (B) in Reynosa, Mexico, from April to December 2021. The upper and the lower line of each plot box shows the IQR, which is a measure of statistical dispersion. The black points outside the box show the outliers, and the black line inside each box is the median of each collection of each hour.

Table 2. The influence of climatic factors (temperature and relative humidity) on the numbers of *Aedes aegypti* and *Culex quinquefasciatus* males, females and blood-fed females collected at hour intervals in wooden resting boxes in Reynosa, Mexico, from April to December 2021, using a GAM univariate analysis.[1]

Sample	Smoothing effect	Est. df	Ref. df	*F*-value	*P*-value	Adj. R^2	Deviance (%)
Cx. quinquefasciatus male	S(Temperature)	6.865	7.939	53.67	<2e-16 ***	0.363	36.8
	S(RH)	7.54	8.371	61.3	<2e-16 ***	0.406	41.2
Cx. quinquefasciatus female	S(Temperature)	7.424	8.371	43.24	<2e-16 ***	0.325	33.2
	S(RH)	7.308	8.206	72	<2e-16 ***	0.441	44.6
Cx. quinquefasciatus blood-fed	S(Temperature)	7.447	8.387	29.4	<2e-16 ***	0.247	25.5
	S(RH)	7.06	8.017	49.78	<2e-16 ***	0.347	35.3
Ae. aegypti male	S(Temperature)	4.728	5.797	18.16	<2e-16 ***	0.122	12.8
	S(RH)	2.954	3.751	15.37	<2e-16 ***	0.0712	7.48
Ae. aegypti female	S(Temperature)	3.419	4.277	7.308	6.66e-06 ***	0.0389	4.33
	S(RH)	1.001	1.002	24.16	1.46e-06 ***	0.0301	3.14
Ae. aegypti blood-fed	S(Temperature)	3.08	3.868	3.961	0.00466**	0.0186	2.26
	S(RH)	1	1	13.28	0.000287***	0.0162	1.75

[1] *** $P < 0.001$, ** $P < 0.01$ level, Adj. R^2: adjusted R square, Est. df: estimated degrees of freedom, Est. df >1 indicates nonlinear relationships, Ref. df: reference degrees of freedom.

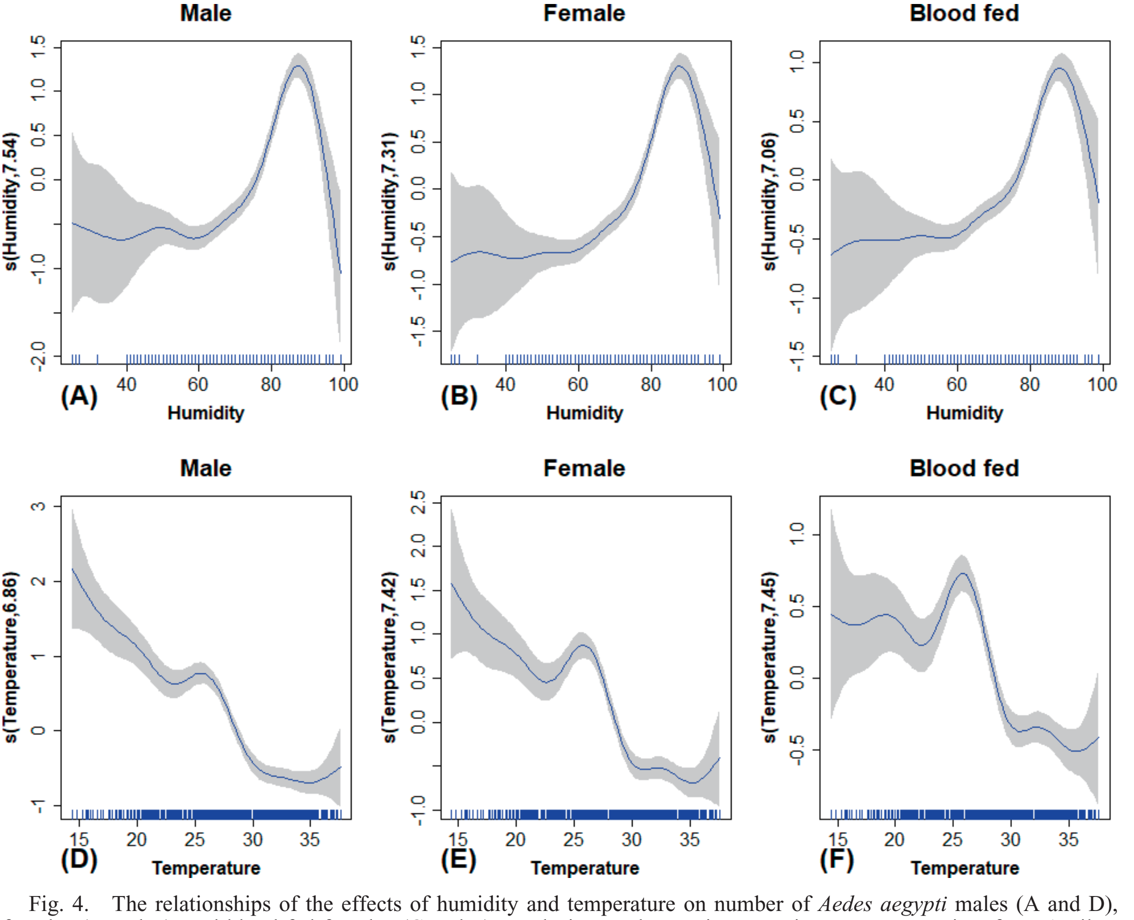

Fig. 4. The relationships of the effects of humidity and temperature on number of *Aedes aegypti* males (A and D), females (B and E), and blood-fed females (C and F) caught in wooden resting traps in Reynosa, Mexico, from April to December 2021. The x-axis is the collected meteorological data, and the y-axis indicates the smoothing components for counts of mosquitos to the fitted values. The gray area represents the 95% confidence interval. This analysis was carried out in R (version 4.2.2) with the mgcv package.

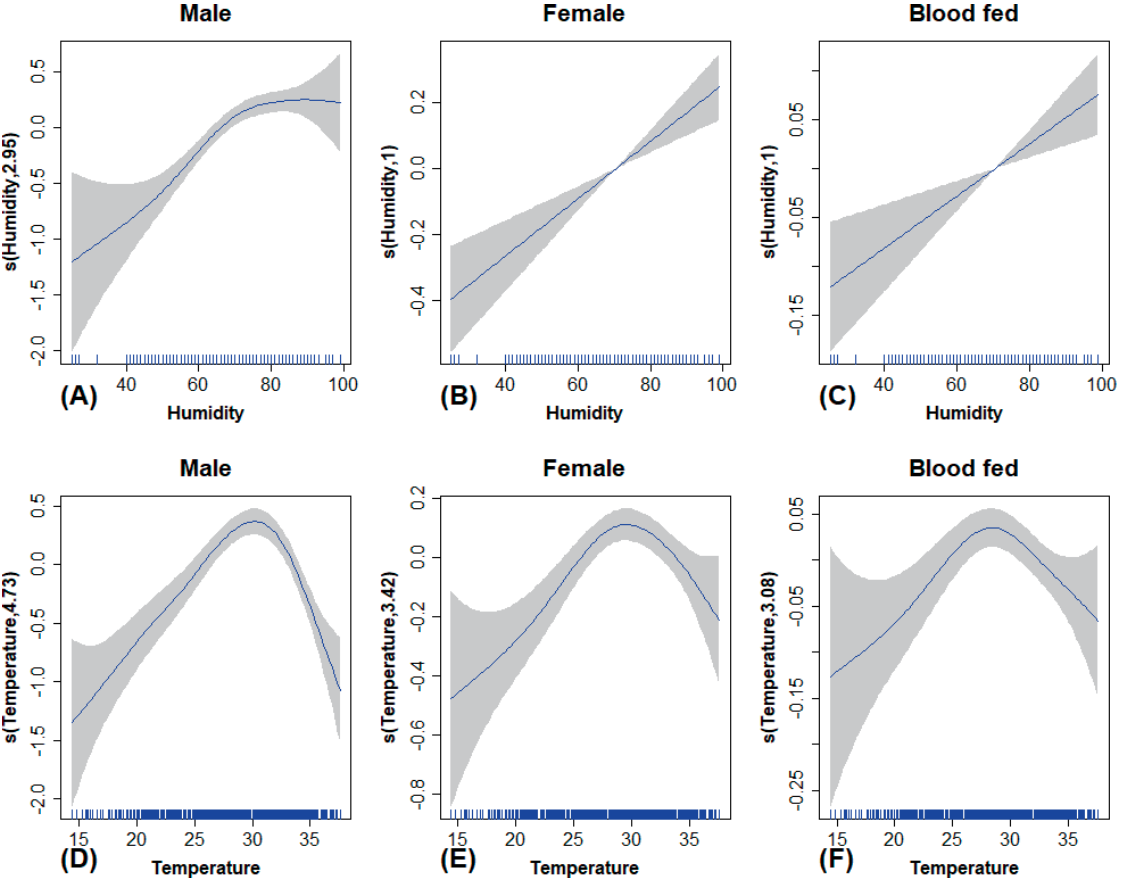

Fig. 5. The relationships of the effects of humidity and temperature on number of *Culex quinquefasciatus* males (A and D), females (B and E), and blood-fed females (C and F) caught in wooden resting traps in Reynosa, Mexico, from April to December 2021. The x-axis is the collected meteorological data, and the y-axis indicates the smoothing components for counts of mosquitos to the fitted values. The gray area represents the 95% confidence interval. This analysis was carried out in R (version 4.2.2) with the mgcv package.

we used hay infusions as odor attractants. Hay infusion has been shown to attract gravid *Ae. aegypti* females (Ponnusamy et al. 2010), which did not agree with our result. It is possible that the ratio of hay to water and age of the infusion water could vary in composition and volatiles among the studies. Dormont et al. (2021) reviewed mosquito attractants and noted various plant odors attractive to *Ae. aegypti*, which could be tested with our resting box trap in the future. *Aedes aegypti* females generally fly 100–500 m (McDonald 1977, Trpis and Hausermann 1986, Juarez et al. 2020). The mean distance traveled per day by female *Ae. aegypti* was only 16.8 m and 24.7 m for indoor and outdoor release, respectively, and the maximum overall distance per day was 160 m (Muir and Kay 1998). For *Cx. quinquefasciatus*, we found a large number of males, females, and blood-fed females resting inside the traps. This can be explained as *Cx. quinquefasciatus*, a member of the *Cx. pipiens* complex, has a maximum flight range from <1.0 to 2.1 km, with a mean distance between 1.27 km and 1.64 km in 36 h (Fussell 1964, Lindquist et al. 1967, Schreiber et al. 1988, Lapointe 2008, Medeiros et al. 2017). Studies investigating the feeding patterns of *Cx. quinquefasciatus* found that they fed primarily on birds, while in many areas of the world, while *Ae. aegypti* is considered anthropophilic, feeding mostly on humans (Sallam et al. 2017, Telang and Skinner 2019, Estrada-Franco et al. 2020, Fikrig and Harrington 2021). Our resting boxes were placed near the trees, which could have been locations of roosting birds. After feeding, *Ae. aegypti* are endophilic and mainly rest inside human dwellings (Diallo and Diallo 2020). A study in Panama found that a low percentage of resting *Ae. aegypti* (24.7%) was found outdoors, while a higher percentage of resting *Ae. aegypti* (75.1%) was found indoors. Janaki et al. (2022) found a higher proportion of resting *Ae. aegypti* females was caught indoors (88%) than outdoors (12%).

Casas Martinez et al. (2013) monitored the daily activity of *Ae. aegypti* males and females using their newly designed BioDiVector tent traps in Tapachula, Chiapas, Mexico. They found that *Ae. aegypti* showed 2 peaks of activity (morning and afternoon) at extra-domicile sites. *Aedes aegypti* female activity was higher between 0600 and 1000 h in the morning and 1600 to 1800 h in the afternoon. The activity of *Ae. aegypti* males was higher between 0800 and 1000 h in the morning and 1600 to 1800 h in the afternoon. Captain-Esoah et al. (2020) reported host-seeking *Ae. aegypti* in northern Ghana with bimodal peaks between 0600 to 0800 and 1500 to 1600 h. These results do not completely correspond to our resting peak results, which may be due to the different type of traps used, hour of collection or geographic features, and environmental variations. Mutebi et al. (2022) conducted a study about the diel activity patterns of host-seeking *Ae. aegypti* females in Brownsville, Texas. They found *Ae. aegypti* females had 2 peaks of diel biting activities, between 0700 and 0800 h and between 1900 and 2000 h. In our study the resting period of *Ae. aegypti* male, female, and blood-fed females was between 0900 and 1300 h. These corroborative results of *Ae. aegypti* populations on both sides of the USA-Mexico border suggest that individuals seek resting habitat later in the morning following a peak in host-seeking activity.

Gjullin et al. (1963) conducted a study about the daily resting cycles of *Culex* species using red box shelter units and found that the daily resting period of *Cx. pipiens* and *Culex tarsalis* extended from 0800 h to sunset, leaving the box after sunset and returning before sunrise, as *Culex* species are often crepuscular in host-seeking behavior. This result is in accordance with our findings as we have noted a unimodal clear peak at 0700 h; it is at that time in the morning when we captured *Culex* species that were done host-seeking and then ready to rest for the rest of the day. To investigate whether mosquitoes rested at night in our red wooden box traps, we inspected the traps during 3 nights from 1900 to 0500 h (data not shown), and no resting mosquitoes were found as reported elsewhere (Zuharah and Sumayyah 2019). As previously mentioned, *Cx. quinquefasciatus* may rest close to hosts at night given their crepuscular feeding behavior. Karlekar and Andrew (2016) found that *Cx. quinquefasciatus* were feeding at dusk (1800 to 2000 h) and resting on the roof of cattle sheds and grain storage, while during the daytime, they preferred resting near water tanks and water puddles.

This GAM analysis helps to explain daytime resting behavior of *Ae. aegypti* and *Cx. quinquefasciatus*. Some discrepancies in temperature and RH were observed between *Ae. aegypti* and *Cx. quinquefasciatus*. For *Cx. quinquefasciatus*, temperature and RH were associated with their resting behavior but not with the resting pattern of *Ae. aegypti*. The GAMs have been used to indicate the distribution and abundance of mosquito larvae in residential areas (Heersink et al. 2016), to evaluate different mosquito collection methods (Câmara et al. 2022), and to analyze the relationship between climate variables with mosquito density (Xu et al. 2017). To our knowledge, no studies have used GAMs to explain the relationship between environmental factors and mosquito daytime resting patterns. However, due to the univariate model construction, this model cannot fully explain the relationship between the environmental factors and the resting behavior of those mosquito species. Adding more variables could better refine the model for interpretation.

Here the resting pattern for the 2 species of mosquitoes peaked at high RH recorded during the day. However, high temperature was less related to mosquito captures. More *Cx. pipiens* and *Ae. aegypti* preferred to stay in the cooler and more humid zone of the cage during the night and day, respectively (Kessler and Guerin 2008), and *Cx. quinquefasciatus* preferred to rest in cool, humid, shady areas such as water tanks, shade near puddles, and trees (Rios et al. 2006, Karlekar and Andrew 2016). In correspondence, we did not observe more mosquitoes at 1300, 1500, and 1700 h, where the temperature was higher than that of 0700 and 1100 h. We suspect mosquitoes were resting more during the highest RH, and for *Culex*, the highest resting counts were first thing in the morning, which was when the humidity was the highest. The time of the day (Fig. 2) shows that male and female *Ae. aegypti* arrived to resting habitat over the course of the morning. However, for *Culex* it is more complicated to interpret since they were already utilizing resting habitat during the first sampling period, and we are not sure how many actually arrived at the first sampling period (0700 h) versus how many arrived before (1900 to 0500 h). We also noted strong day-to-day mosquito variations indicating that other environmental factors influence shelter-seeking behavior, such as wind speed, cloud cover, and rain, which we did not consider in our analysis.

A limitation of this current study was that all wooden box traps were placed on a university campus away from residential homes, which means the results could be influenced by different proximity to larval and host-seeking habitat. We also collected very few *Ae. albopictus* and other mosquito species given that the traps were at the same collection site. Second, all traps are placed on the ground and placing resting habitat in tree canopies could have influenced the results, similar to how traps collecting host-seeking mosquitoes were different at ground level versus forest canopy (Andreadis and Armstrong 2007, Pereira-Silva et al. 2021). Third, we used hay infusion AGO traps to augment the resting box habitat. This odor and the increased humidity from the water might have attracted oviposition seeking females. We did not distinguish gravid from unfed female mosquitoes, which could have been done to distinguish the 2 behaviors. Fourth, additional environmental variables should be recorded to investigate additional abiotic and biotic factors that influence mosquito resting behavior.

The red wooden odor-baited box traps can be used efficiently for monitoring mosquitoes as other traps do for mosquito surveillance such as gravid traps (Reiter et al. 1986), light traps (Sudia and Chamberlain 1962), BG sentinel traps (Maciel-de-Freitas et al. 2006), and oviposition traps (Barbosa et al. 2007). This study may help guide sampling studies aimed at collecting blood-fed females for host-range studies (Estrada-Franco et al. 2020, Olson et al. 2020) and can improve vector surveillance and disease management programs.

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