

SCIENTIFIC NOTE

NEW STATE RECORD FOR *CULEX REJECTOR* AND *CX. RESTRICTOR* IN YUCATAN, MEXICO

JULIO C. TZUC-DZUL,¹ JESUS E. AZCORRA-COUOH,¹ WILBERT A. CHI-CHIM,¹ NOHEMI CIGARROA-TOLEDO,² ELSY NALLELI LORIA-CERVERA,³ JULIAN E. GARCIA-REJON¹ AND CARLOS M. BAAK-BAAK^{1,4}

ABSTRACT. Several known mosquito species occur in the Mexican state of Yucatan, including competent and suspected vectors responsible for transmitting zoonotic arboviruses. Between February and July 2022, mosquito collections were carried out in several forested areas in Yucatan. As part of the ongoing monitoring, we present the first reports of *Culex (Microculex) rejector* and *Cx. (Anoedioparpa) restrictor*. Another 14 species were identified during the monitoring: *Aedes albopictus*, *Ae. bimaculatus*, *Ae. tormentor*, *Ae. cozumelensis*, *Anopheles albimanus*, *Cx. coronator* s.l., *Cx. erraticus*, *Cx. lactator*, *Cx. salinarius*, *Coquillettidia venezuelensis*, *Limatus durhamii*, *Psorophora ciliata*, *Toxorhynchites theobaldi*, and *Wyeomyia mitchellii*. Currently, the mosquito fauna in Yucatan consists of 65 species. The subgenera *Microculex* and *Anoedioparpa* had not been documented in Yucatan State prior to the current investigation.

KEY WORDS *Anoedioparpa*, first record, *Microculex*, mosquito fauna, mosquito update

Culex daumastocampa Dyar and Knab and *Culex rejector* Dyar and Knab, of the subgenus *Microculex*, are reported to occur in Mexico (Ortega-Morales et al. 2018). *Culex imitator* Theobald was first reported in Mexico by Knight and Stone (1977), and subsequent studies reported its occurrence in the Mexican states of Hidalgo, San Luis Potosi, and Tamaulipas (Ortega-Morales et al. 2015, 2018). Following specimen reexamination by the same author, the record for *Cx. imitator* was corrected to *Cx. rejector*. Currently, *Cx. imitator* was removed from the list of mosquitoes in Mexico (Ortega-Morales et al. 2018).

Within the subgenus *Anoedioparpa*, *Cx. conservator* (Dyar and Knab) and *Cx. restrictor* (Dyar and Knab) are recorded to occur in Mexico (Adeniran et al. 2021). In early, now outdated studies, Stone and colleagues (1959) placed *Tinolestes*, *Micraedes*, and *Anoedioparpa* in the subgenus *Aedinus* Bourroul (1904), based on the shared characteristic of males possessing short palps. However, this trait developed independently in several unrelated mosquito species (Belkin 1968); therefore, *Anoedioparpa* was promoted to subgeneric rank. According to Berlin and

Belkin (1980), *Anoedioparpa* can be recognized by a combination of characteristics. Adults have narrow decumbent scales on the vertex; short palpus in both sexes (except in *Cx. restrictor*); a general absence of acrostichal bristles on the mesonotum (except in *Cx. corrigani* Dyar and Knab and *Cx. restrictor*); a generally yellowish pleural integument; and dark tarsi. In male genitalia, a large densely setose area is observed on the inner surface of the sidepiece below the proximal division of the subapical lobe. The siphons of 4th instars sport 4 pairs of subventral hairs, 2 pairs of subdorsal hairs, an apical hook (2-S) with a small submedian branch, dorsal saddle hairs (2,3-X), and a long, single, and ventral brush usually with 5 pairs of hairs. *Culex restrictor* was originally described in Oaxaca, Mexico (Berlin and Belkin 1980), and considered a member of *Microculex* (Stone et al. 1959, Knight and Stone 1977), but reclassified by Berlin and Belkin (1980) after noting characteristics of both adult and immature mosquitoes resembled those of *Anoedioparpa*. At present, 12 species are currently included in the Conservator and Restrictor Groups of the subgenus *Anoedioparpa*. The Restrictor Group is monotypic and contains only *Cx. restrictor* (Berlin and Belkin 1980) due to highly particular traits not shared with other *Anoedioparpa* species (Berlin and Belkin 1980). Adult mosquitoes of the Restrictor Group present acrostichal bristles extending from the anterior promontory to the prescutellar area; and a bristled lower mesepimeron. The male genital lobe is distally divided; has specialized setae with 4 apical leaves; and cereal sclerite of proctiger digitiform. Fourth instars have a ventral brush (4-X) with 6 pairs of hairs; and a sclerotized plate at the base of the 2nd hair on the abdominal segment VIII (Berlin and Belkin 1980). The present study's objective was to monitor the

¹ Laboratorio de Arbovirología, Centro de Investigaciones Regionales “Dr. Hideyo Noguchi,” Universidad Autónoma de Yucatán, Mérida, Yucatán, C.P. 97225, México.

² Laboratorio de Biología Celular, Centro de Investigaciones Regionales “Dr. Hideyo Noguchi,” Universidad Autónoma de Yucatán, Mérida, Yucatán, C.P. 97000, México.

³ Laboratorio de Inmunología, Centro de Investigaciones Regionales “Dr. Hideyo Noguchi,” Universidad Autónoma de Yucatán, Mérida, Yucatán, C.P. 97000, México.

⁴ To whom correspondence should be addressed.

mosquito fauna found in Yucatan's forested areas and update the state occurrence list, if necessary.

Yucatan State (20°50'00"N, 89°00'00"W) is located on Mexico's Yucatan Peninsula, and bordered by the states Quintana Roo and Campeche. The rainy season extends from May to October, with a mean rainfall of 1,000 mm and mean temperature of 27.5°C. The dry season lasts from November to April, with a mean rainfall of 300 mm and a mean temperature of 25.1°C (Baak-Baak et al. 2016). Immature and adult mosquitoes were collected from February to July 2022. Immature mosquitoes from artificial and natural breeding sites were collected by nets, turkey basters, and pipettes, placed into plastic containers that were labeled with collection date, study site, and sample identification number, and transported to the Laboratorio de Arbovirología at Universidad Autónoma de Yucatán. A portion of the larvae were killed in hot water (60°C) and mounted on microscope slides in Euparal (BioQuip®, Galveston, TX). The remaining larvae were individually reared to adults and identified to the species level. Adult mosquitoes were collected using a Mosquito Magnet trap (American Biophysics Corp., North Kingstown, WA) and a mouth aspirator. Stereomicroscopes and taxonomic keys were used to identify species (Dyar and Knab 1906, Berlin 1969, Berlin and Belkin 1980, Clark-Gil and Darsie 1983, Rueda 2004). Voucher specimens were stored in the Arbovirology Laboratory at the Universidad Autónoma de Yucatán, México.

In our study, we identified 16 mosquito species belonging to 8 genera, with *Cx. restrictor* and *Cx. rejelector* as new records for Yucatan State. Table 1 shows the collected number of immature and adult mosquitoes per species, as well as the location and type of breeding grounds.

Culex restrictor was confirmed following the description of Berlin and Belkin (1980). Larvae were recognized by the presence of a ventral brush (4-X) with 6 pairs of hairs, a siphon with both subdorsal (2a-S) and subventral hairs (1, la-S) composed of 5 pairs, and hair 2-VIII on a sclerotized plate. The immature mosquito was found in deciduous forest near the community of Ekmul (20°57'54"N, 89°20'56"W), located 27 km east of Yucatan's capital city of Merida. The positive hatchery was located 15 m from a thoroughfare that connects Ekmul and Hubila in the municipality of Tixkokob. The area is populated by trees typical of the region, such as *Alvaradoa amorphoides* Liebm., *Bursera simaruba* (L.) Sarg., *Ehretia tinifolia* L., *Havardia albicans* (Kunth) Britton and Rose, *Leucaena leucocephala* (Lam.) deWit, *Mimosa bahamensis* Benth., *Piscidia piscipula* (L.) Sarg., and *Bromelia karatas* L. On June 16 and July 7, 2022, 12 mosquito larvae were collected in a 10-liter-capacity plastic bucket filled with dark-colored water with high organic matter, i.e., fallen leaves and decomposing arthropods.

Culex rejelector was confirmed following the description of Dyar and Knab (1906). Larvae were recognized by the presence of setae 6-III-VI of the abdomen with 2 hairs and comb scales with very long spines. Five *Cx. rejelector* larvae were found on July 2, 2022, in the water-filled leaf axils of the bromeliad *Aechmea bracteata* (Swartz) Grisebach. The *Cx. rejelector* was found in the community of Esmeralda (20°42'03"N, 87°36'34"W) located within the states of Quintana Roo and Yucatan, and 240 km east of Yucatan's capital, Merida. The site is represented by trees from the region, such as *Manilkara sapota* (L.) P. Royem, *Brosimum aliscastrum* Sw., *Piscidia piscipula*, *Bursera simaruba*, *Metopium brownei* Roxb., and *Vitex gaumeri* GreenM. The epiphytic and terrestrial forms of *Aechmea bracteata* bromeliad are abundant.

In the present study, we identified 16 mosquito species from sylvan habitats, including 2 species never before reported in Yucatan State: *Cx. restrictor* and *Cx. rejelector*. In the last update of mosquitoes of Yucatan, 52 species were registered (Baak-Baak et al. 2016). In this list, *Wyeomyia mitchellii* (Theobald) was not included (Díaz-Nájera and Vargas 1973). In recent years, the following 12 species have been identified: *Ae. albopictus* (Skuse), *Ae. bimaculatus* (Coq.), *Ae. tormentor*, *Anopheles punctipennis* (Say), *Coquillettidia venezuelensis* (Theobald), *Cx. erythrorothorax* Dyar, *Cx. rejelector* (present study), *Cx. restrictor* (present study), *Cx. taeniopus* Dyar and Knab, *Cx. trifidus* Dyar, *Mansonia dyari* Belkin, Heinemann, and Page, and *Toxorhynchites moctezuma* Theobald (Baak-Baak et al. 2016, Bond et al. 2020). With the addition of our findings, the current mosquito fauna of Yucatan consists of 65 species.

Culex rejelector was originally described from Veracruz, Mexico, from immature mosquitoes discovered and removed from leaf axils of bromeliads (Dyar and Knab 1906). Further distribution is listed in the countries Belize, Costa Rica, Guatemala, Honduras, and Nicaragua (WRBU 2022). *Anoedioporpa* mosquitoes are container breeders (Berlin and Belkin 1980). Their immature stages are typically collected in natural, phytotelma breeding sites such as leaf axils, bamboo internodes, and tree holes (Adeniran et al. 2021; Ortega-Morales et al. 2010, 2019b); they have also been found in discarded tires (Ortega-Morales et al. 2015, 2019a, 2019c). Mosquito adults are generally sylvan, though some have been found in trees in suburban areas (Ortega-Morales et al. 2019b, Adeniran et al. 2021).

Culex restrictor is currently distributed in the Mexican states of Hidalgo, Jalisco, Mexico, Nuevo Leon, Quintana Roo, San Luis Potosi, Tabasco, Tamaulipas, Veracruz, and Yucatan (Berlin and Belkin 1980; Ortega-Morales et al. 2010, 2015, 2019a, 2019b, 2019c; Adeniran et al. 2021). Previous studies reported *Cx. restrictor* from natural and artificial containers cohabiting with *Ae. albopictus*, *Ae. aegypti* (L.), *Ae. epactius* Dyar and Knab, *Ae. muelleri* Dyar, *Ae. podographicus* Dyar and Knab,

Table 1. Mosquito species collected in forest environments in Yucatan State, Mexico, between February and July of 2022, with information on mosquito gender, stage, method of collection, and collection site with coordinates.

Subgenus and species	Female	Male	Collecting method ¹	Immature	Source of collection	Coordinates
<i>Stegomyia</i>						
<i>Aedes albopictus</i>	6	2	MMT, mouth aspirator	Larvae (41) Pupae (17)	Disposable container, tree hole, bromeliad axil, ² sink	20°57'54"N, 89°20'56"W; 20°42'03"N, 87°36'34"W
<i>Ochlerotatus</i>						
<i>Ae. bimaculatus</i>	2	—	Mouth aspirator	—	—	20°42'03"N, 87°36'34"W
<i>Ae. tormentor</i>	1	—	Mouth aspirator	—	—	20°51'24"N, 90°22'32"W
<i>Howardina</i>						
<i>Ae. cozumelensis</i>	—	—	—	Larvae (9)	Sink, rock hole	20°57'54"N, 89°20'56"W; 20°42'03"N, 87°36'34"W
<i>Nyssorhynchus</i>						
<i>Anopheles albimanus</i>	3	—	MMT	—	—	20°42'03"N, 87°36'34"W
<i>Melanoconion</i>						
<i>Culex erraticus</i>	—	—	—	Larvae (14)	Lagoon	20°39'38"N, 87°36'48"W
<i>Culex</i>						
<i>Cx. coronator</i> s.l.	—	—	—	Larvae (40)	Sink, rock hole, drum	20°42'03"N, 87°36'34"W
<i>Cx. salinarius</i>	—	—	—	Larvae (15) Pupae (2)	Disposable container	20°57'54"N, 89°20'56"W
<i>Phenacomyia</i>						
<i>Cx. lactator</i>	—	—	—	Larvae (6)	Rock hole	20°42'03"N, 87°36'34"W
<i>Anoediopora</i>						
<i>Cx. restrictor</i> ³	—	—	—	Larvae (12) Pupae (3)	Disposable container	20°57'54"N, 89°20'56"W
<i>Microculex</i>						
<i>Cx. rejelector</i> ³	—	—	—	Larvae (5)	Bromeliad axil ²	20°42'03"N, 87°36'34"W
<i>Rhynchoetaenia</i>						
<i>Coquillettidia venezuelensis</i>	2	—	MMT, mouth aspirator	—	—	20°42'03"N, 87°36'34"W
<i>Limatus</i>						
<i>Limatus durhamii</i>	1	—	MMT	Larvae (11)	Disposable container	
<i>Psorophora</i>						
<i>Psorophora ciliata</i>	—	—	—	Larvae (2)	Tire	20°28'07"N, 89°42'52"W
<i>Lynchiella</i>						
<i>Toxorhynchites theobaldi</i>	—	—	—	Larva (1)	Bromeliad axil ²	20°42'03"N, 87°36'34"W
<i>Wyeomyia</i>						
<i>Wyeomyia mitchellii</i>	—	—	—	Larvae (20)	Bromeliad axil ²	20°42'03"N, 87°36'34"W

¹ MMT, Mosquito Magnet trap.

² *Aechmea bracteata*.

³ New record for Yucatan State.

Cx. coronator Dyar and Knab, *Cx. corniger* Theobald, *Limatus durhamii* Theobald, *Tx. moctezuma*, and *Wy. mitchellii* (Berlin and Belkin 1980; Ortega-Morales et al. 2010, 2019a, 2019b, 2019c). In a plastic bucket, we found *Cx. restrictor* cohabiting with *Ae. albopictus*. The area of the finding is in new growth in young deciduous forest, although it is also common to find it in oak forest and in suburban areas (Ortega-Morales et al. 2019b, Adeniran et al. 2021). Its distribution range appears to be wide, ranging from the Yucatan peninsula in southeastern Mexico to Tamaulipas in the northeastern part of the country (Berlin and Belkin 1980; Ortega-Morales et al. 2010, 2015, 2019a, 2019b, 2019c; Adeniran et al. 2021). Little is known about the biology of *Cx. restrictor* and *Cx. rejelector* and their vectorial capacity, although they are not currently incriminated as vectors of pathogens. The findings of the present study confirm the broad distribution pattern of the species.

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