Culicidae (Diptera, Culicomorpha) from the western Brazilian Amazon: Juami-Japurá Ecological Station

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ABSTRACT. Culicidae (Diptera, Culicomorpha) from the western Brazilian Amazon: Juami-Japurá Ecological Station. With 312 trap-hours of sampling effort, 1554 specimens of Culicidae (Diptera) were collected, using CDC and Malaise traps, in nine different locations along the Juami River, within the Juami-Japurá Ecological Station, Amazonas State, Brazil. A list of mosquito species with 54 taxa is presented, which includes three new distributional records for the state of Amazonas. The species found belong to the genera *Anopheles*, *Aedeomyia*, *Aedes*, *Psorophora*, *Culex*, *Coquillettidia*, *Sabethes*, *Wyeomyia* and *Uranotaenia*.

KEYWORDS. Amazonia; distribution; mosquitoes.

RESUMO. Culicidae (Diptera, Culicomorpha) da Amazônia Ocidental Brasileira: Estação Ecológica Juami-Japurá. Com um esforço amostral de 312 horas/armadilha, 1.554 espécimes adultos de Culicidae (Diptera) foram coletados, utilizando armadilhas CDC e Malaise, em nove localidades diferentes ao longo do Rio Juami, na Estação Ecológica Juami-Japurá, Amazonas, Brasil. Apresenta-se uma lista de espécies de mosquitos contendo 54 taxa, incluindo três novos registros de distribuição para o Estado do Amazonas. As espécies encontradas pertencem aos gêneros *Anopheles*, *Aedeomyia*, *Aedes*, *Psorophora*, *Culex*, *Coquillettidia*, *Sabethes*, *Wyeomyia* e *Uranotaenia*.

PALAVRAS-CHAVE. Amazônia; distribuição; mosquitos.

Determining the geographical distribution of species is necessary to increase our comprehension of mosquito systematics and to contribute towards our knowledge of their biodiversity. Likewise, it is of epidemiological interest because it permits one to better comprehend the role of mosquito species as vectors thereby facilitating the adoption of control methods (Nielsen 1980). However, very little is known about the mosquito fauna and its distribution within the Amazon region. Cerqueira (1961) registered the presence of 148 mosquito species collected in 24 localities within Amazonas State. Later, Xavier & Mattos (1976) added 25 new records, increasing the number of known species to 175 from 114 localities. Due to its complex geographical nature and difficult access, the Amazon region has areas, such as the interfluve between the Solimões River and Japurá River, in the western part of Amazonas State, where the mosquito fauna is practically unknown.

Therefore, with the objective of serving as a base inventory of the mosquito fauna of that region, this paper reports the species of Culicidae (Diptera, Culicomorpha) collected in the upland (*terra-firme*) rainforest and the floodplain (*varzea*) forest located along the Juami River within the Juami-Japurá Ecological Station, Amazonas State, Brazil.

MATERIAL AND METHODS

Mosquito specimens were collected in nine different localities, within the Juami-Japurá Ecological Station. The collection sites were distributed along a distance of 100 km, upstream from the mouth of the Juami River. The latter is an affluent of the Japurá River on its right margin, at the municipality of Japurá, Amazonas State, Brazil (between 01º44'S 67º36'W and 02º19'S 68º25'W) (Fig 1). The Juami-Japurá Ecological Station (JJES), a Brazilian federal conservation unit, is located within the interfluve of the Solimões River and the Japurá River.

The JJES is characterized by having most of its area covered by dense upland (*terra firme*) ombrophilous tropical forests with low plateaus together with areas of ecological tension with Amazonian white sand (*campinarana*) forests along the Juami River. Other areas of ecological tension between dense upland (*terra firme*) ombrophilous tropical forests and floodplain (*varzea*) forests are present along the mouth of the Juami River and along the Japurá River. Within the JJES, the climate is of the rainy tropical type (*Af* subgroup of the Köppen climate classification system), characterized by being constantly humid, with temperature and precipitation suffering little annual variation. Based on climatic data from Fonte Boa and São Paulo de Olivença (RADARBRASIL 1977), the less rainy season occurs from July to November with lowest monthly precipitation being over 120 mm. The rainy season occurs between December and June, with the maximum precipitation in April. The region's mean annual precipitation is 2687 mm, with a mean monthly relative humidity between 85% and 90% and a mean annual temperature of 25ºC. A more detailed description of the abiotic and biotic characteristics of the JJES can be found in the RADAMBRASIL (1977) publications and the Instituto Chico Mendes de Conservação da Biodiversidade website.

Due to its location in the western part of Amazonas State and the long distances from urban centers (400 km from Tefé and 1200 km from Manaus), the study site is very difficult to access. Consequently, this region has a very low demographic density that is concentrated in small settlements that occupy the margins of the Japurá River. The main form of transportation is riverine.

Mosquito specimens were collected in two different expeditions: September 23-28, 2004 and August 6-18, 2005. Mosquito adults were captured inside the *terra firme* forest along the river and within the *varzea* floodplain forest at the mouth of the river. Two types of CDC traps, each using a different light source (incandescent *vs.* ultraviolet light), placed one meter above the ground and distributed along recently opened trails, were used to collect specimens from 18h00 p.m. to 6h00 a.m. Malaise flight intercept traps, placed along recently opened forest trails, were also used to collect specimens during a 3-day period.

Mosquitoes were identified using the taxonomic keys provided by Lane (1953), Forattini (1965), and Berlin & Belkin (1980). Pecor *et al*. (1992) catalog for *Culex* (*Melanoconion*) was used to identify the species using characters of the male genitalia. Whenever possible, characteristics of the female cibarium were also examined to achieve a more precise species identification for members of the *Culex* (*Melanoconion*). Vouchers are deposited in the Invertebrate Collection of the Instituto Nacional de Pesquisas da Amazônia (INPA-Manaus) and in the Entomological Reference Collection of the Faculdade de Saúde Pública, Universidade de São Paulo (FSP-USP). The collection and specimen data was archived, organized and analyzed using the relational database structure provided by the Biota software (Colwell 1996).

RESULTS AND DISCUSSION

A total of 1554 adult mosquitoes were collected, belonging to 49 species distributed among nine genera (Table I). It should be noted that some of the identifications could not be precise due to the absence of male individuals, whose genitalia would possess the characters that would permit a specific identification. Likewise, it was not possible to identify some female specimens to species level because characters that can be used to separate individuals of the same sex do not exist or the characters that are used to separate these species are damaged or lost. These specimens were identified as morphotypes indicating the species to which they are morphologically similar. Consequently, the number of species could increase to 54 if we take into consideration the two cases for which the identifications are uncertain, i.e., *Anopheles* (*Ste*.) *nimbus / thomasi*, and *Aedes* (*Och*.) *serratus / nubilus* and the three morphospecies (i.e. *Culex* near *eastor* Form 1, *Culex* near *vaxus* Form 3 and *Uranotaenia* (*Ura.*) Form 3).

The genus *Culex*, with 28 species, presented the largest number of species (55%) and of individuals (86%), followed by *Uranotaenia* with 6 species (12%) representing only 1% of the individuals collected. In contrast, the genus *Anopheles* had the second greatest number of collected individuals (11%), which belong to 5 species (10%). More than 97% of the material collected (1512 individuals) belongs to these three genera, which together represent 77% of the identified species.

The most abundant species was *Culex* (*Mel*.) *gnomatos* Sallum, Hutchings & Ferreira (376 individuals, which represent 50% of the 759 specimens identified to species level) followed by *Culex* (*Mel.*) *vaxus* Dyar (with 120 individuals). These two species (4% of the species recorded) represent more than 65% of the specimens collected and identified to the species level. The third most abundant taxon was *Anopheles* (*Nys.*) *oswaldoi s.l.* (with 93 individuals).

It is worthwhile mentioning that the most abundant species at the JJES, *Cx. gnomatos*, is epidemiologically important as a competent vector of multiple arboviruses in the Peruvian Amazon Basin. In Peru, *Cx. gnomatos* was found to be the principal enzootic vector of the Venezuelan Equine Encephalomyelitis virus (VEEV) and also a competent vector of the Eastern Equine Encephalomyelitis virus (EEEV), both viruses belonging to the family Togaviridae, genus *Alphavirus* (Turell *et al.* 2005). Additionally, Turell *et al.* (2005) also indicated *Cx. gnomatos* as a competent vector of the Caraparu, Itaqui, Murutucu, Unidentified Group C, Unidentified Guama Group, and other Group not yet unidentified of viruses belonging to the family Bunyaviridae, genus *Orthobunyavirus*. Furthermore, *Cx. gnomatos* from the

Fig. 1. Location of the study site along the Juami River, within the Juami-Japurá Ecological Station (JJES), municipality of Japurá, Amazonas State, Brazil.

Table I. Species of Culicidae collected along the Juami River, within the Juami-Japurá Ecological Station (JJES), municipality of Japurá, Amazonas State, Brazil.

Subfamily	Tribe	Genus	Subgenus	Species	No.[1]	Sex[2]
Anophelinae		*Anopheles*	(*Anopheles*)	*forattinii* Wilkerson & Sallum,1999	5	F
				mattogrossensis Lutz & Neiva, 1911	1	F
			(*Nyssorhynchus*)	*darlingi* Root,1926	7	F
				oswaldoi s.l.	94	F
				triannulatus (Neiva & Pinto,1922)	3	F
				(*Nys.*) sp.	53	F
			(*Stethomyia*)	*nimbus* / *thomasi*[3]	1	F
				Total genus *Anopheles*	164 (32%)	
Culicinae	Aedeomyiini	*Aedeomyia*	(*Aedeomyia*)	*squamipennis* (Lynch Arribalzaga,1878)	2	F
				Total genus *Aedeomyia*	2 (0%)	
	Aedini	*Aedes*	(*Ochlerotatus*)	*dupreei* (Coquillett,1904)	1	F
				fulvus (Wiedemann,1828)	1	F
				serratus / *nubilus*[3]	11	F
				Total genus *Aedes*	13 (0%)	
		Psorophora	(*Grabhamia*)	*cingulata* (Fabricius,1805)	2	F
				dimidiata Cerqueira,1943	4	3F+1M
			(*Janthinosoma*)	*albipes* (Theobald,1907)	3	F
				amazonica Cerqueira,1960	2	F
				(*Jan.*) sp.	5	Fdam
				Total genus *Psorophora*	16 (31%)	
	Culicini	*Culex*	(*Aedinus*)	*accelerans* Root,1927	2	Mgen
				amazonensis (Lutz,1905)	4	F
				(*Ads.*) sp.	1	Mdam
			(*Culex*)	*declarator* Dyar & Knab,1906	3	F
				mollis Dyar & Knab,1906	5	F
				gr. coronator	3	F
				(*Cux.*) sp.	3	Fdam
			(*Melanoconion*)	*bequaerti* Dyar & Shannon, 1925	1	Mgen
				caudatus Clastrier,1970	10	Mgen
				comatus Sénevet & Abonnenc,1939	4	Mgen
				contei Duret, 1968	1	Mgen
				creole Anduze, 1948	1	Mgen
				eastor Dyar,1920	14	Mgen
				near *eastor* Form 1	5	Mgen
				eknomios Forattini & Sallum,1992	8	F
				fairchildi Galindo & Blanton, 1954	2	Mgen
				foliafer Komp & Rozeboom,1951	1	Mgen
				gnomatos Sallum, Hutchings & Ferreira,1997	376	375F+1Mgen
				johnnyi Duret,1968[4]	5	Mgen
				mesodenticulatus Galindo & Mendez, 1961	1	Mgen
				ocossa Dyar & Knab,1919	2	F
				pedroi Sirivanakarn & Belkin,1980	7	F
				phyllados Hutchings & Sallum, 2008[5]	1	Mgen
				portesi Senevet & Abonnenc,1941	8	F
				putumayensis Matherson,1934	3	Mgen
				spissipes (Theobald,1903)	5	3F+2M
				symbletos Sallum & Hutchings,2003	4	Mgen
				vaxus Dyar,1920	120	110F+10Mgen
				near *vaxus* Form 3	8	Mgen
				vomerifer Komp,1932	1	F
				ybarmis Dyar,1920	1	Mgen
				gr. Pilosus	282	265F+17Mdam
				sec. Melanoconion	13	12F+1Mdam
				sec. Spissipes	8	F
				(*Mel.*) sp.	420	409F+11M
				Total genus *Culex*	1333 (55%)	
	Mansoniini	*Coquillettidia*	(*Rhynchotaenia*)	*arribalzagae* (Theobald,1903)	1	M
				venezuelensis (Theobald,1912)	1	F
				(*Rhy.*) sp.	4	F
				Total genus *Coquillettidia*	6 (67%)	

Table I. Cont.

Subfamily	Tribe	Genus	Subgenus	Species	No.[1]	Sex[2]
	Sabethini	*Sabethes*	(*Sabethes*)	*batesi* Lane & Cerqueira,1941	1	M
				lanei Cerqueira,1961	1	M
				shannoni Cerqueira,1961	1	F
				Total genus *Sabethes*	3 (0%)	
		Wyeomyia		sp.	2	Fdam
				Total genus *Wyeomyia*	2 (100%)	
	Uranotaeniini	*Uranotaenia*	(*Uranotaenia*)	*ditaenionota* Prado,1931	2	F
				geometrica Theobald,1901	3	F
				hystera Dyar & Knab,1913[4]	1	M
				incognita Galindo, Blanton & Peyton,1954[4]	1	F
				leucoptera (Theobald,1907)	2	F
				pallidoventer Theobald,1903	4	3M+1F
				(*Ura.*) sp. Form 3	1	F
				(*Ura.*) sp.	1	F
				Total genus *Uranotaenia*	15 (7%)	
				Total number of individuals	1554 (51%)	

[1] Indicates the number of specimens collected: The value in parenthesis indicates the proportion (%) of individuals that was only possible to identify to genus (section or group) because the characters used for identification were damaged or lost.
[2] Indicates the sex, and condition, of the specimens collected: M = male; F = female; dam = damaged specimen; gen = identified using genitalia dissection.
[3] It was not possible to identify to species level because characters that can be used to separate individuals of the same sex do not exist or the characters that are used to separate these species are damaged or lost.
[4] First published record for Amazonas State.
[5] This specimen is a paratype of this species (Hutchings & Sallum 2008).

Peruvian Amazon basin, near Iquitos, was demonstrated to be the most efficient vector of the subtype IIIC virus in the Venezuelan Equine Encephalomyelitis complex, with a 10-fold higher estimated transmission rate than any other species tested (Turell *et al.* 2006). Based on these literature records of *Cx. gnomatos*, it is possible to speculate that this species may be involved in the transmission of arboviruses in the Brazilian Amazon, including the area of the Juami-Japurá Ecological Station.

Identification of members of the *An. oswaldoi* species complex is not possible based on morphological characters. Currently, this species complex is composed of *An. oswaldoi* (Peryassú) which occurs in areas of the costal lowland of the Mata Atlântica, in the states of Espírito Santo, Rio de Janeiro and São Paulo (Motoki *et al.* 2007) and likely two or three other taxa that occur in localities of Acre State (Sallum *et al.* 2008; Scarpassa 2005), Amazonas State (Marrelli *et al.* 1999), and Colombia (Ruiz *et al.* 2005). However, it is also plausible to assume that among the individuals identified as *An. oswaldoi s.l.* there may be some specimens of *An. konderi s.s.* Galvão & Damasceno or *An. konderi s.l.*, as identified in Acrelândia, Acre State by Sallum *et al.* (2008).

It was not possible to identify 795 individuals (51%) to species level. Consequently, these specimens were recognized to the level of genus, subgenus or other informal inferior groups (sections or groups). Most of the individuals that could not be identified to species level are females (95%) and belong to the genus *Culex* (85%) and *Anopheles* (Table I). It is interesting to note that only 7% of the 1554 specimens collected were males which were indispensable to identify more than 40% of the taxa in this inventory.

Among the taxa identified, there are three new species distribution records for the state of Amazonas (*Culex* (*Mel.*) *johnnyi* Duret, *Uranotaenia* (*Ura.*) *hystera* Dyar & Knab and *Uranotaenia* (*Ura.*) *incognita* (Galindo, Blanton & Peyton) and three morphospecies which may be undescribed taxa (*Cx.* near *eastor* Form 1, *Cx.* near *vaxus* Form 3 and *Ur.* (*Ura.*) sp. Form 3).

With a total combined sampling effort of 312 trap-hours, the CDC traps were responsible for more than 99% of the captured adults while the Malaise trap only captured five mosquitoes. The adult specimens of *Anopheles*, *Aedeomyia*, *Aedes*, *Psorophora*, *Culex*, *Coquillettidia* and *Uranotaenia* were all collected at night using the CDC traps. In contrast, due to their diurnal habits, all the adults of *Sabethes* and *Wyeomyia* were only captured using a Malaise trap. The diurnal mosquitoes are not realistically represented in this inventory because they were not adequately sampled as the only diurnal collecting method used was the Malaise trap.

We must also recognize that it was only possible to sample a small portion of the area and of the habitats that exist in the JJES. Although we were able to collect along the lower portion of the main river, logistical limitations restricted the collections to the areas of ecological tension between the *terra firme* forests and the *campinarana* forests. Consequently, the sampling coverage was concentrated in the fluvial plains and marginal terraces along the margins of the Juami River.

Therefore, in order for further mosquito inventories to be more representative, it will be necessary to plan future collecting so as to: include other methods which sample the diurnal fauna (aspirator and net sweeping) and collect immature forms for rearing; and sample the mid and upper parts of the Juami River, including further sampling of other phytoecological formations and habitats found in the interfluvial regions and in the other areas of ecological tension with *varzea* forests which are located around the mouth of the

Juami River and along the Japurá River.

Likewise, the results of this inventory indicate that many more species of *Culex* and *Anopheles* could have been identified if more male specimens had been collected. Therefore, it becomes evident that future mosquito inventories should also include methodology that maximizes the collection of male specimens (i.e. CDC traps with UV light source and mechanical aspirator) which would significantly increase the number of species which can be identified.

We found no previously published mosquito distributional record for the municipality of Japurá. Therefore, the results of the current inventory represent the first report of these 54 taxa for this municipality. On a larger geographical scale, of the 49 species collected in the Juami-Japurá Ecological Station, four species (8%) represent new records for Amazonas State. In other inventories, we also observed that of the 119 species reported by Hutchings *et al.* (2005) for the Jaú National Park, in the municipality of Novo Airão, 30 species (25%) were new records for Amazonas State. Likewise, of the 44 species reported by Hutchings *et al.* (2002) for the locality of Querari, in the municipality of São Gabriel da Cachoeira, 12 species (27%) were new records for Amazonas State.

Despite the relatively low sampling effort and coverage, compared to that of the other mosquito inventories from different regions of the Amazon, this type of "fast" inventory yielded an important initial contribution to our knowledge of the geographical distribution and diversity of mosquitoes in a very remote, and difficult to access, region of the Western Amazon. Nevertheless, more inventories, in many different areas, continue to be necessary in order to adequately estimate the biodiversity of Amazonian mosquitoes.

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