

Diversity of *Culicoides* (Diptera: Ceratopogonidae) in the National Forest of Caxiuanã, Melgaço, Pará State, Brazil

Diversidade de *Culicoides* (Diptera: Ceratopogonidae) na Floresta Nacional de Caxiuanã, Melgaço, Estado do Pará, Brasil

Diversidad de *Culicoides* (Diptera: Ceratopogonidae) en el Bosque Nacional de Caxiuanã, Melgaço, Estado de Pará, Brasil

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ABSTRACT

The genus *Culicoides* is distributed worldwide and includes nearly 82 species recorded in the Brazilian Amazon. In spite of the sanitary and economic relevance of the genus, few studies have been addressed to its members in this region. This study provides a survey of *Culicoides* species in a research plot of the Brazilian Biodiversity Research Program (PPBio) in the Amazon region in Caxiuanã National Forest. Collections were carried out with CDC light traps at five terrestrial sampling points from February 9 to February 13 in 2007 on the ground and at the sub-canopy level. After identifying the collected species, the frequency of species in each stratus and the Simpson's diversity index were calculated. A total of 542 specimens were collected, belonging to six species: *C. foxi* Ortiz, *C. fusipalpis* Wirth & Blanton, *C. glabrior* Macfie, *C. guerrai* Wirth & Blanton, *C. hylas* Macfie and *C. vernoni* Wirth & Blanton. The diversity of species was low in both strata ($\lambda = 0.91$ on the ground; $\lambda = 0.89$ in the sub-canopy). *C. fusipalpis* was the most abundant species at all sampling points and in both strata, corresponding to 94.8% of the collected specimens. This species can be found in both floodplain ecosystems and modified areas, such as hydroelectric dams, in the Amazon Region, feeding on humans. In spite of its hematophagous activities, the species is not implicated as a vector of tropical diseases.

Keywords: Ceratopogonidae; Biodiversity; Insect Vectors.

INTRODUCTION

Ceratopogonidae are small nematoceran Diptera that can present different habits. Some of the genera in this group are ectoparasites of large insects, while others are important pollinators. Haematophagous midges are known to transmit three types of organisms, including 66

types of viruses, 15 species of protozoa and 26 species of filarial nematodes, to a diversity of hosts, including humans and domestic and wild animals¹. Additionally, their intensive biting activity leads to significant nuisance and may cause dermatological problems when these insects are present in abundance. Animal diseases, such as bluetongue, encephalitis and equine onchocerciasis, are transmitted by different species of biting midges. The human disease mansonellosis (*Mansonella ozzardi*) is transmitted by *Culicoides* in Central America, on Caribbean islands and in South America, and Shelley and Coscarón² recently suggested its transmission by *C. lahillei* (Iches) in Argentina. Oropouche fever is one of the most important arboviral diseases in the Americas³. It is transmitted to humans by *C. paraensis* (Goeldi) in urban

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sites of the Brazilian Amazon Region, and more than 30 outbreaks have been reported in the Amazon and Central Plateau regions, reaching up to 100,000 human infections in same cases⁴.

The genus *Culicoides* includes 1,311 extant species distributed worldwide¹. Borkent and Spinelli⁵ listed 266 species from the Neotropical Region and nearly 82 in the Brazilian Amazon. In spite of the sanitary and economic relevance of the genus, few studies have been carried out on its members in this region. Wirth and Blanton⁶ described 15 new species in a list of 60 species recorded for the Amazon Basin. Two new species were described posteriorly by Felipe-Bauer et al^{7,8}, *C. kampa* Felipe-Bauer, Veras & Castellón and *C. baniwa* Felipe-Bauer, from the Brazilian states of Acre and Amazonas, and Spinelli et al⁹ described *C. felippebaueri* Spinelli from Amazonas. Surveys of *Culicoides* were performed by Castellón¹⁰ and Castellón and Ferreira¹¹ in the Ducke Forest Reserve, Manaus, Amazonas State, and Trindade and Gorayeb^{12,13} in coastal areas in the Itatupã-Baquiá Reserve in Pará State, Brazil.

The aim of this study is to provide a survey of *Culicoides* species in a research plot of the Caxiuanã National Forest, a forested freshwater ecosystem, in collaboration with the Brazilian Biodiversity Research Program (*Programa de Pesquisa em Biodiversidade – PPBio*) aimed at improving our knowledge about the biodiversity of the Brazilian Amazon Region.

MATERIALS AND METHODS

The Caxiuanã National Forest is a typical dense rain forest located in the north central region of Pará State, Brazil, characterized by "terra firme", or upper level forest (80%), a small floodplain and several "igapó" (flooded forest) areas (20%). The climate is tropical humid, with a short dry period. Meteorological studies show two defined periods in the area: humid or rainy, from January to March, and dry, from September to November. The average temperature is approximately 26° C, with a minimum of 22° C and a maximum of 32° C¹⁴. This study was developed at the Ferreira Penna Research Station, which covers an area of 33,000 ha within the National Forest of Caxiuanã (Figure 1) and is under the administration of the Museu Paraense Emílio Goeldi (MPEG). The plot at Caxiuanã of PPBio had an area of 25 km² and was divided into 30 sampling sites. Collections were performed using ten CDC light traps, with two in each of the five terrestrial sites of the plot (1, 4, 7, 8 and 9), from February 9 to February 13 in 2007 between 6 pm and 6 am on the ground and in the sub-canopy stratus (around 20 m of height).

The specimens collected by the Fundação Oswaldo Cruz and MPEG team were preserved dry using naphthalene and sent to the Ceratopogonidae Collection of the Laboratory of Diptera of the Instituto Oswaldo Cruz for identification. Individual specimens were mounted on microscope slides in phenol-balsam as described by Wirth and Marston¹⁵. For identification, we used the reports on *Culicoides* by Wirth and Blanton⁶ for the Amazon Basin,

Spinelli et al¹⁶ for the *guttatus* group, Felipe-Bauer et al¹⁸ for the *hylas* group of the *Hoffmania* subgenus, and the atlas of wing photographs of neotropical species produced by Wirth et al¹⁷. Voucher specimens were deposited in the collection of the MPEG.

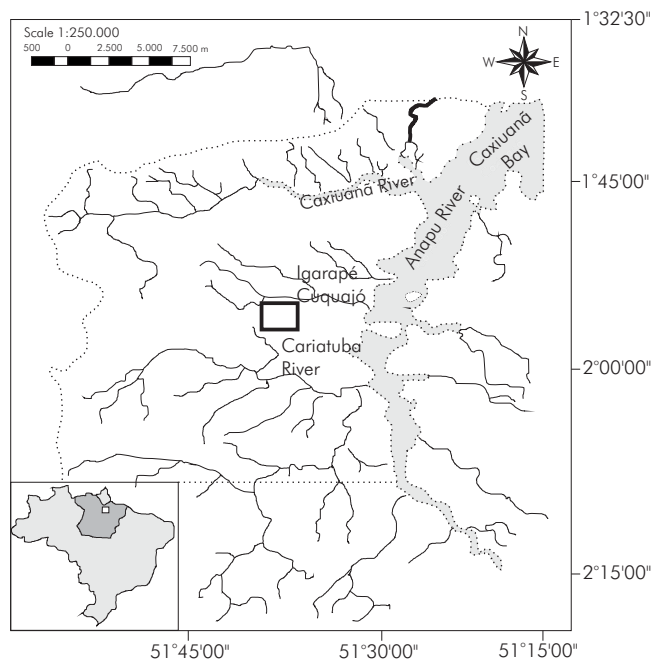


Figure 1 – Research plot (square) of Caxiuanã National Forest, in the Brazilian Biodiversity Research Program – PPBio

The frequencies of the *Culicoides* species were calculated based on the abundance of each species in relation to the total abundance of all of the collected species from the ground and the sub-canopy strata. The diversity of each stratus and site was assessed by Simpson's diversity index.

RESULTS

A total of 542 specimens of *Culicoides* were collected during the sampling period on the ground and in the sub-canopy stratus, with 99.6% female and 0.4% male specimens being captured. Six species were collected: *C. foxi* Ortiz, *C. fusipalpis* Wirth & Blanton, *C. glabrior* Macfie, *C. guerrai* Wirth & Blanton, *C. hylas* Macfie, and *C. veroni* Wirth & Blanton. The most abundant species were *C. fusipalpis* (94.8%), *C. hylas* (2.6%) and *C. foxi* (2%). The remaining species represented 0.6% of the sample.

The distributions of the species for each stratus and site are shown in table 1. The frequency of the most abundant species, *C. fusipalpis*, was similar in both strata (95.3% on the ground; 94.4% in the sub-canopy). The diversity of species was low in both strata ($\lambda = 0.91$ on the ground; $\lambda = 0.89$ in sub-canopy) and in all sites ($\lambda = 0.85$ in site I; $\lambda = 0.96$ in site IV; $\lambda = 0.94$ in site VII; $\lambda = 0.80$ in site VIII; $\lambda = 0.78$ in site IX). For *C. fusipalpis* and *C. hylas*, we obtained only females both on the ground and at the sub-canopy level, while for *C. foxi*, females were found in both strata, and a single male was found in the sub-canopy (Table 1).

Table 1 – Abundance of species on the ground and sub-canopy (20 m) stratus in the research plot of Caxiuanã National Forest

Species	Terrestrial sites										Total
	Ground					Subcanopy					
	I	IV	VII	VIII	IX	I	IV	VII	VIII	IX	
<i>C. fusipalpis</i>	15 ♀	55 ♀	99 ♀	40 ♀	35 ♀	31 ♀	86 ♀	106 ♀	35 ♀	12 ♀	514 ♀
<i>C. foxi</i>	–	–	2 ♀	–	2 ♀	1 ♀	1 ♀	2 ♀; 1 ♂	1 ♀	1 ♀	10 ♀; 1 ♂
<i>C. hylas</i>	–	–	–	5 ♀	3 ♀	3 ♀	–	–	3 ♀	–	14 ♀
<i>C. glabrior</i>	–	–	–	–	–	–	1 ♀	–	–	–	1 ♀
<i>C. vernoni</i>	–	–	–	–	–	–	1 ♂	–	–	–	1 ♂
<i>C. guerrai</i>	–	–	–	–	–	–	–	1 ♀	–	–	1 ♀
TOTAL	15 ♀	55 ♀	101 ♀	45 ♀	40 ♀	35 ♀	88 ♀; 1 ♂	109 ♀; 1 ♂	39 ♀	13 ♀	540 ♀; 2 ♂

Conventional sign used: – Numeric data equal to zero is not due to rounding.

DISCUSSION

Aitken (nec. Wirth and Blanton⁶) found the same six species collected in this study in Caxiuanã National Forest while working in floodplain areas in the Área de Pesquisas Ecológicas do Guamá (APEG) forest, Pará State, Brazil. Both of these localities in Pará are freshwater ecosystems, which may explain the similarity of the results.

In agreement with our data, Veras and Castellón¹⁸ observed *C. fusipalpis*, *C. hylas* and *C. foxi* on the ground and at different levels (1, 5, 10 and 15 m) and obtained a few male specimens in Ducke Reserve in Manaus, Amazonas State. However, Aitken (nec. Wirth and Blanton⁶) collected a greater number of *C. fusipalpis* on the ground than in the sub-canopy stratus in APEG Forest, and Castellón et al¹⁹ found male specimens corresponding to 31% of the *C. hylas* collected at all levels (1, 5 and 10 m) in hydroelectric dam areas in Amazonas.

C. fusipalpis has been observed feeding on mammals and birds as *Didelphis* sp. (opossum), *Mesocricetus* sp. (hamster) and *Gallus* sp. (chicken), as well as feeding on humans^{6,18,19,20,21}, which indicates an eclectic biting habit of this species.

In previous reports, it has been observed that *C. fusipalpis* is abundant in floodplain ecosystems in the Amazon Region, such as natural preserved areas, and areas subjected to considerable human interference, like hydroelectric dams^{6,19}. These areas are enriched by the mixture of estuarine waters with sediments, favoring the development of the *Culicoides* fauna in such regions⁶.

CONCLUSION

The species collected in this study in Caxiuanã National Forest are common in different ecosystems in the Amazon Region, including environments impacted by human

activities, where *C. fusipalpis* has been observed in great numbers. This species can be found from the ground up to heights of 15 m and exhibits eclectic haematophagous habits, biting humans and other mammals and birds. In spite of its anthropophilic behavior, this species is not implicated as vector of tropical diseases.

We suggest that the reason that we observed females outnumbering male specimens may be due to having performed collections in a dry terrestrial area. If the captures had been carried out near breeding sites, where sources of water are present, more males would have been found.

Additional studies regarding bio-ecological aspects of *Culicoides*, such as their spatial distribution and breeding sites, in preserved areas like Caxiuanã National Forest are necessary to understand the natural cycles of the species of this genus in forest/freshwater ecosystems. Such studies will provide data that can be useful for future management strategies for the control of biting midges.

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RESUMO

O gênero *Culicoides* apresenta distribuição global e abrange cerca de 82 espécies registradas na Amazônia Brasileira. Apesar de sua relevância nas áreas sanitária e econômica, poucas pesquisas têm sido realizadas sobre suas espécies na região. Este estudo apresenta um levantamento das espécies de *Culicoides* em um sítio de pesquisa do Programa de Pesquisa em Biodiversidade (PPBio) na Floresta Nacional de Caxiuanã, na Região Amazônica. As coletas foram realizadas com armadilhas luminosas tipo CDC em cinco pontos de amostragem de 9 de fevereiro a 13 de fevereiro de 2007, nos níveis do solo e subdossel. Após a identificação das espécies coletadas, a sua frequência em cada estrato e seu índice de diversidade de Simpson foram calculados. Foram coletados 542 espécimes, pertencentes a seis espécies: *C. foxi* Ortiz, *C. fusipalpis* Wirth & Blanton, *C. glabrior* Macfie, *C. guerrai* Wirth & Blanton, *C. hylas* Macfie e *C. vernoni* Wirth & Blanton. A diversidade de espécies foi baixa em ambos os estratos ($\lambda = 0,91$ no solo; $\lambda = 0,89$ em nível subdossel). *C. fusipalpis* foi a espécie mais abundante em todos os pontos de amostragem e em ambos os estratos, correspondendo a 94,8% dos espécimes coletados. Na Região Amazônica, esta espécie pode ser encontrada tanto em ecossistemas de várzea como em áreas modificadas, como as localizadas no entorno de usinas hidrelétricas, alimentando-se em seres humanos. Apesar de seus hábitos hematófagos, a espécie não é apontada como vetor de doenças tropicais.

Palavras-chave: Ceratopogonidae; Biodiversidade; Insetos Vetores.

Diversidad de *Culicoides* (Diptera: Ceratopogonidae) en el Bosque Nacional de Caxiuanã, Melgaço, Estado de Pará, Brasil

RESUMEN

El género *Culicoides* presenta distribución global y abarca cerca de 82 especies registradas en la Amazonía Brasileña. A pesar de su relevancia en las áreas sanitaria y económica, han sido realizadas pocas investigaciones sobre sus especies en la región. Este estudio presenta un registro de las especies de *Culicoides* en un local de investigación del Programa de Investigación en Biodiversidad (PPBio) en el Bosque Nacional de Caxiuanã, en la Región Amazónica. Las colectas se realizaron con trampas luminosas de tipo CDC en cinco puntos de muestreo entre 9 y 13 de febrero de 2007, a niveles del suelo y subdossel. Luego de la identificación de las especies colectadas, fueron calculados su frecuencia en cada estrato y su índice de diversidad de Simpson. Se colectaron 542 especímenes, pertenecientes a seis especies: *C. foxi* Ortiz, *C. fusipalpis* Wirth & Blanton, *C. glabrior* Macfie, *C. guerrai* Wirth & Blanton, *C. hylas* Macfie y *C. vernoni* Wirth & Blanton. La diversidad de especies fue baja en ambos estratos ($\lambda = 0,91$ en el suelo; $\lambda = 0,89$ a nivel subdossel). *C. fusipalpis* fue la especie más abundante en todos los locales de muestreo y en ambos estratos, correspondiendo a 94,8% de los especímenes colectados. En la Región Amazónica, esta especie puede ser encontrada tanto en ecosistemas de várzea (húmedales) como en áreas modificadas, como las localizadas en el entorno de usinas hidroeléctricas, alimentándose en seres humanos. A pesar de sus hábitos hematófagos, la especie no se señala como vector de enfermedades tropicales.

Palavras clave: Ceratopogonidae; Biodiversidad; Insectos Vectores.



REFERENCES

- 1 Borkent A. World species of biting midges (Diptera: Ceratopogonidae). 2009. Available from: <http://www.inhs.illinois.edu/research/FLYTREE/Borkent.html>.
- 2 Shelley AJ, Coscarón S. Simuliid blackflies (Diptera: Simuliidae) and ceratopogonid midges (Diptera: Ceratopogonidae) as vectors of *Mansonella ozzardi* (Nematoda: Onchocercidae) in Northern Argentina. Mem Inst Oswaldo Cruz. 2001 May;96(4):451-8.
- 3 Pinheiro FP, Travassos da Rosa APA, Vasconcelos PFC. An overview of Oropouche fever epidemics in Brazil and neighbor countries. In: Travassos da Rosa APA, Vasconcelos PFC, Travassos da Rosa JFS, editors. An overview of arbovirology in Brazil and neighbouring countries. Belém: Instituto Evandro Chagas; 1998. p. 186-92.
- 4 Figueiredo LTM. Emergent arboviruses in Brazil. Rev Soc Med Trop. 2007 Mar-Apr;40(2):224-9.
- 5 Borkent A, Spinelli GR. Neotropical Ceratopogonidae (Diptera: Insecta). In: Adis J, Arias JR, Rueda-Delgado G, Wantzen KM, editors. Aquatic biodiversity in Latin America. Moscow: Pensoft Publishers; 2007. 198 p.
- 6 Wirth WW, Blanton FS. A review of the maruins or biting midges of the genus *Culicoides* (Diptera: Ceratopogonidae) in the Amazon Basin. Amazoniana. 1973;4:405-70.
- 7 Felipe-Bauer ML, Veras RS, Castellon EG, Moreira NA. A new *Culicoides* from the Amazonian Region, Brazil (Diptera: Ceratopogonidae). Mem Inst Oswaldo Cruz. 2000 Jan-Feb;95(1):35-7.
- 8 Felipe-Bauer ML, Damasceno CP, Py-Daniel V, Spinelli GR. *Culicoides baniwa* sp.nov. from the Brazilian Amazon Region with a synopsis of the *hylas* species group (Diptera: Ceratopogonidae). Mem Inst Oswaldo Cruz. 2009 Sep;104(6):851-7.

- 9 Spinelli GR, Ronderos MM, Marino PI, Carrasco DS, Ferreira RLM. Description of *Culicoides* (*Mataemyia*) *felippebaueri* sp.n., *Forcipomyia musae* immatures, and occurrence of *F. genualis*, breeding in banana stems in Brazilian Amazonia (Diptera: Ceratopogonidae). Mem Inst Oswaldo Cruz. 2007 Sep;102(6):659-9.
- 10 Castellón EG. *Culicoides* (Diptera: Ceratopogonidae) na Amazônia brasileira. II. Espécies coletadas na Reserva Florestal Ducke, aspectos ecológicos e distribuição geográfica. Acta Amaz. 1990;20:83-93.
- 11 Castellón EG, Ferreira RLM. *Culicoides* Latreille (Diptera: Ceratopogonidae) da Amazônia. III. Resultados de coletas noturnas, na Reserva Florestal Ducke, Estado do Amazonas, Brasil. Bol Mus Para Emilio Goeldi. 1991;7(2):117-23.
- 12 Trindade RL, Gorayeb IS. Maruins (Ceratopogonidae: Diptera) do estuário do Rio Pará e do litoral do Estado do Pará, Brasil. Entomol Vect. 2005 jan-mar;12(1):61-74.
- 13 Trindade RL, Gorayeb IS. Maruins (Diptera: Ceratopogonidae: *Culicoides*), após a estação chuvosa, na Reserva de Desenvolvimento Sustentável Itatupã-Baquiá, Gurupá, Pará, Brasil. Rev Pan-Amaz Saude. 2010 Jun;1(2):121-30.
- 14 Silva SS. Vespas sociais da Floresta Nacional de Caxiuanã, Melgaço, Pará. Descrição da fauna numa grade de 25 km² e comparação entre protocolos de amostragem (Hymenoptera, Vespidae) [dissertação]. Belém (PA): Universidade Federal do Pará, Museu Paraense Emílio Goeldi; 2007. 102 p.
- 15 Wirth WW, Marston N. A method for mounting small insects on microscope slides in Canada balsam. Ann Entomol Soc Am. 1968;61(3):783-4
- 16 Spinelli GR, Greiner EC, Wirth WW. The neotropical bloodsucking midges of the *Culicoides guttatus* group of the subgenus *Hoffmania* (Diptera: Ceratopogonidae). Contrib Am Entomol Inst. 1993;27(3):1-91.
- 17 Wirth WW, Dyce AL, Spinelli GR. An atlas of wing photographs, with a summary of the numerical characters of the neotropical species of *Culicoides* (Diptera: Ceratopogonidae). Contrib Am Entomol Inst. 1988;25:1-72.
- 18 Veras RS, Castellón EG. *Culicoides* Latreille (Diptera: Ceratopogonidae) in Brazilian Amazon. V. Efficiency of traps and baits and vertical stratification in the forest reserve Adolpho Ducke. Rev Bras Zool. 1998;15(1):145-52.
- 19 Castellón EG, Ferreira RLM, Silva MNT. *Culicoides* (Diptera: Ceratopogonidae) da Amazônia Brasileira. I. Coletas na usina hidrelétrica (UHE) de Balbina, usina hidrelétrica (UHE) Cachoeira Porteira e Cachoeira dos Espelhos (Rio Xingú). Acta Amaz. 1990;20:77-81.
- 20 Castellón EG, Ferreira RM, Silva MNT. *Culicoides* (Diptera: Ceratopogonidae) in the Brazilian Amazon. IV. Species collected with CDC light trap in the Ducke Forest Reserve (RFD), Amazon State, Brazil. Acta Amaz. 1993;23(2-3):309-10.
- 21 Tikasingh ES. Seasonal and diurnal activities of four species of Trinidadian *Culicoides* (Diptera: Ceratopogonidae). Mosq News. 1972;32(3):447-52.

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