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Inventory

Diversity of Biting Midges *Culicoides* (Diptera: Ceratopogonidae) in Pacoti Municipality, Baturité Highland, Ceará State, Northeast of Brazil

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Abstract: *Culicoides* is a small dipteran genus of Ceratopogonidae Family with worldwide distribution. Due to their hematophagous habits, species within this genus are a biting nuisance to humans and domestic animals . They also transmit protozoa, filarial worms, and several arboviruses. The Northeastern region of Brazil is characterized by a semi-arid climate and few bodies of water, which does not favor breeding sites for ceratopogonids. In addition to the climatic factor, studies on Biting Midges *Culicoides* are scarce in this geographic region. The municipality of Pacoti, located in Serra de Baturité, in Ceará, is an important area for agricultural production and ecotourism; also, Pacoti is located in an environmental protection area. Due to the medical and veterinary importance of these biting midges, a survey was conducted in Pacoti. For capture, CDC-type light traps were used near forested areas, where collections were carried out during two periods: March 2015, and August 2022. Of the 17 species found, 12 are new records for the municipality of Pacoti, nine are new records for the state of Ceará, and four are recorded for the first time for the Northeast region. Of the registered incriminated vectors, we found *C. paraensis*, *C. insignis*, and *C. pusillus*. The knowledge of biodiversity and geographic distribution update of *Culicoides* may contribute to prevention and control measures for vector-borne diseases in Ceará.
Keywords: *Anthropophilic behaviour; Culicomorpha; hematophagous; Northeast, vectors.*

Diversidade de Maruins *Culicoides* (Diptera: Ceratopogonidae) no Município de Pacoti, Planalto de Baturité, Estado do Ceará, Nordeste do Brasil

Resumo: O gênero *Culicoide*s é um diminuto diptero da família Ceratopogonidae com distribuição mundial. Devido aos seus hábitos hematófagos, as espécies desse gênero são um incômodo para os humanos e animais domésticos transmitem protozoários, vermes filariais, bem como, diversos arbovírus. A região Nordeste do Brasil é caracterizada por clima semiárido e poucos corpos d'água, o que não favorece criadouros de ceratopogonídeos. Além do fator climático, estudos sobre mosquitos picadores são escassos nesta região geográfica. O município de Pacoti, localizado na Serra de Baturité, no Ceará, é uma importante área de produção agrícola e ecoturismo; além disso, Pacoti está localizada na área de preservação ambiental. Devido à importância médica e veterinária desses maruins, foi então realizada uma pesquisa em Pacoti. Para a captura das amostras foram utilizadas armadilhas luminosas do tipo CDC em áreas florestadas is, onde as coletas foram realizadas em dois períodos: março de 2015 e agosto de 2022. Das 17 espécies encontradas nesse estudo, 12 são novos registros para o município de Pacoti, nove são novos registros para o estado do Ceará e quatro são registrados pela primeira vez para a região Nordeste. Das espécies vetoras incriminadas temos: *C. paraensis*, *C. insignis* e *C. pusillus*. O conhecimento da biodiversidade e a atualização da distribuição geográfica dos *Culicoides* poderão contribuir para medidas de controle de doenças transmitidas por vetores no Ceará.
Palavras-chave: *Comportamento antropofílico; Culicomorpha; Hematófagos; Nordeste; Vetores.*

Introduction

Culicoides is a small dipteran from the family Ceratopogonidae (Culicomorpha: Chironomoidea) with worldwide distribution. Due to their hematophagous habits, species of this genus are a nuisance to humans, causing acute allergic dermatitis in horses, and are proven or putative vectors of multiple pathogens of veterinary and public health importance, and can transmit protozoa, filarial worms, such as several arboviruses (Mellor et al. 2000; Borkent & Spinelli 2007). Worldwide, there are 1399 described *Culicoides* species of this genus, 301 of which are found in the neotropical region, 151 in Brazil, with most occurrences being recorded in the Amazon, with 123 species (Borkent & Dominiak 2020; Santarém & Felippe-Bauer 2023). The Northeastern region of Brazil is characterized by a semi-arid climate and few bodies of water, which does not favor breeding sites for ceratopogonids. In addition to the climatic factor, studies on Biting Midges *Culicoides* are scarce in this geographic region, where 37 species have been recorded for the Northeast. Currently, 12 species of *Culicoides* have been recorded in Ceará state (Santarém & Felippe-Bauer 2023).

In the Northeastern region of Brazil, certain mountainous areas adjacent to the coastline experience high humidity levels, fostering the growth of dense vegetation typical of the Atlantic Forest biome, characterized by its lush, humid environment, abundant with streams and rivers that contribute to its rich biodiversity. The humid mountain ranges of Northeastern Brazil, locally called 'altitudinal wetland forests', form islands of humidity and evergreen forests (humid forest) (Bétard et al. 2008). These humid areas in the Northeast are a center of endemism and the greatest richness of animal species in the region, e.g., Pessoa et al. (2005, 2012). The municipality of Pacoti, located in Serra de Baturité, in Ceará, is one of these areas of endemism and an important area for agricultural production and ecotourism; also, Pacoti is located in an environmental protection area (Ribeiro et al. 2022). Due to the public health importance of these biting midges, entomological collections for *Culicoides* was conducted in Pacoti and new occurrences of species are presented for the state and region.

Material and Methods

The *Culicoides* were collected in a forested area of Jordão farm 4°13'02"S 38°57'10"W 823 m, and of Museum station 4°13'35"S 38°55'19"W 742 m, Pacoti municipality, Baturité Highland, Ceará State, Northeast of Brazil (Figure 1), covering an area of 108.55 km², and 56.20% of its area being included in the Environmental Protection

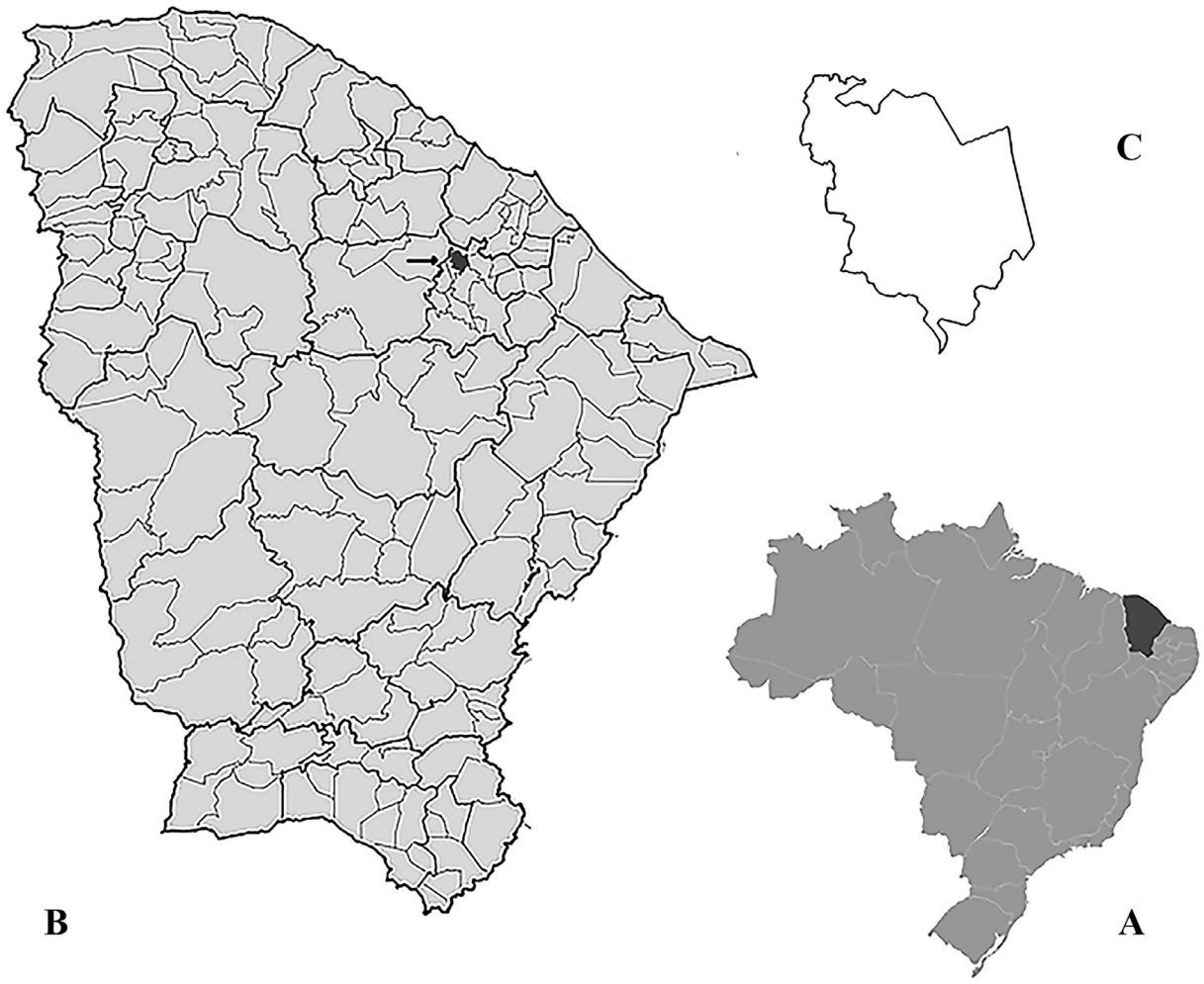

Figure 1. A. Map of Brazil; **B.** Ceará State; **C.** Pacoti municipality.

Area of Baturité (Oliveira et al. 2017). The municipal headquarters is located at the bottom of the valley, and it is characterized by intensive land use according to the layout of the relief. Its growth is accompanied by the expansion of occupation on steep slopes and the banks of the Pacoti River and tributary streams. The vegetation is characterized by humid forest enclaves in the semi-arid region that functioned as ecological refuges for flora and fauna, providing natural shelter for several endangered species and new species not yet described (Andrade-Lima 1982). They are characterized by having plants with Amazon, Atlantic, and Caatinga distribution (Santos et al. 2007).

To capture samples, two CDC-type light traps, during five night collections e used in a forests areas of Jordão farm and Museum station, these area are distanced 10 kilometers between then, during two periods, March 2015, and August 2022. Both sample areas in rural areas of Pacoti (Figure 2A–C). The samples were stored in 70% alcohol. The *Culicoides* were separated from the other families of dipterans specimens and separated into morphotypes. Then, the *Culicoides* were assembled following the methodology of Wirth & Marston (1968), where the specimen was dissected to be mounted on a slide and coverslip with phenol-balm (50%–50%). Subsequently, the insects were observed in detail under an optical microscope (Leica DM 1000, Frankfurt, Germany) for morphological identification. The wings were photomicrographed using a digital system (SynopticsTM, Cambridge, UK) coupled to the microscope. For identifying morphology, the mapping of structures was based on Wirth & Blanton (1959) and Spinelli et al. (1993); subgeneric classifications were based on Borkent (2015a, b). The specimens studied were housed in the Ecology Laboratory of Infectious Disease in Amazon, Leonidas and Maria Deane Institute (ILMD).

Results

A total of 527 *Culicoides* specimens were screened, 467 females (88.61%) and 60 males (11.39%) distributed into 17 species (Table 1): one for the subgenus *Avaritia* Fox, 1955 (*C. pusillus*); one for the subgenus *Diphaomyia* Vargas, 1960 (*C. freitasi*); one for the subgenus *Drymodesmyia* Vargas, 1960 (*C. poikilonotus*); six species for the subgenus *Haematomyidium* Goeldi, 1905 (*C. crucifer, C. darlingtonae, C. debilipalpis, C. martyrius, C. paraensis*, and *C. peruvianus*); three for the subgenus *Hoffmania* Fox, 1948 (*C. diabolicus, C. insignis*, and *C. lutzi*); one for the unplaced *fluvialis*, species group (*C. leopoldoi*); one for the subgenus unplaced, *leoni* species group (*C. gabaldoni*); one for the Subgenus unplaced, *limai* species group (*C. limai*); one for the subgenus unplaced, *reticulatus* species group (*C. pifanoi*); and an unidentified male *C.* sp1.

The most abundant species were *C. limai* (261 – 49,53%), followed by *C. diabolicus* (76 – 14,42%), *C. insignis* (42 – 7,97%), *C. pifanoi* (32 – 6.07%) and *C. paraensis* (31 – 5.88%). The others had an abundance of less than 5% (Table 1).

Species composition, presented by subgenera/group, were:

Subgenus *Avaritia* Fox

Avaritia Fox 1955: 218 (as subgenus of *Culicoides*). Type species: *Ceratopogon obsoletus* Meigen, by original designation

Figure 2. A – landscape of rural area of Pacoti municipality. **B** and **C** – Forest landscap of collection and CDC light trap installed for *Culicoides* collections in forested area of the study.

Table 1. List of species of *Culicoides* collected in Pacoti municipality, Baturité Hills, Ceará State, Northeast of Brazil.

Species	male	%	female	%	total	%
***C. crucifer* Clastrier	0	0.00	1	0.21	1	0.19
***C. darlingtonae* Wirth & Blanton	0	0.00	1	0.21	1	0.19
**C. debilipalpis* Lutz	6	10.00	2	0.43	8	1.52
***C. diabolicus* Hoffman	1	1.67	75	16.06	76	14.42
***C. freitasi* Wirth & Blanton	1	1.67	2	0.43	3	0.57
***C. gabaldoni* Ortíz	7	11.67	11	2.36	18	3.42
**C. insignis* Lutz	0	0.00	42	8.99	42	7.97
C. leopoldoi Ortíz	9	15.00	16	3.43	25	4.74
***C. limai* Barretto	13	21.67	248	53.10	261	49.53
***C. lutzi* Costa Lima	0	0.00	1	0.21	1	0.19
***C. martyrius* Trindade & Felippe-Bauer	0	0.00	1	0.21	1	0.19
C. paraensis (Goeldi)	11	18.33	20	4.28	31	5.88
***C. peruvianus* Felippe-Bauer	0	0.00	3	0.64	3	0.57
C. pifanoi Ortíz	8	13.33	24	5.14	32	6.07
**C. poikilonotus* Macfie	0	0.00	1	0.21	1	0.19
C. pusillus Lutz	3	5.00	19	4.07	22	4.17
C. sp1	1	1.67	0	0.00	1	0.19
TOTAL	60	11.39	467	88.61	527	100.00

(*) indicates that the taxon is reported for the first time in the Municipality of Pacoti, Ceará; (**) indicate that the taxon is reported for the first time in Ceará State, (***) indicate that the taxon is reported for the first time from the Northeastern region.

Culicoides pusillus Lutz

Culicoides pusillus Lutz (1913): 52.

Identification: Small size, bare; wing as shown in figure 3A, without marginal pale spots, the very short, entirely dark second radial cell, blackish mesonotum with two faints darker vittae and relatively pale. This species is an exception among the related species of the subgenus *Culicoides* (*Avaritia*) due to the apex of the very short second radial cell entirely dark or only very slightly encroached on by the poststigmatic pale spot (Wirth & Blanton 1959).

Distribution: Florida, USA to Central and South America to Northeastern Argentina and Brazil (Acre, Amapá, Amazonas, Bahia, Ceará, Minas Gerais, Pará, Rondônia, Rio de Janeiro, São Paulo, and Santa Catarina).

Material examined: 3 males and 19 females collected in Pacoti municipality, Baturité Highland, Ceará State, Northeast of Brazil; CDC light trap; 21.VIII.2022; collector FACP.

Subgenus *Diphaomyia* Vargas

Diphaomyia Vargas 1960: 40 (as subgenus of *Culicoides*). Type species: *Culicoides baueri* Hoffman, by original designation.

Culicoides freitasi Wirth & Blanton

Culicoides freitasi Wirth & Blanton 1973: 434.

Identification: Eyes narrowly separated, bare; third palpal segment moderately swollen, with a round deep sensory pit; wing as shown in figure 3B, and similar in pattern to *C. iriatei* Fox 1952, but differs due to two pale spots in the distal portion of the anal cell and only pale spot lying adjacent to the proximal portion of vein M_1; halter pale; two spermathecae (Wirth & Blanton 1973).

Distribution: Brazil (Amazonas, Ceará, and Pará).

New record: Ceará (Municipality of Pacoti).

Material examined: 1 male and 1 female collected in Pacoti municipality, Baturité Highland, Ceará State, Northeast of Brazil; CDC light trap; 21.VIII.2022; collector FACP. 1 female collected in Pacoti municipality, Baturité Highland, Ceará State, Northeast of Brazil; CDC light trap; 01.III.2015; collector FACP.

Subgenus *Drymodesmyia* Vargas

Drymodesmyia Vargas 1960: 40 (as subgenus of *Culicoides*). Type species: *Culicoides copiosus* Root & Hoffman, by original designation.

Culicoides poikilonotus Macfie

Culicoides poikilonotus Macfie 1948: 82.

Identification: Eyes contiguous above, bare; AR 1,26; palpal third segment short and swollen, PR 1.4 – 1.8, with a very deep, large sensory pit opening by a very small distal pore; mandible with 10 – 11 very small, almost vestigial teeth; wing as shown in figure 3C, pale spot over r-m crossvein very small but extending in full width to costal margin; Length of wing 0.76 – 0.92 mm; halter pale. The great reduction in the mandibles, as well as the wing pattern and antennal and palpal structure, ally *C. poikilonotus* with *C. panamensis* Barbosa, which, however, can readily be recognized by the pale spots straddling veins M_1 and M_2

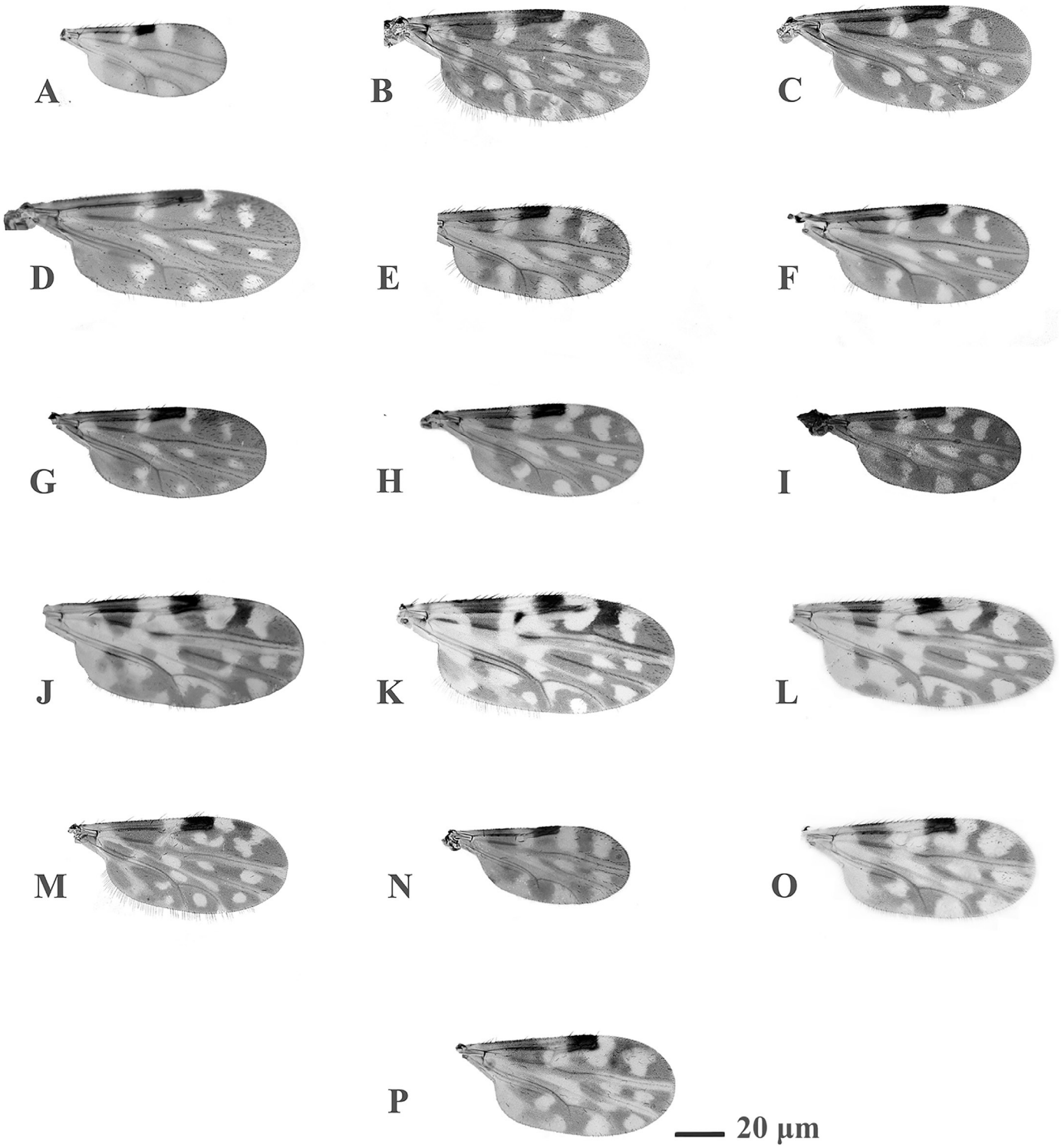

Figure 3. Wings of females of 17 species of *Culicoides* collected in Pacoti-Ceará. One for the subgenus *Avaritia*: **A.** *C. pusillus*; one for the subgenus *Diphaomyia*: **B.** *C. freitasi*; one for the subgenus *Drymodesmyia*: **C.** *C. poikilonotus*; six species for the subgenus *Haematomyidium*: **D**. *C. crucifer,* **E.** *C. darlingtonae,* **F.** *C. debilipalpis,* **G.** *C. martyrius,* **H.** *C. paraensis,* and **I.** *C. peruvianus*; three for the subgenus *Hoffmania*: **J.** *C. diabolicus,* **K.** *C. insignis,* and **L.** *C. lutzi*; one for the unplaced *fluvialis*, species group: **M.** *C. leopoldoi*; one for the subgenus unplaced, *leoni* species group: **N.** *C. gabaldoni*; one for the Subgenus unplaced, *limai* species group: **O.** *C. limai*; one for the subgenus unplaced, *reticulatus* species group: **P.** *C. pifanoi*.

and by the very unequal, retort-shaped spermathecae. Except for the difference in the extent of the pale spot in front of (*C. cacozelus*) or straddling (*C. poikilonotus*) the base of vein M_2, *C. cacozelus* Macfie seems to be identical with *C. poikilonotus* (Wirth & Blanton 1959).

Distribution: Mexico, Guatemala, Honduras (Lancetilla), Nicaragua (Villa Somoza), Panama, Venezuela, and Brazil (Bahia, Ceará, and Rio de Janeiro).

New record: Municipality of Pacoti (Ceará).

Material examined: 1 female collected in Pacoti municipality, Baturité Highland, Ceará State, Northeast of Brazil; CDC light trap; 01.III.2015; collector FACP.

Subgenus Haematomyidium Goeldi

Haematomyidium Goeldi (1905): 137. Type species: *Haematomyidium paraensis* Goeldi, by original designation.

Culicoides crucifer Clastrier

Culicoides crucifer Clastrier (1968): 85.

Identification: A large species; third palpal segment slender with broad shallow pit; wing as shown in figure 3D, with distal pale spot in cell m1 lying far from wing margin, no pale spot present in cell m_2 lying adjacent to midportion of mediocubital stem; aedeagus conspicuously cleft distad with two sharps laterally directed teeth on each side; parameres with simple twisted tips without fringing spines (Vitale et al. 1981).

Distribution: Brazil (Amazonas, Ceará, and Pará), French Guiana, Guyana, and Trinidad.

New record: Ceará (Municipality of Pacoti).

Material examined: 1 female collected in Pacoti municipality, Baturité Highland, Ceará State, Northeast of Brazil; CDC light trap; 21.VIII.2022; collector FACP.

Culicoides darlingtonae Wirth & Blanton

Culicoides darlingtonae Wirth & Blanton 1971: 39.

Identification: Eyes narrowly separated, with short interfacetal hairs; third palpal segment considerably swollen its full length, with shallow and round sensory pit; proboscis short; thorax dark brown; legs brown, knee spots blackish; wing as shown in figure 3E, second radial cell included in a dark spot to its apex, cell r_3 with contiguous poststigmatic pale spots, cell m_1 with two pale spots, cell m_2 with small pale spot; two spermathecae and rudimentary third, with sclerotized ring (Wirth & Blanton 1971).

Distribution: Costa Rica, Trinidad and Brazil (Ceará, and Rondônia)

New record: Ceará (Municipality of Pacoti).

Material examined: 1 female collected in Pacoti municipality, Baturité Highland, Ceará State, Northeast of Brazil; CDC light trap; 21.VIII.2022; collector FACP.

Culicoides debilipalpis Lutz

Culicoides debilipalpis Lutz 1913: 60.

Identification: Third palpal segment stout, PR 2.00– 2.60; flagellomere 8 longer than 9; wing as shown in figure 3F, with distal pale spot in cell m_1 separated from wing margin by a distance approximate to its length, macrotrichia distributed on distal two-thirds of wing, extending in at least two rows to base of cell m_2; halter brown (Spinelli et al. 2005).

Distribution: From the south of the United States in North America to Argentina, in Brazil (Acre, Amapá, Amazonas, Ceará, Goiás, Espírito Santo, Mato Grosso do Sul, Pará, Pernambuco, São Paulo, Santa Catarina, Rondônia, and Roraima).

New record: Municipality of Pacoti (Ceará).

Material examined: 1 male and 2 females collected in Pacoti municipality, Baturité Highland, Ceará State, Northeast of Brazil; CDC light trap; 21.VIII.2022; collector FACP.

Culicoides martyrius Trindade & Felippe-Bauer

Culicoides martyrius Trindade & Felippe-Bauer 2011: 62.

Identification: Wing as shown in figure 3G, this species easily distinguished from the other species of the subgenus *Haematomyidium* by the single poststigmatic pale spot in cell r_3, eyes bare and slightly separated, palpus moderately long with a shallow pit, fore-mid femur with a faint, irregular subapical pale bands, hind tibia without pale band in apical portion, unequal spermathecae. It has wing pattern similar to *C.* germanus Macfie, from which it can be distinguished by its slightly longer wing 0.89 mm (0.80 in *C. germanus*), greater PR 2.4 (2.0 in *C. germanus*), by the single post-stigmatic pale spot (double in *C. germanus*), fore-mid femur with subapical pale bands (dark femurs in *C. germanus*) and by the apical portion of hind tibia dark (with broad pale area in *C. germanus*) (Trindade & Felippe-Bauer 2011).

Distribution: Brazil (Ceará, and Pará).

New record: Ceará (Municipality of Pacoti).

Material examined: 1 female collected in Pacoti municipality, Baturité Highland, Ceará State, Northeast of Brazil; CDC light trap; 01.III.2015; collector FACP.

Culicoides paraensis (Goeldi)

Culicoides paraensis (Goeldi) 1905:137.

Identification: Third segment palpus longer and more slender, PR 2.1–2.8; wing as shown in figure 3H, with three pale spots in cell m_1, distal pale spot distinctly separated from the subapical pale spot; third rudimentary spermatheca shorter; sclerotized ring long and curved; male with paramere uniformly slender in midportion, with elongate lobe (Felippe-Bauer et al. 2003).

Distribution: USA to Argentina and Brazil (Acre, Amazonas, Pará, Rondônia, Roraima, Bahia, Ceará, Maranhão, Pernambuco, Mato Grosso do Sul, Epírito Santo, Minas Gerais, Rio de Janeiro, São Paulo, and Santa Catarina).

Material examined: 11 males and 20 females collected in Pacoti municipality, Baturité Highland, Ceará State, Northeast of Brazil; CDC light trap; 01.III.2015; collector FACP.

Culicoides peruvianus Felippe-Bauer

Culicoides peruvianus Felippe-Bauer, in Felippe-Bauer et al. 2003: 1054.

Identification: Wing as shown in figure 3I, wing length 0.73–0.86 mm; AR 0.68–0.78; sensillar pattern 1,6-8; mandible with 18 teeth; palpus brown with sensory pit; PR 2.0–2.4; P/H ratio 0.74–0.90; CR 0.53–0.60; halter pale; two sub-spherical, equal-sized spermathecae, plus a long rudimentary third and sclerotized ring. Can be distinguished from the other species of the *paraensis* group by the distal pale spot in cell r_3, which gradually extends towards the subapical pale spot and is often joined with it. Also, the equal-sized spermathecae with long, stout necks and a high P/H ratio (0.84) are all characteristics of the *paraensis* species group (Felippe-Bauer et al. 2003).

Distribution: Peru and Brasil (Ceará, Rio de Janeiro, and São Paulo).

New record: Ceará (Municipality of Pacoti).

Material examined: 3 females collected in Pacoti municipality, Baturité Highland, Ceará State, Northeast of Brazil; CDC light trap; 01.III.2015; collector FACP.

Subgenus *Hoffmania* Fox

Hoffmania Fox 1948: 21 (as subgenus of *Culicoides*). Type species: *Culicoides inamollae* Fox & Hoffman (=*Culicoides insignis* Lutz), by original designation

Culicoides diabolicus Hoffman

Culicoides diabolicus Hoffman 1925: 294.

Identification: Eyes contiguous by a distance equal to diameter of 1.5–2.5 ommatidial facets; antenna brown, bases of flagellar segments 3–10 pale; third palpal segment slender, slightly broad in middle, pit irregular; wing as shown in figure 3J, with r-m crossvein pale (faintly darkened in some topotypic specimens), one large, transverse, distal pale spot in cell r_3 broadly meeting wing margin, two distal pale spot in cell m1, the distal one smaller, apices of veins M_1 and M_2 pale, apices of veins CuA_1 and CuA_2 dark; halter pale; spermathecae subspherical to ovoid, unequal (Spinelli et al. 1993).

Distribution: Mexico to Ecuador and Brazil (Amazonas, Ceará, Rondônia, Roraima, and Maranhão).

New record: Ceará (Municipality of Pacoti).

Material examined: 1 male and 75 females collected in Pacoti municipality, Baturité Highland, Ceará State, Northeast of Brazil; CDC light trap; 21.VIII.2022; collector FACP.

Culicoides insignis Lutz

Culicoides insignis Lutz 1913: 51.

Identification: Eyes contiguous by a distance equal to the diameter of 1.5 ommatidial facets; third palpal segment with a definite, irregular pit; wing as shown in figure 3K, r–m crossvein dark on anterior half; vein CuA_2 dark up to the point where it turns abruptly forward to meet the costa; only one distal pale spot in cell m1; halter dark (Spinelli et al. 1993).

Distribution: USA, Mexico, Central America, and the Caribbean; Brazil (Alagoas, Amazonas, Pará, Bahia, Ceará, Maranhão, Paraíba, Pernambuco, Goiás, Mato Grosso, Mato Grosso do Sul, Espírito Santo, Minas Gerais, Rondônia, Rio de Janeiro, Rio Grande do Sul, Santa Catarina, and São Paulo).

New record: Municipality of Pacoti (Ceará).

Material examined: 42 females collected in Pacoti municipality, Baturité Highland, Ceará State, Northeast of Brazil; CDC light trap; 21.VIII.2022; collector FACP.

Culicoides lutzi Costa Lima

Culicoides lutzi Costa Lima 1937: 419.

Identification: Brown species; eyes contiguous by a distance equal to the diameter of 2–3 ommatidial facets; third palpal segment stout, with irregular pit; wing as shown in figure 3L, crossvein r-m pale, one transverse distal pale spot in cell r_3 reaching wing margin, only one distal pale spot in cell m1, apices of veins M_1, M_2 and CuA_1 broadly pale, apex of vein CuA_2 dark; halter knob pale (Spinelli et al. 1993).

Distribution: Colombia, French Guiana, Northeastern Argentina, and Brazil (Acre, Amazonas, Ceará, Pará, Rondônia, Roraima, Maranhão, Goiás, Mato Grosso, Minas Gerais, Rio de Janeiro, São Paulo, Paraná, and Santa Catarina).

New record: Ceará (Municipality of Pacoti).

Material examined: 1 female collected in Pacoti municipality, Baturité Highland, Ceará State, Northeast of Brazil; CDC light trap; 21.VIII.2022; collector FACP.

Subgenus unplaced, *fluvialis* species group

Culicoides leopolodoi Ortíz

Culicoides leopoldoi Ortíz 1951: 579.

Identification: Wing as shown in figure 3M, with pale spot on crossvein r–m extensive; pale spot present behind second radial cell, proximal to the poststigmatic pale spots; poststigamtic pale spots longitudinally aligned, posterior one smaller; distal pale spot in cell r_3 large, with narrow proximal extension; one spermatheca (Spinelli et al. 2005).

Distribution: Guatemala and Belize to Bolivia, northeastern Argentina, Trinidad, and Brazil (Amapá, Amazonas, Ceará, Pará, Rondônia, Roraima, Bahia, Ceará, Maranhão, Pernambuco, Espírito Santo, Minas Gerais, Rio de Janeiro, Rio Grande do Sul, and São Paulo).

Material examined: 9 males and 16 females collected in Pacoti municipality, Baturité Highland, Ceará State, Northeast of Brazil; CDC light trap; 01.III.2015; collector FACP.

Subgenus unplaced, *leoni* species group

Culicoides gabaldoni Ortíz

Culicoides gabaldoni Ortíz 1954: 221.

Identification: Eyes broadly separated, with long interfacetal hairs. AR 0.85–0.96. Palpal third segment moderately swollen, PR 1.7–2.1, with a small, verydeep sensory pit. Mandible with 10–15 teeth. Wing as shown in figure 3N, Length of wing 0.55–0.79 mm. Halter pale. Spermathecae two, ovoid, slightly unequal. In pinned material, females of *C. gabaldoni* are practically inseparable from *C. leoni* Barbosa and *C. glabellus* Wirth and Blanton. However, in *C. gabaldoni,* the legs are paler, and there may be a few more hairs at the apex of the wing, arranged in rows. *Culicoides leoni* differs from the other two in having an AR of only 0.75. Females of *C. leoni* and *C. glabellus* differ from *C. gabaldoni* in having only one spermatheca (Wirth & Blanton 1959).

Distribution: Venezuela; Guatemala; Honduras; Nicaragua; Panama and Brazil (Bahia, Ceará, Pará, Rondônia, Roraima, and São Paulo).

New record: Ceará (Municipality of Pacoti).

Material examined: 7 males and 11 females collected in Pacoti municipality, Baturité Highland, Ceará State, Northeast of Brazil; CDC light trap; 21.VIII.2022; collector FACP.

Subgenus unplaced, *limai* species group

Culicoides limai Barretto

Culicoides limai Barretto 1944: 99.

Identification: Small; the much broader third palpal segment; wing as shown in figure 3O, with distal pale spot in cell m1 broadly abutting wing margin, second radial wholly included in a dark spot, no pale spot straddling vein M_2, this vein usually dark to apex, apices of veins M_1, M_2 dark; subapical pale band on the hind femur and pale apex of the hind tibia; presence of a small ventral lobe on the male parameres (Wirth & Blanton 1959).

Distribution: El Salvador to Ecuador, northeastern Argentina, and Brazil (Amapá, Amazonas, Pará, Rondônia, Roraima, Maranhão, Mato Grosso do Sul, Minas Gerais, Rio de Janeiro, São Paulo, and Santa Catarina)

New record: Ceará (Municipality of Pacoti).

Material examined: 13 males and 248 females collected in Pacoti municipality, Baturité Highland, Ceará State, Northeast of Brazil; CDC light trap; 21.VIII.2022; collector FACP.

Subgenus unplaced, *reticulatus* species group

Culicoides pifanoi Ortíz

Culicoides pifanoi Ortíz 1951: 588.

Identification: Eyes narrowly separated, bare. AR 0.84–0.97. Palpal third segment moderately swollen, PR 1.6–1.9, with a large, shallow sensory pit. Mandible with 12–15 teeth. Wing as shown in figure 3P, pale spot over r-m crossvein extensive. Among those species with dark second radial cells and prominent mesonotal pattern and large distal pale spot in cell r_3 broadly meeting wing margin, this species is readily identified by the extensive pale margins to veins M_1 and M_2, which are connected to the pale spots in cell m_1, and in apex of cell r_3, with the distal spot in cell M_1 much smaller than its distance from the wing margin. Halter pale. Spermathecae two, ovoid, unequal (Wirth & Blanton 1959).

Distribution: Venezuela, Florida, Honduras (Lancetilla), Nicaragua (Villa Somoza), Panama, Trinidad, and Brazil (Pará, Bahia, Ceará, and Rio de Janeiro).

Material examined: 8 males and 24 females collected in Pacoti municipality, Baturité Highland, Ceará State, Northeast of Brazil; CDC light trap; 01.III.2015; collector FACP.

Discussion

Some *Culicoides* species found in Pacoti were found just in Brazil in the Amazon basin l- *C. crucifer*, *C. darlingtonae, C. diabolicus, C. freitasi* and *C. martyrius* (Santarém & Felippe-Bauer 2023). During the Quaternary period, the Brazilian region was covered by Amazon and Atlantic rainforests, forming a vast, interconnected, continuous forest (Morley 2000). However, this continuum underwent historical compression and fragmentation due to the influence of open areas (semi-arid formations), which emerged as a consequence of the Pleistocene glacial cycles (Haffer 1969; Gentry 1982). Additionally, the presence of the Baturité hills forest, encompassing the Pacoti municipality, has been identified as a microrefugia area capable of supporting rainforest habitats and harboring reminiscent fauna, explaining the continued presence of certain species within these regions. *Culicoides peruvianus*, collected in Pacoti, distributed in Amazonian Peru and the Atlantic Forest of Rio de Janeiro and São Paulo states (Santarém & Felippe-Bauer 2023) is another example.

Of the 17 species found, 12 are new records for the municipality of Pacoti, nine for the state of Ceará, and four are recorded for the first time in the Northeast region of Brazil. *Culicoides* species found in Pacoti, *C. gabaldoni*, *C. limai*, and *C. lutzi* are widespread in many areas of Brazil (Santarém & Felippe-Bauer 2023).

This study identified *C. limai* as the most abundant species in Pacoti, representing almost 50% of two specimens collected. This species ranges from El Salvador to Ecuador, northeastern Argentina and was registered in 11 Brazilian states, being a new record for the state of Ceará. Acccording Williams (1964) in Trindade, larval habitats of *C. limai* were found in edge of tidal drainage ditch, short grass, riparian forest streams and other environments. In Brazil, an example of *C. limai* was pointed out with L3 filaria in the mouthparts (Forattini 1957), and tailed with human appeal Trindade & Gorayeb (2010) observed this species biting humans.

It was collected in this the study approximately 8x more females than males. Most part surveys done with CDC light traps for *Culicoides*, usually is collected low densities of males (e.g. Farias et al. 2020, Carvalho et al. 2016); and probably this trap is not very effective of males of *Culicoides*.

Some species collected of *Culicoides* found in Pacoti exhibit anthropophilic behavior: *C. debilipalpis*; *C. insignis*; *C. leopondoi*; *C. limai*; *C. lutzi*; *C. paraensis*; *C. peruvianus*, *C. pusillus* (Aitken et al. 1975; Bermúdez 1986; Castellón & Ferreira 1991; Trindade & Gorayeb 2005, 2010, Felippe-Bauer et al. 2003), of which three are incriminated as vectors relevant to both humans and veterinary health, as notable carriers of certain etiological agents: *C. paraensis*, *C. insignis*, and *C. pusillus* (Mellor 2004).

Culicoides paraensis, is a species with wide distribution in the New World, occurring as far north as the southeastern United States to Uruguay (Salto) (Felippe-Bauer et al. 2003). This species has a daytime habit and is often found stinging humans. In Salvador, Bahia, it was considered a plague because the bites cause allergic dermatitis (Sherlock 1965). This species transmits the Oropouche Virus (OROV) to humans (Nunes et al. 2007). The OROV is transmitted through wild transmission and urban cycles, with the primary vector in the urban cycle being the anthropophilic biting midges *C. paraensis* (Sakkas et al. 2018). Humans likely serve as a bridge between wild and urban transmission cycles; upon entering forested areas, humans may become infected and subsequently return to urban settings during the viremic period. (Pinheiro et al. 1981). Since the early 1960s, OROV has been implicated in more than 30 epidemics (Sakkas et al. 2018). In Northeastern Brazil, OROV was detected in patients in Bahia (Fonseca et al. 2020). So far in 2024, four countries in the Region of the Americas will have reported cases of OROV: the Plurinational State of Bolivia, Brazil, Colombia, and Peru. In Brazil, OROV was detected in 2,104 samples, 1,821 in Amazonas, 172 in Rondônia, 51 in Acre and 12 in Roraima (PAHO/WHO 2024) However, it is only recently that OROV fever has attracted more research attention for reasons associated with climate change, the geographic expansion of arthropod vectors, the globalization of human and animal transport, and the emergence of other arboviral diseases (Sakkas et al. 2018). *Culicoides paraensis* has also been incriminated as a vector of *Mansonella ozzardi* Manson by Shelley & Coscarón (2001) in northern Argentina.

In Brazil, *C. insignis* and *C. pusillus* are present in all regions, with a wide distribution throughout the Brazilian states; of the 26 states, *C. insignis* is present in 17 and *C. pusillus* in 11, both species have been recorded in the state of Ceará. In studies by Rebêlo et al. (2016), *C. insignis* was found to be naturally infected with *Leishmania (Vianna) braziliensis* DNA in Northeastern Brazil. Experimental in vivo infections in *Culicoides* spp. demonstrated that biting midges can act as competent vectors for *Leishmania* (Kaewmee et al. 2023).

Culicoides insignis and *C. pusillus* are suspected vectors of the Bluetongue Virus (BTV) in South America (Mellor 2004), an importante problema to veterinary health. In Brazil, BTV is found throughout the national territory, from the extreme South to the North, with several surveys that address its seroprevalence (Scolari et al. 2011, Dorneles et al. 2012, Cunha-Filho et al. 2019). In the state of Ceará, BTV infections have been reported in ruminant populations, as evidenced by studies conducted by Brown et al. (1989), Silva (2002), and Pinheiro et al. (2013). Occurrences of clinical signs of Bluetongue in South America

occurred in 2001 and 2002 in Paraná (Clavijo et al. 2002; Lager 2004); later, in 2013, there was an outbreak of Bluetongue in Vassouras, Rio de Janeiro state (Balaro et al. 2014). It is caused by the association of the endemic with climatic conditions, with high temperatures and humidity that promote the proliferation of *Culicoides* biting midges (Cunha-Filho et al. 2019).

The low number of previously known *Culicoides* species in the Northeastern region of Brazil is probably due to the lack of studies in that state, which now has 21 recorded species, corresponding to 51% of the *Culicoides* species recorded for the Northeast of Brazil and 14% of the Brazilian fauna species of biting midges.

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Author Contributions

Emanuelle de Sousa Farias: contributed to screening and assembly of slides for morphological identification; contributed to taxonomic identification at species level; contributed to data analysis and manuscript preparation.

Carlos Rafael Lima de Souza: contributed to screening and assembly of slides for morphological identification; contributed to data analysis and manuscript preparation.

Rute Cristina Marques Costa: contributed to screening and assembly of slides for morphological identification; contributed to data analysis and manuscript preparation.

Sheila Patrícia Carvalho-Fernandes: contributed to data collection and manuscript preparation; contributed to data analysis and manuscript preparation.

Eddie William de Pinho Santana: contributed to data collection and manuscript preparation; contributed to data analysis and manuscript preparation.

Leonia da Conceição de Oliveira: contributed to data analysis and manuscript preparation.

Claudia María Ríos-Velásquez: contributed to data collection and manuscript preparation.

Felipe Arley Costa Pessoa: contributed to data collection and manuscript preparation.

Conflicts of Interest

The authors declare that they have no conflict of interest related to the publication of this manuscript.

Ethics

This study did not involve human beings and/or clinical trials that should be approved by any Institutional Committee.

Data Availability

Supporting data are available at < https://doi.org/10.48331/scielodata.8MUGMH>.

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