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Mosquito fauna (Diptera: Culicidae) of high Andean mountain ecosystems in Colombia

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Abstract

An annotated list of the mosquitoes (Diptera: Culicidae) known to occur in Colombia above 2,000 m is analyzed in present study. To study the mosquito fauna present in high Andean mountain ecosystems in the Departments of Antioquia and Quindío, sampling was carried out by rapid biodiversity assessment during May 2008 and September 2016. In addition, all records of Culicidae obtained from the scientific literature were included in the data analysis. As a pioneering work, this checklist updates the mosquito fauna above 2,000 m to a total of 35 species representing 11 genera and 15 subgenera. This list adds valuable information to the knowledge of biodiversity in Colombia and contributes to the establishment of a baseline for future research.

Keywords: Biodiversity, mosquitoes, mosquito fauna, highland, Colombia

Introduction

Colombia is known globally for its mega diversity, including mosquito fauna, with an estimation of 353 species [1]. High altitude sites are a specific category of ecosystem with a diversity of habitats such as lakes, ponds, rivers, water-containing plants [2]. In general, in Colombia, there is little information for this insect group over 2,000 m although there are some records of Diptera in three different ecological sites in the Parque Nacional Natural Chingaza at 3,200 m [3, 4], the Usme locality in Bogotá, between 3,080-3,315 m [5], and the Páramo Andino of Monserrate at 3,230 m [6], all sites in the Department of Cundinamarca. In the Parque Nacional los Nevados of the Caldas Department, some dipterans have been studied that occur at 4,000-4,400 m, the highest altitude recorded in Colombia [7]. However, the four records mentioned above do not include any specimens from the family Culicidae. Recently, one culicid morphospecies was collected in the municipality of Tenjo, Cundinamarca Department at 2,714- 2,765 m [8].

Specifically, for mosquitoes, in three localities of the Cundinamarca Department; Mosquera (2,516 m), Fusagasugá (1,728 m), and Girardot (269 m), specimens of *Culex* (*Culex*) *quinquefasciatus* Say, 1823 were collected for studies of development time, survival, mortality and life expectancy under laboratory conditions similar to their place of origin. Significant differences were found only in development time between Mosquera (15.05 days) and Girardot (7.72 days). Garcia *et al.* 2010 suggested that differences in life expectancy for a recently oviposited egg in Girardot (26.06 days), Fusagasugá (24.12 days) and Mosquera (40.77 days) demonstrate adaptation to lower temperatures at higher elevations [9]. Recently, *Aedes* (*Stegomyia*) *aegypti* (Linnaeus, 1762) was reported in the Bello municipality in the Antioquia Department, within a range in altitude from 1,882-2,659 m. Also, dengue virus was detected in *Ae. aegypti* collected in the same municipality at 1,984 m [10]. In another study, species of the tribe Sabethini were reported above 2,000 m in the northern Andean coffee-growing regions of Colombia [11]. Considering the new distribution records for mosquitoes and their importance, the objective of the present study was to compile a list of Culicidae in high altitude zones of the country to establish a baseline for future research with these insects. This paper presents the results of recent surveys of mosquitoes above 2,000 m in two Departments of Colombia, Antioquia and Quindío with a review of past records from publications and reports in the country.

Materials and methods

Study site and data collection: From May 2008 to September 2016 mosquito collections were made in six representative municipalities of Antioquia Department and one locality of Quindío Department. Elevation of the sampling sites ranged from 2,000-2,802 m. Mosquito sampling was based on the methodology proposed for rapid biodiversity assessment [12–14]. Mosquito larvae were found in several distinct microhabitats, such as phytotelmata, as well as other natural and artificial aquatic habitats. Adult mosquitoes were captured using Shannon traps, oral aspirators and insect nets. All collection sites in the field were geo-referenced. All field collections were transported to laboratories of the Programa de Estudio y Control de Enfermedades (PECET) at the Universidad de Antioquia and Insectary of Grupo de Investigación en Sistemática Molecular (GSMUN) at the Universidad Nacional de Colombia sede Medellín. Larval and pupal exuviae were obtained under laboratory conditions, following a standard protocol [15].

Data analysis: Specimens were identified using morphological keys and reviews available for Neotropical Culicidae [16–19]. For each genus and subgenus, we used keys as follows: *Aedes (Howardina)* [20]; *Aedes (Ochlerotatus)* [21]; *Anopheles (Kerteszia)* [22–24]; *Culex* [25–27]; *Johnbelkinia* [28], *Trichoprosopon* [28,29]; and *Wyeomyia (Nunezia)* [30,31]. Generic and subgeneric names of aedine mosquitoes follow the most recent classification proposed by Wilkerson *et al.* 2015 [32].

Identified specimens were deposited in the Entomological Museum Francisco Luis Gallego-MEFLG at the Universidad Nacional de Colombia, Medellín campus, collection codes NC36977-NC38176. In addition, mosquito records were obtained from publications and reports located *via* a review of relevant literature. This literature survey was completed in July 2017.

Results

A total of 1,200 specimens were collected, representing eight genera and 10 subgenera. The most diverse tribe was Culicini (493 specimens) followed by Aedini (479 specimens). This study in addition to literature records is the first checklist of mosquitoes above 2,000 m in Colombia with at least 35 species belonging to 11 genera and 15 subgenera. Previously, based only on published records, 11 genera, 11 subgenera and 30 species had been recorded. The mosquito list from high Andean mountain ecosystems in Colombia provided below is based on records from 60 collection sites in seven Departments between 2,000-3,250 m: Antioquia, Caldas, Cundinamarca, Huila, Nariño, Quindío and Santander (Figure 1). The lowest altitude was for *Aedes (Ochlerotatus) euiris* (Dyar, 1922), *Aedes* sp. and *Culex* Subgenus *Culex* in Chipaque at 2,000 m and the highest for *Ae. (Och.) euiris* in Choachí at 3,250 m, both municipalities in the Cundinamarca Department [33]. Species from the tribes Sabethini and Uranotaeniini have been collected only as high as 2,977 m and 2,700 m respectively (Table 1).

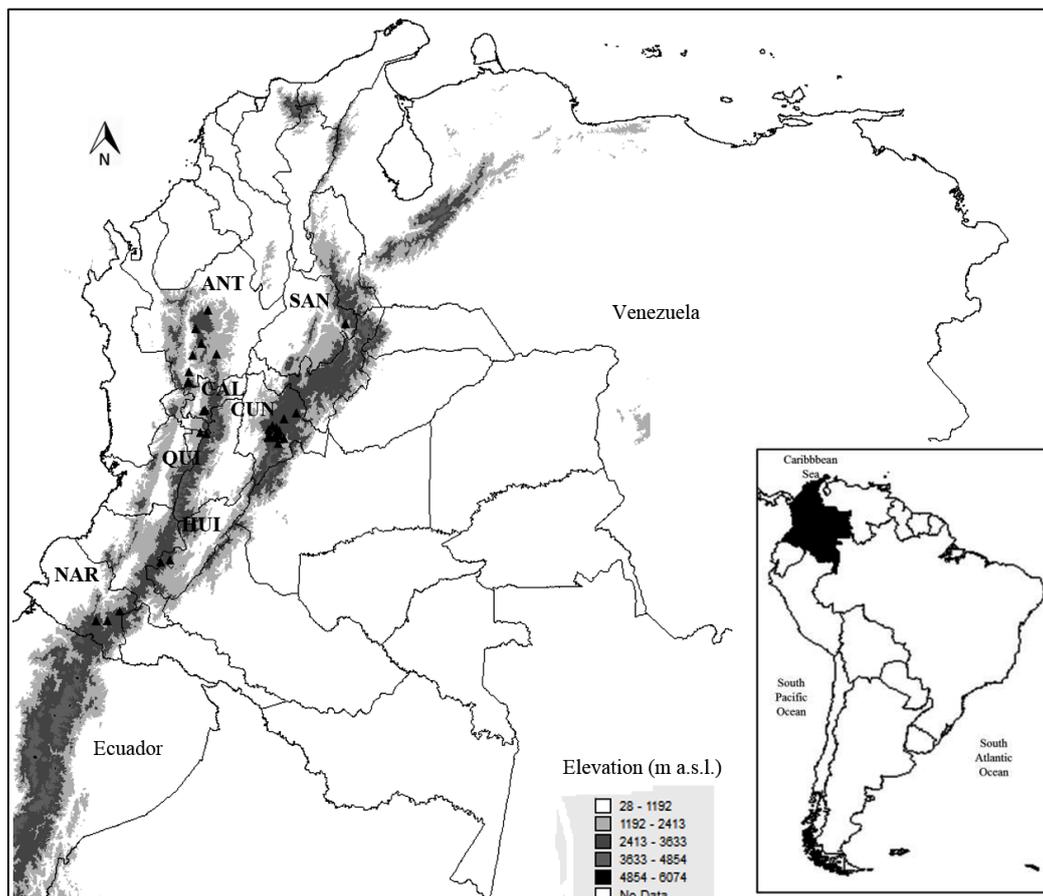


Fig 1: Map of sites for mosquito records above 2,000 m in Colombia. ANT: Antioquia, CAL: Caldas, CUN: Cundinamarca, HUI: Huila, NAR: Nariño, QUI: Quindío, SAN: Santander.

Some immature specimens collected in rock holes in Bogotá at 2,650 m, and adult females biting humans in partial forest in Chipaque at 2,000 m, included in the present list, were not available for the Project “Mosquitoes of Middle America” [33]. Records with changes in nomenclature are depicted by*.

Subfamily Anophelinae

1. *Anopheles (Anopheles) pseudopunctipennis* Theobald, 1901
2. *Anopheles (Kerteszia) Boliviensis* Group (Navarro *et al.* 2010)
3. *Anopheles (Lophopodomyia) oiketorakras* Osorno-Mesa, 1947*
4. *Anopheles (Nyssorhynchus) argyritarsis* Robineau-Desvoidy, 1827

Subfamily Culicinae

Tribe Aedini

5. *Haemagogus* sp.
6. *Aedes (Howardina)* sp.
7. *Aedes (Howardina)* sp. Eleanorae Group (Berlin, 1969)
8. *Aedes (Howardina) eleanorae* (Berlin, 1969)
9. *Aedes (Howardina) leei* (Berlin, 1969)
10. *Aedes (Howardina) marinkellei* (Berlin, 1969)
11. *Aedes (Howardina) marinkellei/ osornoi* hybrid (Berlin, 1969)
12. *Aedes (Howardina) osornoi* (Berlin, 1969)
13. *Aedes (Howardina) quadrivittatus* (Coquillett, 1902)*
14. *Aedes (Ochlerotatus)* sp.
15. *Aedes (Ochlerotatus) bogotanus* (Arnell, 1976)*
16. *Aedes (Ochlerotatus) deficiens* (Arnell, 1976)*
17. *Aedes (Ochlerotatus) euiris* (Dyar, 1922)*
18. *Aedes (Ochlerotatus) milleri* (Dyar, 1922)*
19. *Aedes* sp.
20. *Aedes (Stegomyia) aegypti* (Linnaeus, 1762)

Tribe Culicini

21. *Culex* sp.
22. *Culex (Anoedioparpa) conservator* Dyar & Knab, 1906
23. *Culex* subgenus *Carrollia*
24. *Culex (Carrollia) bihaicola* Dyar & Nuñez Tovar, 1928*
25. *Culex (Carrollia) secundus* Bonne-Wepster & Bonne, 1920
26. *Culex* subgenus *Culex*
27. *Culex (Culex) acharistus* Root, 1927
28. *Culex (Culex) coronator* Dyar & Knab 1906
29. *Culex (Culex) chitae* Duret, 1967
30. *Culex (Culex) quinquefasciatus* Say, 1823
31. *Culex* Subgenus *Microculex*
32. *Culex (Microculex) aureus* Lane & Whitman, 1951
33. *Culex (Microculex) imitator* Theobald, 1903
34. *Culex* Subgenus *Neoculex*

Tribe Orthopodomyiini

35. *Orthopodomyia* sp.

Tribe Sabethini

36. *Johnbelkinia* sp.
37. *Sabethes* sp.
38. *Sabethes* Subgenus *Sabethoides*
39. *Shannoniana fluviatilis* (Theobald, 1903)
40. *Trichoprosopon* sp.

41. *Trichoprosopon evansae* Antunes, 1942
42. *Trichoprosopon pallidiventer* (Lutz, 1905) s.l.
43. *Trichoprosopon* sp. A (Suaza-Vasco *et al.* 2015)
44. *Trichoprosopon* sp. B (Suaza-Vasco *et al.* 2015)
45. *Wyeomyia* sp.
46. *Wyeomyia* aff. *phroso* Howard, Dyar & Knab 1915
47. *Wyeomyia* Subgenus *Nunezia*
48. *Wyeomyia (Nunezia)* sp. A (Suaza-Vasco *et al.* 2015)
49. *Wyeomyia (Nunezia)* sp. B (Suaza-Vasco *et al.* 2015)

Tribe Uranotaeniini

50. *Uranotaenia* Subgenus *Uranotaenia*
51. *Uranotaenia riverai* Duret, 1970

Some relevant aspects of general distribution, main characters used in the morphological identification, altitude records and ecological or health importance of these species are described below and summarized in Table 1. For remarks on the identification of mosquitoes reported in the literature, we recommend checking the original paper in which the species were reported.

1. *Anopheles pseudopunctipennis*: This species is recognized as a complex of at least three species, and it is an important vector of malarial plasmodia in the foothills and mountainous regions of the Andean countries of South America [34–36]. In Bolivia, for example, this species has always been found in exposed, clear, slow-moving water, almost always associated with filamentous algae. Above 1,000 m *An. pseudopunctipennis* is the only species considered to be a vector of malarial parasites in Bolivia and is a priority in control efforts [37]. In Colombia, is a secondary vector of *Plasmodium* spp. [38, 39] and has been reported above 2,000 m in the Department of Caldas [40].

2. *Anopheles (Kerteszia) sp.*: The specimen reported here appears to be a species of the Boliviensis Group, which includes *Anopheles boliviensis* (Theobald, 1905), *Anopheles gonzalezrinconesi* (Cova García, Pulido F. & Escalante de Ugueto, 1977) and *Anopheles rollai* (Cova García, Pulido F. & Escalante de Ugueto, 1977). These species have been reported in sympatry in high altitude areas associated with forest bromeliads at 2,232 m in the Parque Nacional Dinira in Venezuela [41–43]. Currently, the three species cannot be separated using morphological characters [23]; moreover, the adult male and immature stages of *An. boliviensis* are unknown and the adult male of *An. rollai* has not been completely studied [23, 24, 44]. Studies of this group in Colombia are uncommon, old and questionable, with some records of *An. boliviensis* in the Departments of Meta, Huila, Tolima, Cundinamarca, Caldas and Chocó [22]. *Anopheles boliviensis* is the only species recorded in Colombia and it is considered to be a seasonal vector [38, 39, 45]. This species was detected infected with Anopheles A and Anopheles B viruses in Horizonte, Villavicencio of the Meta Department [46] and carrying the eggs of *Dermatobia hominis* (Linnaeus, 1781), a large fly that is important in public health [47]. Recently, in the Antioquia Department third- and fourth-instar larvae of Boliviensis Group were collected in forest bromeliads at 2,600 m in the Reserva forestal El Romeral, Angelópolis [48]. Additionally, adult females were collected by Shannon trap in Jardín at 2,400 m [49] and entomological nets in Páramo de

Santa Inés, Belmira at 2,977 m^[50], both municipalities in the Antioquia Department. In the present study, six larvae were collected in bromeliads at 2,521 m; nine females were collected by Shannon trap at 2,403 m and 15 females by oral aspirator at 2,100 m in Jardín. In addition, one larva was collected in a bromeliad at 2,755 m in Yarumal in the Antioquia Department. The characters for identification of these specimens were spots on the palpi, legs and wings, as well the general coloration of the adult females, abdominal terga II-VII with numerous dark decumbent scales and sterna with few white scales^[23, 24].

3. *Anopheles oiketorakras*: Type-loc: Bogotá, Cundinamarca, Colombia (USNM-United States National Museum, now National Museum of Natural History; Smithsonian Institution; Washington, DC). This species, known only from Colombia, was described from larvae collected in la hoya hidrográfica del río San Francisco, a mountainous region at east of Bogotá between 2,700-2,840 m^[51]. The habitats for immature stages are shaded stream pools rich in organic matter^[51]. The other record of this species is from Túquerres at 3,104 m in the Nariño Department^[22]. Recently, two females were collected in the Páramo de Santa Inés, Belmira employing a CDC trap at 2,862 m^[50].

4. *Anopheles argyritarsis*: In Bolivia, this species shares the same geographical distribution as *An. pseudopunctipennis* above 1,000 m^[37]. Although the exact role of this species in the transmission of *Plasmodium* parasites is unclear^[52], some publications reported that it may contribute as a vector when present at high densities^[53]. In the Nariño Department, this species was collected in a pond along a road in Funes at 1,720 m, and the highest record is at 2,800 m in the Laguna de la Cocha, Pasto^[33]. Other records for this species above 2,000 m are described in table 1 and correspond to the Departments of Antioquia and Caldas^[40].

5. *Haemagogus* sp.: Some species of this genus are vectors of the sylvan cycle of yellow fever virus^[54-56]. Of the eight species recorded from Colombia, six are lowland species and two could be highland species^[54, 57-59]. *Haemagogus (Haemagogus) andinus* Osorno-Mesa, 1944 and *Haemagogus (Haemagogus) equinus* Theobald, 1903 are known to occur at 1,746 m in a coffee plantation near Fusagasugá, in the Cundinamarca Department^[54]. Recently, species of *Haemagogus* were collected from bromeliads in two localities in the Cundinamarca Department above 3,000 m^[60, 61].

6. *Aedes (Howardina)* sp.: A total of 35 species of this subgenus were recognized: five species in one group in the Fulvithorax Section; 11 species in five groups in the Walkeri Section; and 19 species in six groups in the Sexlineatus Section, which is primarily associated with the mountain regions of Central America and western South America^[20]. Little information is available on the medical importance of species of *Ae. (Howardina)*, and more investigations should be conducted to understand the actual role of these species as vectors^[20]. In a recent study, in the Antioquia Department, 83 specimens were collected in the Páramo de Santa Inés, Belmira between 2,734-3,098 m using different collecting methods and 33 females were identified as *Ae. (How.) aff. brevis/spinosus*^[50]. In addition, a total of 230 specimens were

identified as *Ae. (Howardina)* sp. and were collected from bromeliads: three larvae, 10 females and 10 males between 2,704-2,802 m in the Reserva Ecológica Cuchilla Alto El Romeral, Angelópolis and 23 larvae, 48 females and six males between 2,092-2,521 m in Jardín. Also, adult females were collected using different methods: entomological nets at 2,253 m (10 females), oral aspirator at 2,100 m (three females) and Shannon trap between 2,251-2,403 m (108 females) in Jardín. Only one larva was collected from a bromeliad at 2,173 m in Támesis. Lastly, seven specimens were collected from bromeliads (five larvae, two males) at 2,755 m in Yarumal and two females were collected by oral aspirator at 2,100 m in Jardín that were identified as belonging to the Fulvithorax Section. In the adults, the most important taxonomic characters pertain to the ornamentation of the mesonotum, which usually consists of lines of pale scales superimposed on a background of dark scales. The fourth-instar larvae show many taxonomic characters at the section and group levels that are usually concordant with those of adult ornamentation, and many reliable specific differences^[20].

7. *Aedes (Howardina) Eleanorae* Group: This group includes species that are difficult to identify based on morphological characters: *Ae. (How.) marinkellei* is morphologically similar to *Ae. (How.) osornoi* and *Ae. (How.) leei*. Although Berlin (1969) mentions the possibility of a natural hybrid between these two species have not been collected together in the same bromeliad^[20]. Other species of this group include *Ae. (How.) eleanorae*, *Ae. (How.) ecuadoriensis* and *Ae. (How.) brevivittatus*. Recently, 30 specimens were identified as Eleanorae Group that were collected at 2,734-3,098 m using different collecting methods in Páramo Santa Inés, Belmira, in the Antioquia Department^[50]. In addition, 18 females morphologically identified as members of this group were collected as follows: one specimen from a bromeliad at 2,802 m in the Reserva Ecológica Cuchilla Alto El Romeral, Angelópolis and other specimens by oral aspirator at 2,100 in Jardín. The main morphological characters in adults are based on usually creamy to golden lines of scales on the mesonotum, except in *Ae. (How.) ecuadoriensis* in which inner dorsocentral and lateral marginal lines are white. In larvae, seta 15-C is usually single or double, moderate in length, reaching the level of the middle of the dorsomentum^[20].

8. *Aedes (Howardina) eleanorae*: Type-loc: Buriticá, Antioquia, Colombia (USNM). This species, known only from Colombia, is found in epiphytic bromeliads at elevations around 1,524 m. At present it is known from Buriticá in the northern part of the Cordillera Occidental, Buena Vista in the upper part of the Orinoco basin on the eastern slope of the Cordillera Oriental^[20] and adult females collected in Páramo de Santa Inés, Belmira between 2,734-2,862 m^[50].

9. *Aedes (Howardina) leei*: Type-loc: Puracé, Huila, Colombia (USNM). The larvae and adults of this species are known from bromeliads and human landing collections, respectively, from the type locality at 2,926-3,100 m^[20, 33]. Another definite record is a single adult female from Zongo near La Paz, Bolivia and probably this species will eventually be found in areas between Ecuador and Perú^[20].

10. *Aedes (Howardina) marinkellei*: Type-loc: Suba, Bogotá, Cundinamarca, Colombia (USNM). This species is known to occur only in Colombia. Immature stages have been found in bromeliads at elevations between 2,580-2,650 m^[20, 33]. Adult females of this species are apparently at least occasionally attracted to humans^[20].

11. *Aedes (Howardina) marinkellei/osornoi* hybrid: A single reared adult female of *Ae. (How.) marinkellei* collected in the Cundinamarca Department at 2,650 m^[20, 33]. This species exhibits features of *Ae. (How.) osornoi* in adult ornamentation and in the immature stages, suggesting hybridization between the species. Although found in the same general locality, *Ae. (How.) osornoi* and *Ae. (How.) marinkellei* have not been collected together in the same bromeliad axils^[20].

12. *Aedes (Howardina) osornoi*: Type-loc: Chorro de Padilla, Rio San Francisco, Bogotá, Cundinamarca, Colombia (USNM). The known larval habitats of this species are epiphytic bromeliads above 2,650 m^[20, 33]. Remarkably, three females and one male identified as *Ae. (How.) aff. osornoi* and two larvae as *Ae. (How.) osornoi* were collected in the Páramo de Santa Inés, Belmira between 2,734-3,098 m^[50]. In addition, 13 females were collected by oral aspirator at 2,100 m in Jardín and were identified as *Ae. (How.) aff. osornoi*. In general, the morphological characters were compatible with *Ae. (How.) osornoi*, but all mesonotal golden lines were thicker than in *Ae. (How.) osornoi* and *Ae. (How.) eleanorae*.

13. *Aedes (Howardina) quadrivittatus*: Apparently, the larval habitats of this species are exclusively epiphytic bromeliads above 1,219 m^[20]. Adult females of this species are attracted to humans. An unidentified arbovirus was isolated from specimens collected in Panamá^[62]. Specimens of this species were collected above 2,700 m in Bogotá^[51].

14. *Aedes (Ochlerotatus) sp.*: Co-evolutionary relationships have been suggested between some species of the *Ochlerotatus* and Orthobunyavirus in North America^[63]. *Aedes (Ochlerotatus) scapularis* (Rondani, 1848) is a potential vector of microfilariae and arboviruses in the southeastern region of Brazil^[64]. However, in a recent study it was not possible to find a statistically significant effect of the Orthobunyavirus-*Ochlerotatus* pairings on experimental eVT rates^[65]. Recently, one female was collected at 2,862 m by oral aspirator in the Páramo de Santa Inés, Belmira^[50]. In the present study, a total of 152 specimens of *Aedes (Ochlerotatus)* species were collected by oral aspirator (one female) and in a single rock pool at 2,100 m in Jardín. In addition, seven larvae were collected in an artificial container at 2,799 m in Yarumal. Adult females of *Ae. (Ochlerotatus)* were recognized from specimens of other subgenera of *Aedes* by the basal bands on the abdomen and the color pattern formed by scales covering the mesonotum. Other characters for both sexes were the narrow decumbent head scales in a rather broad median patch and the erect scales usually numerous and extending onto the disc of the vertex. For larvae, the most important characters were the complete saddle and a ventral brush with seven to nine pairs of setae; siphon usually over twice as long as its breadth at the base^[17, 66]. In most cases, *Aedes* mosquitoes share a combination of morphological features and are best separated at the species level^[67].

15. *Aedes (Ochlerotatus) bogotanus*: Type-loc: Ogamora, Soacha, Cundinamarca, Colombia (USNM) at 2,400 m. This species is known only from the vicinity of Bogotá in Soacha at 2,400 m and in Usaquen at 2,640 m^[21, 33]. Larvae were collected in a small ground pool in a garden with abundant vegetation and a volcanic rock hole in a pasture, with temporary water and without vegetation^[21, 33].

16. *Aedes (Ochlerotatus) deficiens*: Type-loc: Suba, Cundinamarca, Colombia (USNM). This species is only known to occur in Colombia. It has been collected only at 2,580 m^[21, 33].

17. *Aedes (Ochlerotatus) euiris*: Type-loc: Bogotá, Cundinamarca, Colombia (USNM). In Venezuela, this species was collected as an adult in Páramo Batallón and La Negra at 3,133 m^[41, 68, 69]. In Colombia, two females collected in Bogotá between 2,700-2,840 m in a Magoon trap^[51] and one female collected in Belmira at 3,200 m in a CDC light trap^[70] were originally identified as *Ae. (Och.) euiris*. This species has the distinction of having been collected at the highest altitude in Colombia, at 3,250 m^[33], and in Venezuela at 3,133 m in the Páramo La Negra, Táchira State^[41].

18. *Aedes (Ochlerotatus) milleri*: Type-loc: Bogotá, Cundinamarca, Colombia (USNM). In Ecuador, this species was collected in El rio Tomebamba, Cuenca, Provincia del Azuay at 2,600 m in ponds with clear water and abundant *Spyrogirae* algae^[71]. In Colombia, 54 females were collected in Bogotá between 2,700-2,840 m in a Magoon trap and were identified as *Aedes milleri*^[51].

19. *Aedes sp.*: The immature stages of *Aedes* species are typically found in natural and artificial containers. Other typical habitats are tree holes and small amounts of water contained in dead and fallen plant parts^[72]. In Chipaque, specimens of *Aedes* species were collected in tree holes near a stream in a domestic area at 2,000 m^[33]. Recently, 30 larvae were collected from two tires (11 larvae) and a clothing washing pond (19 larvae) in Jericó in the Antioquia Department at 1,960 m. The following combination of characters were used to identify larvae of *Aedes sp.*; metathoracic pleural group of setae and siphon well developed and a single pair of seta 1-S inserted near or beyond the middle of the siphon^[17, 73].

20. *Aedes (Stegomyia) aegypti*: This species is an important vector in the transmission of dengue, yellow fever, and chikungunya viruses^[73,74]. It has not been recorded above 2,000 m in Venezuela, an altitude considered to be a theoretical barrier to the transmission of dengue virus^[41]. However, in Mexico, it was commonly encountered up to 1,700 m and there are rare records from 1,700-2,130 m^[75]. In Costa Rica, *Ae. aegypti* can be found up to 2,600 m^[76] and in Colombia the highest altitude registered was 2,200 m in Málaga, Santander^[77]. Recently, a study in the municipality of Bello in the Antioquia Department, increased the altitudinal range of *Ae. aegypti* from 2,252 to 2,302 m, previously unrecorded heights in Colombia^[10]. Notably, for the first time in Jericó, 22 specimens of *Ae. aegypti* were collected by oral aspirator (six females) and in tires (eight females and eight males) at 1,960 m. The main characters for morphological

identification are: clypeus with white scales, midfemur with an anterior white stripe from base to tip, and mesonotum with a lyre-shaped pattern of silvery scales [17].

21. *Culex* sp.: Larvae occur primarily in semi-permanent or permanent bodies of ground water, but many species are found exclusively in leaf axils, tree holes, rock-holes and crab-holes. Some species utilize artificial containers as well as the normal ground-water habitats. Several species of the subgenera *Culex* and *Melanoconion* are of medical importance [73]. Recently, three specimens identified as *Culex* species were collected at 2,862 m in the Páramo Santa Inés, Belmira [50]. In the present study, a total of 120 specimens were identified as *Culex* species collected as follows: in bromeliads at 2,092-2,481 m (23 larvae, three reared to obtain two females and one male) in Jardín; in leaf axils of *Xanthosoma* (one male) at 2,009 m; inside tires (95 larvae, 63 reared to obtain 37 females, 26 males) at 1,960 m in Jericó; and in one bromeliad at 2,642 m (eight larvae, three females and five males) in Salento.

22. *Culex (Aneodiopora) conservator*: This species is frequently found in tree holes and bamboo internodes in tropical areas far from forests [78]. Only one larva was identified as *Culex* aff. *conservator* and it was collected in a bromeliad at Jardín at 2,092 m. The diagnostic characters were lateral abdominal setae single on segments III to V [26]. Considering the single larva collected, future collections should be conducted to confirm this record.

23. *Culex* Subgenus *Carrollia*: The immature stages are found commonly in natural containers; however, some species can also be found in artificial containers. The habitats most frequently used by species of this subgenus are broken bamboo and tree holes [16, 27]. This subgenus is probably the most distinctive subgenus of *Culex* and includes the only species in the entire genus with conspicuous metallic ornamentation in the adults [27]. Species of this subgenus are not of medical importance to humans [73]. The abdomen with iridescent basolateral markings was particularly useful for the identification of two specimens (two larvae reared to obtain one female and one male) collected as larvae in the leaf axil of *Xanthosoma* at 2,100 m in Jardín.

24. *Culex (Carrollia) bihaicola*: This species utilizes an extremely wide range of aquatic habitats [27]. Some larvae of this species were collected in artificial containers in “El Chorro” at Sierra de San Luis at 1,200 m in Venezuela [79]. In Colombia, the only occurrence records published are from Anorí, Antioquia Department at 1,535 m and the Rio Raposo, Valle del Cauca Department, at low altitude from metal boxes [80]. Interestingly, specimens of *Cx. bihaicola* were collected in a palm-leaf (10 larvae reared to obtain three females) and in a tire (13 larvae, 10 reared to obtain three females and seven males) from Jardín at 2,526 m and in Salento at 2,095 m respectively. Adult characters [27]: erect scales of vertex of head yellowish to white and abdominal sterna entirely pale-scaled. Male genitalia: distal division of subapical lobe of gonocoxite relatively broad and with two specialized setae with recurved apex. Fourth-instar larvae: head seta 6-C single.

25. *Culex (Carrollia) secundus*: Larvae do not appear to occupy a wide range of aquatic habitats and have been found primarily in cut or broken bamboo, fallen leaves and palm spathes [27]. Particularly, in Colombia this species has been recorded in the Departments of Caldas, Huila, Meta, Valle del Cauca and Antioquia at elevations between 15-1,700 m [33]. In addition, there are some records of *Cx. secundus* at 1,500 m collected in fallen and broken bamboo in a forest of Manizales in the Caldas Department [33]. Recently, five specimens (five larvae reared to obtain one female and four males) were collected in one bromeliad at 2,173 m in Támenes in the Antioquia Department. Morphological characters [27]: midlobe of scutellum with linear dark scales; mesonotal vestiture predominantly dark bronzy; male with a single pair of long subapical; maxillary palpus of male entirely dark-scaled.

26. *Culex* Subgenus *Culex*: Specimens of this subgenus were collected in bromeliads in the coffee plantation “Finca La Esperanza”, Iles municipality on the road between Pasto and Ipiales at 1,900 m [33]. Recently, *Culex stenolepis* (Dyar & Knab 1908) was the first species found in a ground pool within, and downslope from, the crater of the Pululahua Volcano at 2,101 m, 80 km from the capital Quito, in Ecuador [81]. In Colombia, the maximum altitude recorded for this species is in Bogotá at 3,150 m [33] and in Venezuela, six species belonging this subgenus were reported above 2,000 m [41]. In the present study, specimens were identified principally by the presence/absence of a few salient anatomical features of the adults based on characters available in Forattini 2002 [16]. A total of 17 specimens belonging this subgenus were identified as *Cx. aff. habilitator*. They were collected in bromeliads at 2,092 m (three larvae) and 2,416 m (17 larvae, five reared to obtain two females and three males) in Jardín and Salento respectively. In addition, six species were identified as species 1, 3-7. Further studies are necessary each species the exact taxonomic status assignment of each species.

27. *Culex (Culex) acharistus*: Eighty-four larvae, 75 reared to obtain 40 females and 35 males, were morphologically identified as *Cx. acharistus* and were collected exclusively from cattle footprints between 2,576-2,642 m in Salento. The main morphological characters for larvae, adult females and male genitalia are based on the description of this species provided by Forattini 2002 [16].

28. *Culex (Culex) coronator*: In Brazil, Saint Louis encephalitis virus has been isolated from *Cx. coronator* [82] and in Florida, USA, this species is a competent vector of West Nile virus under certain conditions, such as high temperatures [83]. Most recently, 15 specimens were identified as *Cx. coronator* collected from one bromeliad (one female) and an animal drinking bowl (14 larvae, five reared to obtain three females and two males) at 2,092 m in Jardín. The main character in larvae was the siphon with strong apical spines. This species differs from other members of the Coronator Complex by the arrangement and number of setae on the subapical lobe of the gonocoxite in the male genitalia [25].

29. *Culex (Culex) chitae*: Type-loc: Monserrate, nr. Bogota, Colombia (USNM). The only information about this species is that two males were collected in the type locality at 2,900 m. Adult female, larval and pupal characters are unknown [84].

30. *Culex (Culex) quinquefasciatus*: This species is recorded above 2,000 m from various municipalities of the Cundinamarca Department [9, 85-87]. *Culex quinquefasciatus* is an urban problem with health importance in Bogotá and its surroundings [88]. In its natural environment, larvae develop in polluted ponds rich in organic matter [86, 89]. In the list of mosquitoes above 2,000 m in Venezuela, two records exist for this species at 2,000 and 2,327 m [41]. A total of 119 specimens *Cx. quinquefasciatus* were collected in bromeliads (84 larvae, 46 reared to obtain 17 females and 29 males) and one tire (35 larvae, 29 reared to obtain nine females and 20 males) in Jardín at 2,092-2,317 m and one tire in Salento at 2,019 m.

31. *Culex* Subgenus *Microculex*: The immature stages inhabit mainly epiphytic bromeliads. Moreover, some specimens have also been found less frequently in tree holes and bamboo internodes. Species of this subgenus are not of medical importance to humans [16, 73]. *Culex (Microculex) daumastocampa* Dyar & Knab, 1908 was reported at 2,550 m in Venezuela [41]. The subgenus *Microculex* includes 33 species [73] and currently no attempt has been made to develop a taxonomic key for all species [90]. Specimens of this subgenus were collected in Jardín between 2,261-2,403 m (17 larvae, 11 reared to obtain five females and six males) and in Yarumal (three larvae) at 2,800 m.

32. *Culex (Microculex) aureus*: In the present study, we collected 21 specimens (21 larvae, nine reared to obtain seven females and two males) identified as *Cx. aureus* in bromeliads at 2,095 m in Salento. The morphological characters are available for the identification of the pupa, larva and adult male [91]. This is the first record of this species in Colombia.

33. *Culex (Microculex) imitator*: The larval habitat for this species is exclusively bromeliads [92, 93]. We identified 20 specimens as probable *Cx. imitator* using molecular taxonomy [94]. In the Páramo de Santa Inés, Belmira, one female reared from a larva collected in a bromeliad at 2,862 m [50, 94]. The other 19 were larvae collected in bromeliads at 2,481 m in Jardín. These are potential records as the species was not definitively identified.

34. *Culex* Subgenus *Neoculex*: Species of this subgenus are not known to be of medical or economic importance [73]. In Suba in the Cundinamarca Department, specimens collected at 2,580 m were identified belonging this subgenus as species 60 [33]. In the present study, one specimen of this subgenus was collected in a pond in the Parque Ecoturístico Arvi, Santa Elena, Medellín in the Antioquia Department at 2,600 m. Considering that only one specimen was collected, future collections should be conducted to expand and identify the species.

3.5 *Orthopodomyia* sp.: Most species of this genus occur in the Neotropical and Oriental regions. Recently, in Colombia, *Orthopodomyia albicosta* (Lutz, 1905)

was collected in areas with coffee plantations in Hispania in the Antioquia Department at 1,045 m and Anserma in the Caldas Department at 875 m [95]. In the present study, for the first time in Salento, three larvae reared to males were identified as *Orthopodomyia* species and were collected in bromeliads at 2,642 m. Little is known about the biology of *Orthopodomyia* and none of the species are of medical or economic importance to humans [73].

36. *Johnbelkinia* sp.: Species of *Johnbelkinia* are considered to be potential vectors of pathogens of human diseases. [73]. Recently, specimens of *Johnbelkinia ulopus* (Dyar & Knab, 1906) were collected in the north of the Colombian Andes between 848-1,751 m [11]. In addition, two females were identified as *Johnbelkinia* sp. and were collected by oral aspirator at 2,100 m in Jardín. Both specimens were identified using the following combination of characters: “dorsal head scales with brilliant silver and blue reflections, proboscis distinctly longer than forefemur, scutal scales moderately broad and flat and dull to moderately iridescent, postpronotum with one or two posterior setae, lower mesokatepisternal setae usually not extended above lower edge of mesepimeron and mid- and hindtarsi with pale markings” [28].

37. *Sabethes* sp.: Species of this genus are forest mosquitoes and some species are of medical importance to humans [73]. Recently, *Sabethes (Sabethinus) intermedius* (Lutz, 1904) and *Sabethes (Peytonulus) soperi* Lane & Cerqueira, 1942 were collected in the province of Napo, Ecuador in the Natural Reserve of Colonso-Chalupas at 1,200 m altitude [96]. In the current study, four females of a *Sabethes* species were collected by oral aspirator at 2,100 in Jardín. The morphological characters of all specimens were: “pronotal lobe quite developed and united above; postnotum developed and nearly perpendicular to the axis of scutellum; prealar setae absent; mosquitoes with shining metallic scales” [17].

38. *Sabethes* subgenus *Sabethoides*: In Jardín, four females of this subgenus were reared from larvae obtained from bromeliads at 2,403 m [11].

39. *Shannoniana fluviatilis*: In Jardín, one female was collected by insect net at 2,253 m [11].

40. *Trichoprosopon* sp.: Species of this genus are basically forest mosquitoes, the larvae of which are found in a variety of habitats: bamboo, cacao pods, coconut shells, flower bracts of heliconians, leaf axils of plants, tree holes and artificial containers. *Trichoprosopon digitatum* (Rondani, 1848) is regarded as a potential vector of arboviruses to humans [73]. In the present study, 72 specimens were identified as species of *Trichoprosopon*, two females were collected by oral aspirator at 2,253 m in Jardín, 52 larvae in bamboo stumps (45 reared to obtain 31 females and 14 males) between 2,002-2,200 m, three larvae in one bromeliad (reared to obtain two females and one male) at 2,009 m, 14L in the leaf base of a *Xanthosoma* between 2,002-2,009 m in Jericó and one larvae in a bamboo stump at 2,471 m in Salento. Molecular identification tools will be used to aid the identification of these specimens because it was not possible to identify them to species based on morphology.

41. *Trichoprosopon evansae*: Type-loc: Restrepo, Meta, Colombia (IOC). In Jardín, one male and six females were obtained by different collecting methods in a forest habitat between 2,253-2,413 m ^[11]. In addition, one female was identified as *Tr. evansae*, which was collected at 2,253 m by entomological net in Jardín. Future collections should be conducted to expand this record.

42. *Trichoprosopon pallidiventer* s.l.: In 13 localities from northern Andean coffee-growing regions of Colombia, 346 males and 416 females were obtained by different collecting methods between 798-2,515 m ^[11]. In the current study, 11 larvae of this species were collected in a bamboo stump (five reared to obtain two females and three males) and bromeliads (six reared to obtain three females and three males) at 2,642 m in Salento. The specimens were identified using available morphological keys ^[17, 28]. These specimens belong to the Pallidiventer Complex of Zavortink (1981) which includes unknown species ^[11].

43. *Trichoprosopon* sp.: A. This morphospecies was tentatively identified among 28 females from a high valley area of Jardín municipality, part of the Colombian coffee growing region. The habitat is a cloud forest at an elevation of 2,253 m with numerous bromeliads and other epiphytes. Mosquitoes were attracted to humans during the day and they were collected using a mouth aspirator. Morphologically, these specimens seem to belong to the *Trichoprosopon pallidiventer* Complex ^[11].

44. *Trichoprosopon* sp. B: This morphospecies was found at 2,202 m in a Colombian Andean region. The mosquitoes were tentatively identified from three reared adult females obtained from larvae and pupae inhabiting a *Xanthosoma* spp. leaf base. The plants were in a small vegetation patch above the border of a road in the rural municipality of Jericó. This species is similar to species belonging to the genus *Johnbelkinia* but there are some morphological differences, especially in the pleural setae, allowing for the recognition of *Trichoprosopon* sp. B as part of the genus *Trichoprosopon* ^[11].

45. *Wyeomyia* sp: Species of this genus are not known as vectors of disease agents and have little or no economic importance to humans ^[73]. In the Páramo de Santa Inés, Belmira, eight females were collected by entomological nets at 2,977 m ^[50]. In the present study, 74 specimens were identified as *Wyeomyia* sp. In Jardín, five larvae (two reared to obtain two females) were collected in bromeliads between 2,092-2,526; two females were collected by entomological net at 2,253 m; three females by oral aspirator between 2,100-2,253 m; and one female by Shannon trap at 2,403 m. In Jericó, three larvae (reared to obtain three females) were collected from one bromeliad at 2,200 m and 51 larvae (reared to obtain 43 females and 8 males) in bromeliads at 2,018 m. For Salento, six larvae (reared to obtain four females and two males) were collected in bromeliads at 2,642 m. Finally, three larvae (one reared to obtain one female) were collected in Yarumal at 2,800 m. Molecular identification will be used in this case because most of the material consists of larvae and adult females, and this group is taxonomically difficult with morphologically similar species that require revision ^[73].

46. *Wyeomyia* aff. *phroso*: Recently, this species was collected in the adult stage in the forest habitat of three municipalities of the northern Andean coffee-growing regions of Colombia, in Jardín (Antioquia Department) and Anserma and Chinchiná (Caldas Department) between 1,313-2,253 m ^[11].

47. *Wyeomyia* (*Nunezia*): The subgenus *Nunezia* has not been well characterized, but the species possess significant differences from specimens ^[31]. Recently, *Wy. bicornis* was recorded at 2,550 m in Venezuela ^[41]. An undescribed species, consisting of 101 adult females and eight males, is known to occur in Jardín at 1,313 and in Chinchiná at 2,247 m ^[11]. Seven larvae of this subgenus were collected in bromeliads between 2,317-2,526 m in Jardín. Some characters of the of the females reared from the larvae were curved proboscis similar in length to the forefemur, antennal flagellum about 0.6 length of the proboscis, frequent presence of silvery scales on the scutellum, and other characters provided in Porter 2014 ^[31].

48. *Wyeomyia* (*Nunezia*) sp.: A. Records correspond to three males reared from larvae collected in bromeliads in forest habitat in Chinchiná (1,514 m) and Jardín (2,247 m), mentioned above ^[11].

49. *Wyeomyia* (*Nunezia*) sp.: B. In Jardín, this species was provisionally identified from two males reared from larvae collected in a bromeliad forest at 2,316 m ^[11].

50. *Uranotaenia* Subgenus *Uranotaenia*: In Bogotá, specimens collected at 2,550 m were identified belonging to this subgenus as species 4 ^[33].

51. *Uranotaenia* (*Uranotaenia*) *riverai*. Type-loc: Bogotá, Colombia (USNM). This species was described from two males and two females collected in Bogotá at 2,700 m. The larva and pupa are unknown ^[97].

Table 1: Mosquito species recorded above 2,000 m in Colombia.

# Taxon	Reference	Municipality/Neighborhood	Department	Coordinates	Collecting method	Altitude
19	^[33]	Chipaque	Cundinamarca	04°26'33"N, 74°02'39"W	Seepage/tree holes/ground pool	2,000
40	Present study	Jericó/ Quebrada cerca al casco urbano	Antioquia	05°47'18"N, 75°47'26"W	Bamboo stump/ Leaf axils of <i>Xanthosoma</i>	2,002
1, 4	^[40]	Villa Maria	Caldas	05°02'44"N, 75°30'55"W	Not reported	2,005
21 / 40	Present study	Jericó/ Quebrada cerca al casco urbano	Antioquia	05°47'17"N, 75°47'27"W	Leaf axils of <i>Xanthosoma</i> / Bromeliad	2,009
45	Present study	Jericó/Reserva la Cascada	Antioquia	05°47'50"N, 75°46'49"W	Bromeliad	2,018
30	Present study	Salento	Quindio	04°38'14"N, 75°34'15"W	Tire	2,019
6, 21, 22, 26, 30, 45 / 28 // 30	Present study	Jardín/Forest	Antioquia	05°37'18"N, 75°49'35"W	Bromeliad /Animal drinking bowl // Tire	2,092
24 / 32	Present study	Salento	Quindio	04°37'50"N, 75°28'08"W	Tire / Bromeliad	2,095
2, 6, 7, 12, 36, 37, 45 / 12 //23	Present study	Jardín/Reserva forestal La Playa	Antioquia	05°35'03"N, 75°46'02"W	Oral aspirator / rock pool // Leaf axils of <i>Xanthosoma</i>	2,100
4	^[40]	Santuario	Antioquia	06°08'15"N, 75°15'50"W	Not reported	2,150
1, 4	^[40]	Manizales	Caldas	05°03'58"N, 72°29'05"W	Not reported	2,153
6, 25	Present study	Támesis	Antioquia	05°43'19"N, 75°44'45"W	Bromeliad	2,173
40, 45	Present study	Jericó/Río Frío	Antioquia	05°43'19"N, 75°44'45"W	Bromeliad / Bamboo stump	2,200
44	^[11]	Jericó	Antioquia	05°43'19"N, 75°44'45"W	Leaf axils of <i>Xanthosoma</i>	2,202
20	^[77]	Málaga	Santander	06°41'58"N, 72°43'58"W	Not reported	2,200
4	^[40]	Aguadas	Caldas	05°36'33"N, 75°27'23"W	Not reported	2,214
30	Present study	Jardín/Reserva forestal La Playa	Antioquia	05°35'51"N, 75°47'40"W	Bromeliad	2,230
6	Present study	Jardín/Reserva forestal La Playa	Antioquia	05°36'49"N, 75°48'57"W	Entomological nets, bromeliads	2,251
47, 48	^[11]	Jardín/Reserva forestal La Playa	Antioquia	05°36'49"N, 75°48'57"W	Bromeliad	2,247
20	^[10]	Bello/Vereda Tierradentro, zona rural	Antioquia	06°19'55"N, 75°33'29"W	Ovitrap	2,252
39, 41, 43, 46/ 40, 41, 45	^[11] / Present study	Jardín/Forest	Antioquia	05°35'54"N, 75°49'11"W	Insect net	2,253
31	Present study	Jardín/Reserva forestal La Playa	Antioquia	05°35'52"N, 75°47'40"W	Bromeliad	2,261
26, 30	^[33]	Santa Leticia	Huila	02°14'25"N, 76°10'12"W	Swamp	2,300
20	^[10]	Bello/Vereda Tierradentro, zona rural	Antioquia	06°19'55"N, 75°33'29"W	Ovitrap	2,302
49	^[11]	Jardín/Forest	Antioquia	05°35'52"N, 75°47'39"W	Bromeliad	2,316
30, 47	Present study	Jardín/Reserva forestal La Playa	Antioquia	05°35'13"N, 75°46'18"W	Bromeliad	2,317
6	Present study	Jardín/Reserva forestal La Playa	Antioquia	05°35'03"N, 75°46'02"W	Shannon trap	2,376
15	^[21, 33]	Soacha/Ogamora town	Cundinamarca	04°34'41"N, 74°12'52"W	Volcanic rockhole in pasture	2,400
2	^[49]	Jardín/Reserva forestal La Playa	Antioquia	05°35'03"N, 75°46'02"W	Shannon trap	2,400
2, 6, 31, 45 / 38	Present study / ^[11]	Jardín/Reserva forestal La Playa	Antioquia	05°37'59"N, 75°49'48"W	Shannon trap, bromeliad	2,403
6	Present study	Jardín/Reserva forestal La Playa	Antioquia	05°37'59"N, 75°49'48"W	Bromeliad	2,412
41	^[11]	Jardín	Antioquia	05°36'49"N, 75°48'57"W	Bromeliad, <i>Xanthosoma</i> spp. leaf base,	2,413
26	Present study	Salento	Quindio	04°38'06"N, 75°28'58"W	Bromeliad	2,416
40	Present study	Salento	Quindio	04°37'50"N, 75°28'08"W	Bamboo stump	2,471
21, 33	Present study	Jardín/Reserva forestal La Playa	Antioquia	05°35'03"N, 75°46'02"W	Bromeliad	2,481
42	^[11]	Jardín/Reserva forestal La Playa	Antioquia	04°37'50"N, 75°28'08"W	Insect net, bromeliad, <i>Xanthosoma</i> spp. leaf base	2,515
26	Present study	Salento	Quindio	04°37'50"N, 75°28'08"W	Bamboo stump/ <i>Xanthosoma</i> spp. leaf base	2,515
2, 6	Present study	Jardín/Reserva forestal La Playa	Antioquia	05°35'03"N, 75°46'02"W	Bromeliad	2,521
24 / 45, 47	Present study	Jardín/Reserva forestal La Playa	Antioquia	05°35'13"N, 75°46'18"W	Palm-leaf / Bromeliad	2,526
30	^[85]	Bogotá/ Río Tunjuelito	Cundinamarca	04°32'43"N, 74°07'32"W	Shannon trap	2,540

30	[86]	Mosquera/Centro Agropecuario Marengo	Cundinamarca	04°42'00"N, 74°14'00"W	Aspirator, artificial containers, Shannon trap	2,543
50	[33]	Bogotá/Canal del Rio Negro	Cundinamarca	04°35'56"N, 74°4'51"W	Swamp/seepage	2,550
30	[33]	Bogotá/Ciudad Universitaria	Cundinamarca	04°38'08"N, 74°04'58"W	Ground pool	2,555
26	[33]	Soacha/Chusacá town	Cundinamarca	04°33'00"N, 75°15'00"W	Swamp	2,570
27	Present study	Salento	Quindio	04°37'50"N, 75°28'08"W	Cattle footprint	2,576
10, 16, 34	[20, 21, 33]	Bogotá/Suba	Cundinamarca	04°44'28"N, 74°05'02"W	Bromeliad	2,580
30	[87]	Bogotá/Urban area	Cundinamarca	04°36'43"N, 74°04'07"W	Not reported	2,600
26	[33]	Tangua/Vereda El Cebadal	Nariño	01°05'44"N, 77°23'38"W	Ground pool	2,600
17, 33	[33]	Sopó	Cundinamarca	04°54'29"N, 73°56'25"W	Pond	2,600
2	[48]	Angelopolis/Reserva Ecológica	Antioquia	06°06'47"N, 75°43'01"W	Bromeliad	2,600
34	Present study	Medellín/ Parque Ecoturístico Arví	Antioquia	05°15'56"N, 75°29'49"W	Pond	2,600
15	[21, 33]	Usaquen/Las Acacias	Cundinamarca	04°43'11"N, 74°02'12"W	Small ground pool in garden lawn	2,640
21, 35, 42, 45/ 26 // 42	Present study	Salento	Quindio	04°37'50"N, 75°28'08"W	Bromeliad/ Cattle footprint // Bamboo stump	2,642
10-12	[20, 33]	Bogotá	Cundinamarca	04°44'28"N, 74°05'02"W	Bromeliad	2,650
15	[21]	Bogotá/Usaquen, Las Acacias	Cundinamarca	04°41'20"N, 74°01'31"W	Ground pool	2,690
3, 13, 17, 18 / 51	[51]/[97]	Bogotá	Cundinamarca	04°35'56"N, 74°04'51"W	Ground pool / Magoon trap	2,700
6	Present study	Angelopolis/El Romeral	Antioquia	06°05'30"N, 75°44'05"W	Bromeliad	2,704
6-8, 12	[50]	Páramo de Santa Inés, Belmira	Antioquia	06°37'22"N, 75°39'19"W	Bromeliad	2,734
2, 6	Present study	Yarumal/La Maconia	Antioquia	06°57'48"N, 75°25'02"W	Bromeliads	2,755
12	Present study	Yarumal/La Maconia	Antioquia	06°57'48"N, 75°25'02"W	Artificial container	2,799
4	[33]	Pasto/Laguna de la Cocha	Nariño	01°16'00"N, 77°09'07"W	Swamp	2,800
31, 45	Present study	Yarumal/La Maconia	Antioquia	06°57'48"N, 75°25'02"W	Bromeliad	2,800
6, 7	Present study	Angelopolis/ El Romeral	Antioquia	06°07'55"N, 75°45'45"W	Bromeliad	2,802
3, 17, 18	[51]	Bogotá/Rio San Francisco	Cundinamarca	04°35'01"N, 74°02'25"W	Ground pool / Magoon trap	2,840
3, 8, 14, 21 / 33	[50]	Páramo de Santa Inés, Belmira	Antioquia	06°37'10"N, 75°39'26"W	CDC trap, oral aspirator /bromeliads	2,862
29	[84]	Bogotá/Cerro de Monserrate	Cundinamarca	04°36'21"N, 74°03'23"W	Aspirator/stream pool	2,900
6, 9	[50]	Páramo de Santa Inés, Belmira	Antioquia	06°38'44"N, 75°42'16"W	Oral aspirator, bromeliad	2,905
32	Present study	Salento	Quindio	04°37'50"N, 75°28'08"W	Bromeliad	2,905
6	[50]	Páramo de Santa Inés, Belmira	Antioquia	06°38'44"N, 75°42'16"W	Oral aspirator	2,928
2, 45	[50]	Páramo de Santa Inés, Belmira	Antioquia	06°37'30"N, 75°42'25"W	Entomological nets, bromeliads	2,977
6, 9	[50]	Páramo de Santa Inés, Belmira	Antioquia	06°38'40"N, 75°42'23"W	Oral aspirator, bromeliad	3,000
5	[60]	Chocontá	Cundinamarca	05°01'00"N, 73°42'00"W	Bromeliad	3,000
6, 7	[50]	Páramo de Santa Inés, Belmira	Antioquia	06°38'48"N, 75°42'35"W	Oral aspirator, CDC trap, bromeliad	3,098
5	[61]	Guasca	Cundinamarca	04°46'00"N, 74°10'00"W	Bromeliad	3,100
9	[20, 33]	Puracé/Parque Nacional Natural Puracé	Huila	02°12'00"N, 76°21'00"W	Bromeliad, Human Landing Catch	3,100
3	[22]	Túquerres	Nariño	01°05'14"N, 77°37'08"W	Not reported	3,104
26	[33]	Bogotá/Rio San Francisco	Cundinamarca	04°35'01"N, 74°02'25"W	Stream pool	3,150
13,17,18,21/ 9,12, 33	[51] / [20], [33]	Bogotá/Rio San Francisco	Cundinamarca	04°35'01"N, 74°02'25"W	Magoon trap/Bromeliad/Stream pool/Pond	3,186
17	[70]	Páramo de Santa Inés, Belmira	Antioquia	06°36'18"N, 75°39'57"W	CDC light trap	3,200
17	[33]	Choachí	Cundinamarca	04°32'00"N, 73°56'00"W	Ground pool	3,250

4. Discussion

This paper provides the most complete, up-to-date list of mosquito species known to occur above 2,000 m in Colombia, with a total of at least 35 species belonging to 11 genera and 15 subgenera according to the latest nomenclature and classification. The occurrence of some species mentioned only from their type locality in Colombia, for example, *An. oiktorakras*, *Ae. (How.) marinkellei*, *Ae. (Och.) bogotanus*, *Ae. (Och.) deficiens*, *Cx. chitae* and *Ur. riverai*, constitutes valuable information for overall country diversity. Twelve records, *An. (Kerteszia) sp. Boliviensis Group*, *Ae. (How.) Eleanorae Group*, *Ae. (How.) marinkellei/osornoi* hybrid, *Ae. (How.) quadrivittatus*, *Cx. (Microculex) aureus*, a species of *Culex* subgenus *Neoculex*, *Trichoprosopon sp. A*, *Trichoprosopon sp. B*, *Wy. aff. phroso*, *Wy. (Nuz) sp. A*, *Wy. (Nuz) sp. B* and a species of *Wy.* subgenus *Nunezia*, were not included in a recent publication about mosquitoes of Colombia [98]. For *Trichoprosopon* this is probably because the study with the most recent new records for the tribe Sabethini is was published the same year [11]. About the species of the *An. (Kerteszia) Boliviensis Group*, the records for Colombia are recent [48–50, 94]. The taxonomic information for *An. boliviensis* is incomplete due to the absence of descriptions of the immature stages and adult male. Moreover, using the latest taxonomic key to adult females, it is possible to identify this species only as a member of the Boliviensis Group because of the high similarity of morphological characters. Given the incomplete information on species in this group and their taxonomic status, additional investigation is highly desirable [48, 49].

Species of subgenus *Lophopodomyia* of the genus *Anopheles* have not been studied in detail. These are forest mosquitoes and little is known about their bionomics. Even though species of this subgenus are not of medical importance to humans, adult females are apparently active during twilight periods and will feed on humans and other animals that enter their realm [73]. For *An. pseudopunctipennis* (subgenus *Anopheles*) and *An. argyritarsis* (subgenus *Nyssorhynchus*) some authors describe a strong association between them because the species co-occur, although in different proportions, in the same larval habitats at < 1,000 m [99–102]. In Bolivia, both species were found above 2,000 m, which is considered to be “of mid-range altitude” compared with the other *Anopheles* species. The minimum and maximum altitudes are 206 and 2,732 m for *An. pseudopunctipennis* and 348 m and 2,323 m for *An. argyritarsis* [37].

In general, *Anopheles* species present in high mountain ecosystems of Colombia are not significant vectors, however, above 1,000 m *An. pseudopunctipennis* is the only species considered to be a malaria vector and has priority in control efforts in Bolivia [37]. Moreover, *Anopheles (Nyssorhynchus) albimanus* (Wiedemann, 1820), one of the most important vectors of human malaria in Colombia [38,39,103] is typically found in coastal land below 500 m; nevertheless, an earlier report indicated its presence at 1,941 m in Morelia, Mexico [104]. Furthermore, highland expansion of *An. albimanus* has been demonstrated in Ecuador, up to 1,541 m [105]. For countries in Mesoamerica and the Caribbean, near present and future climate data suggest that *An. albimanus* is likely to invade high-altitude areas (>2,000 m) by 2080 as a result of global warming [106].

Remarkably, species belonging to some genera of the tribe Aedini have not been well studied. Therefore, it is presumed that additional species could occur above 2,000 m in Colombia. In Ecuador, the *Ae. (How.)* species are reported mainly from the Andean highlands. Recently *Aedes (Howardina) fulvithorax* (Lutz, 1904) was found in bromeliads in a small town at 1,475 m in Napo Province [96]. Some species of this subgenus attack humans readily and are very abundant [20]. Isolation of an unidentified virus was reported for *Aedes (Howardina) sp.* [62]. Another important subgenus of *Aedes* is *Ochlerotatus*, which includes species apparently endemic to the Colombian Andes and are relatively poorly known [21]. *Aedes (Och.) euiris* is the mosquito with the maximum altitude (3,250 m) in Colombia [33] and in Venezuela (3,133 m) [41]. The best studied aedine species is *Ae. aegypti*, which occurs up to 2,302 m in Colombia [10], up to 2,130 m in Mexico [75] and up to 2,600 m in Costa Rica [76]. Moreover, some studies have suggested that the geographic range of *Ae. aegypti* could be increasing due to climate warming [107]. A more thorough investigation of distribution at high altitudes and the role of these aedine species as vectors of arboviruses should be conducted. This is especially pertinent in Colombia, due to the presence of *Ae. aegypti* at 1,960 m in Jericó and up to 2,302 m in Tierradentro, Bello and positive dengue infections in specimens collected in the Paris neighborhood of Bello at 1,984 m [10].

The genus *Culex* is the second largest genus of the family Culicidae after genus *Aedes*. It has a cosmopolitan distribution with 768 formally recognized species divided into 26 subgenera. In the Americas members of this extensive genus are subdivided into 14 subgenera [90]. The subgenera *Culex* and *Melanoconion* are the most important because of their abundance, diversity and epidemiological importance [16]. Above 2,000 m, species of three subgenera, *Culex*, *Neoculex* and *Microculex*, are present in Colombia. Altitude does not seem to limit the distribution of *Cx. quinquefasciatus*, since it has been found commonly in municipalities of the Cundinamarca Department between 2,540–2,600 m [9, 33, 85–87]. *Culex quinquefasciatus* is a competent vector of filarial infections, West Nile virus and St. Louis encephalitis virus. Although recent studies have demonstrated the circulation of West Nile virus in Colombia [108, 109], this species has not yet been implicated as a vector in this country [74].

Finally, species from the subgenus *Uranotaenia* of the genus *Uranotaenia* are not of medical or economic importance but adult females of some species are known to feed on frogs, birds and mammals [73]. For the tribe Sabethini, an important study was undertaken in 17 representative municipalities of the northern Andes in Colombia, the most important coffee-growing region in the country recorded 10 species above 2,000 m. However, in some cases it was not possible to identify some specimens to species. Many of these species exhibited close similarity to described species but possessed morphological differences that require direct comparison with type material [11]. In general, this tribe has not been well studied, and additional and/or undiscovered species may occur over 2,000 m in Colombia. The mosquito fauna and their ecology above 2,000 m in Colombia requires more research, and we consider that the findings reported here to be a good starting point.

5. Conclusion

A total of 35 species of mosquitoes are reported from high Andean mountain ecosystems in Colombia with at least five of medical importance. The presence of these species along with epidemiological data are part of the baseline data for surveillance programs. Some species of the subgenera *Howardina* are reported for the first time from this area. This is a taxonomic complex group that deserve more detailed studies. The annotated list of mosquitoes given in this study is the most complete one for mosquitoes collected above 2,000 m in the country.

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