Check List the journal of biodiversity data

NOTES ON GEOGRAPHIC DISTRIBUTION

Check List 18 (2): 275–280
https://doi.org/10.15560/18.2.275

PENSOFT

Wyeomyia abebela Dyar & Knab, 1908 and *W. coenonus* Dyar & Knab, 1913: new mosquito records (Diptera, Culicidae) in the Choco Biosphere Reserve, Ecuador, a biodiversity hotspot

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Abstract

We report the first records of two mosquito species, *Wyeomyia* (*Wyeomyia*) *abebela* Dyar & Knab, 1908 and *W.* (*Hystatomyia*) *coenonus* Dyar & Knab, 1913, from the Choco Biosphere Reserve, a global biodiversity hotspot in Ecuador. We collected these mosquitoes in the Mashpi Lodge Reserve, a conserved natural area. Specimens were collected during their immature stages in bromeliads using a 375-ml absorber dropper. Our new data for *W. abelela* increases the geographical distribution of this species to include seven countries of America (Belize, Costa Rica, Guatemala, Honduras, Mexico, Panama, and Ecuador); *W. coenonus* is now known from two countries of America (Panama and Ecuador), and our records of *W. abelela* and *W. coenonus* are the first from South America.

Keywords

Bromeliads, distribution expansions, Mashpi, natural reserve

Academic editor: Fabio Laurindo da Silva | Received 9 July 2021 | Accepted 4 March 2022 | Published 17 March 2022

Citation: Vaca-Moyano F, Duque PL, Enríquez S, Herrera V, Sánchez-Murillo M, Rivadeneira E, Molina CA (2022) *Wyeomyia abebela* Dyar & Knab, 1908 and *W. coenonus* Dyar & Knab, 1913: new mosquito records (Diptera, Culicidae) in the Choco Biosphere Reserve, Ecuador, a biodiversity hotspot. Check List 18 (2): 275–280. https://doi.org/10.15560/18.2.275

Introduction

The Andes Mountain range in tropical Ecuador favors diverse landscapes, such as Amazonia and the Choco rainforest (Morrone 1999). The Ecuadorian Andean Choco is a global biodiversity hotspot, with some of the highest rates of endemism in the world and a high diversity of species per area (Myers 1988; Myers et al. 2000). The mosquito fauna is diverse and abundant in tropical and subtropical regions (Linton et al. 2013; Navarro

et al. 2015), but because many areas in the Ecuadorian Choco are unexplored, the local diversity and distribution of mosquitoes are underestimated. Some natural environments have undergone dramatic modifications and declines that affect the behavior of mosquito populations (Weaver 2013).

The genus *Wyeomyia* F.V. Theobald, 1901 includes sylvatic mosquitoes from tropical and subtropical regions of the Americas, with 139 species divided among 17 subgenera and at least 28 species without a subgeneric classification (Belkin et al. 1970; WRBU 2021a). The immature stages of *Wyeomyia* prefer natural breeding sites in tropical rainforests, including bamboos, bromeliads, and *Heliconia* L. leaf axils, tree holes, and flower bracts, but they also use artificial containers in rural and urban areas (WRBU 2021a). Several *Wyeomyia* species are potential vectors of arboviruses. However, these pathogens do not have drastic consequences for human populations (Hall and Fish 1974; Pajot 1980; Chowdhary et al. 2012).

Wyeomyia is considered one of the most challenging mosquito groups to identify, with identification based on the scale ornamentation in adult stages (Judd 1996). However, it is feasible to identify some species by the morphological characters of the male genitalia (Belkin et al. 1970; Petersen et al. 2017). Here, we identify two species, *Wyeomyia* (*Wyeomyia*) *abebela* Dyar & Knab, 1908 and *W.* (*Hystatomyia*) *coenonus* Dyar & Knab, 1913, by studying the male genitalia. Our study is part of a comprehensive analysis of arthropod vectors and environmental microbiomes in the tropical rainforests of Choco and Amazonia with a public health approach.

Methods

We carried out our research in primary and secondary preserved forest in the western foothills of the Andes; our collections of mosquitoes were made in the natural and touristic areas of the Mashpi Lodge Reserve, which is over 2,500 ha and located near the Metropolitan District of Quito (Benavides et al. 2010). The Mashpi Lodge Reserve is within the Piedmont evergreen forest in the biodiverse Choco-Darien hotspot in northwestern Ecuador (Sierra 1999; Benavides et al. 2010) (Fig. 1). Historically, Mashpi Lodge Reserve has been affected by wood extraction since the 1970s. The biological diversity of several groups of organisms and the high level of endemism are due to the altitude gradient, the transition between Andean and tropical biotas, and high rainfall rates (Ron 2000).

Our study was performed during three field collections in 2020 and 2021. Our methods for sampling of immatures stages of mosquitoes were based on standard protocols (Belkin 1965; Gaffigan and Pecor 1997;

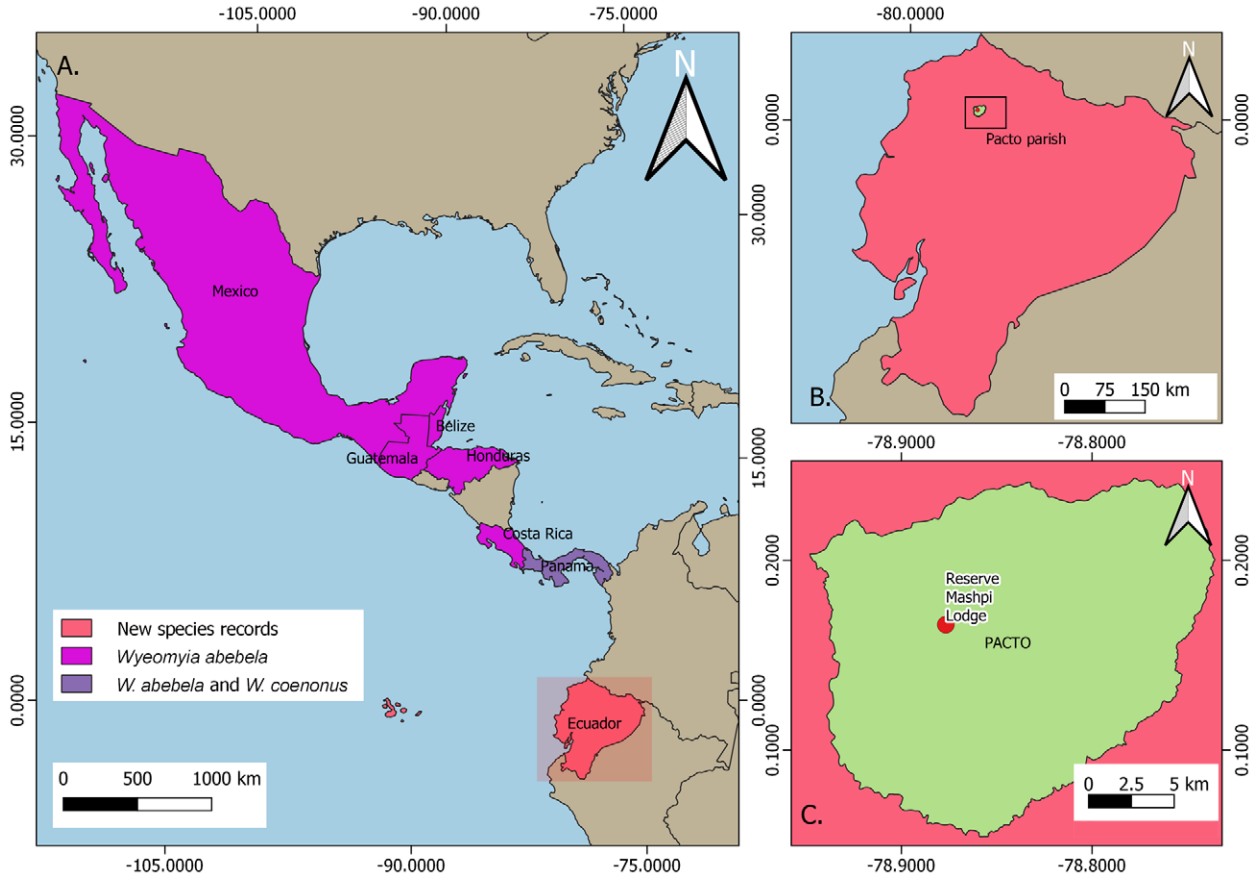

Figure 1. A. Geographical distribution of *Wyeomyia abelela* and *W. coenonus* in South America. **B.** Northwestern Pichincha province, within Andean Choco, Biosphere Reserve of Ecuador. **C.** Pacto County, showing the location of the Mashpi Lodge Reserve, where the new records of *W. abelela* and *W. coenonus* were collected.

Navarro et al. 2015; Duque et al. 2019). We used a 375-ml absorber dropper to collect immature mosquitoes from natural breeding sites (bromeliads) at heights of 1.8–2.5 m. We also sampled from bromeliads at 30 m high above the ground by using a zip line installed over the forest canopy in the reserve. The biological samples were stored in 100-ml Whirl-Pak bags and taken to the Applied Entomology Unit laboratory, located at the Universidad Central del Ecuador, Quito, Ecuador. The mosquitoes were reared under laboratory conditions to adults; then the adult samples were preserved in entomology boxes (Belkin 1965). Identifications were made using the keys and descriptions by Dyar et al. (1917), Lane (1953), and Harbach and Knight (1980); the genitalia protocols followed those established by Belkin (1965) and Gaffigan and Pecor (1997). Finally, the individuals were registered in the National Reference Collection of Arthropods of Zoonotic Importance of the Zoonosis Investigation Center, Quito, Ecuador.

Results

***Wyeomyia* (*Wyeomyia*) *abebela* Dyar & Knab, 1908**
Figure 2A–D

New records. ECUADOR – **Pichincha** • Metropolitan District of Quito, Mashpi Lodge Reserve; 00°09′58″N, 078°52′36″W; 945 m alt.; 01.III.2021; Franklin Vaca-Moyano, Paul L. Duque, Sandra Enríquez, Erika Rivadeneira, Marco Sánchez, Vanessa Herrera, C. Alfonso Molina leg.; bromeliads/absorber dropper; 1 ♂, BPML-SC-BR9-100.

Identification. Male genitalia: in the subgenus *Wyeomyia* s.s., gonocoxite (Gc) with setae and scales, and gonostylus (Gs) end with complex and variable structures. *Wyeomyia abebela* is characterized by having the Gs with a conspicuous appendix, the apical lobes much developed, and the most important characteristic is the ninth tergum (IX-Te), forming two rows of setae on each lobe. These characteristics are used to separate *W. abebela* from its most similar congeners, like *W. arthrostigma* Lutz, 1095; *W. medioalbipes* Lutz, 1904; *W. gaudians* Dyar & Nuñez Tobar, 1927, and *W. vanduzeei* Dyar & Knab, 1906 (Lane 1953). Overall, the male genitalia of *W. abebela* displays unique and very well-marked anatomical characteristics. The Gc is elongate and curved, with three long, thin setae inserted horizontally on the basal third. The Gs is larger than the Gc, and the Gs has a downward displaced lobe (Fig. 2A); it is curved and, at its base, it has two spines and numerous hairs on the upper external margin (Fig. 2B). The ninth tergum (IX-Te) has the convex middle part with two lobes; each ninth tergum lobe (IX-TL) has thick setae in two rows (Fig. 2C). Tergum X (X-Te) is apically sclerotized and has four terminal teeth (Fig. 2D).

Females are characterized by the dark mid-lobe of the scutellum (Stm); dark scales on the abdominal tergites and yellowish scales on the sternites, both separated by

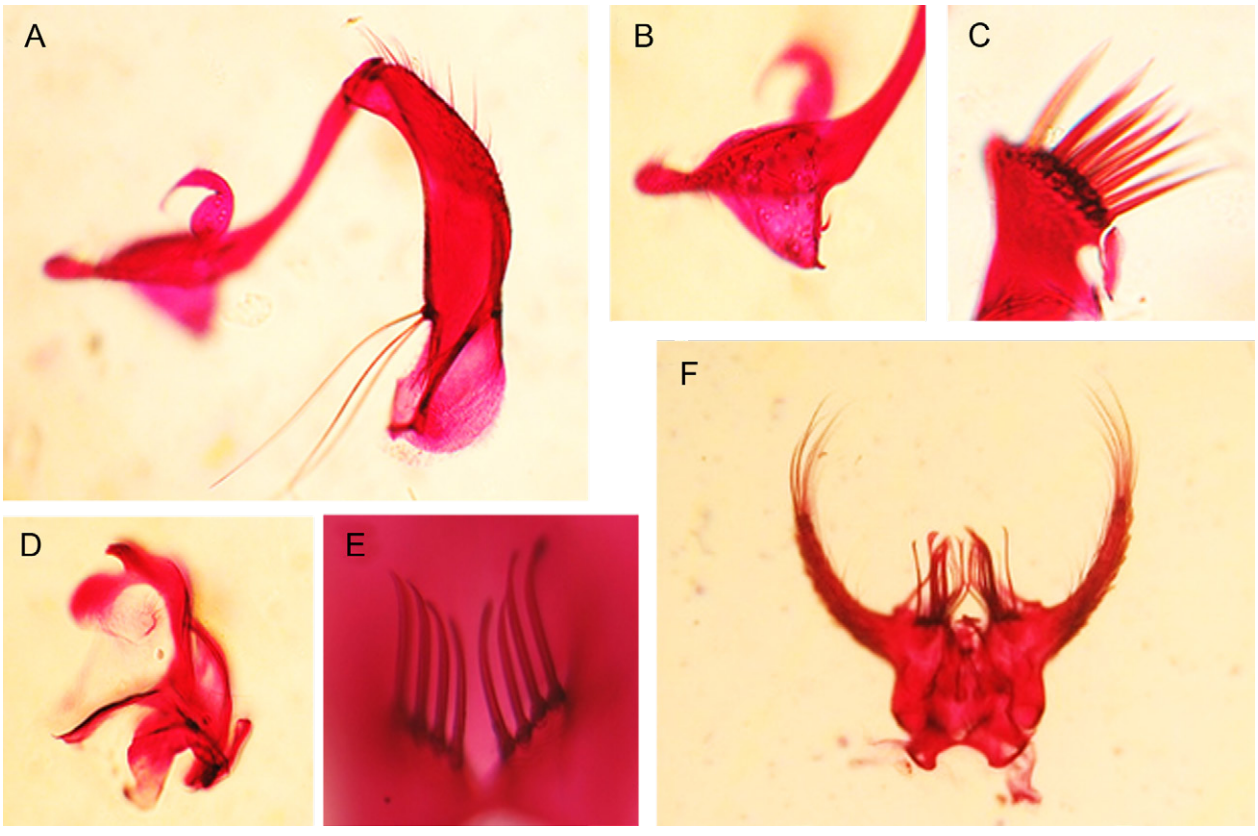

Figure 2. Male gentialia. **A–D.** *Wyeomyia abebela*: (**A**) ventral view of gonocoxite (Gc) and gonostylus (Gs); (**B**) curved lobe in the external margin of gonostylus (Gs); (**C**) tergum IX lobe (IX-TL); (**D**) tergum X (X-Te). **E, F.** *Wyeomyia coenonus*: (**E**) tergum IX lobe (IX-TL); (**F**) ventral view of Gc and Gs.

a straight line; legs with dark scales; the dark pronotum (PrN) with white scales on top and violaceus scales on the middle; and the scutum (Scu) with dark brown integument. These characters differentiate *W. abebela* from *W. arthrostigma* and *W. pertinans* Williston, 1896; but not from *W. melanopus* Dyar, 1919. Males of *W. abebela*, which are similar to the females, have slightly more plumose antennas (Ant), and shorter maxillary palpus (MPlP). However, the male is almost impossible to separate from *W. melanopus* and *W. pertinans* (Lane 1953).

***Wyeomyia* (*Hystatomyia*) *coenonus* Dyar & Knab, 1913**

Figures 2E, F, 3A–C

New records. ECUADOR – **Pichincha** • Metropolitan District of Quito, Mashpi Lodge Reserve; 00°09′58″N, 078°52′36″W; 945 m alt.; 19.II.2020, Franklin Vaca, Marco Sánchez, C. Alfonso Molina leg.; bromeliads/absorber dropper; 1 ♂, BPML-RL-BR01-100 • same locality; 00°10′01″N, 078°52′44″W; 852 m alt.; 13.XI.2020; Paul L. Duque, Sandra Enríquez, Vanessa Herrera, Erika Rivadeneira leg; bromeliads/canopy/absorber dropper; 1 ♂, SA-B1-02-103.

Identification. Male genitalia: the subgenus *Hystatomyia* is easily differentiated based on the subapical position of the Gs. *Wyeomyia coenonus* can be separated from other similar *Hystatomyia* species by the thin shape of Gs and the conic apex, which is oriented towards the midline. Additionally, IX-Te has four short, slender setae that are curved outwards. *Wyeomyia coenonus* can be confused with other species like *W. lamellata* Bonne-Wepster & Bonne, 1919, which has four or five setae on IX-Te (Bruijning 1959), although Lane (1953) only mentioned five setae on IX-Te of the latter. *Wyeomyia coenonus* can be separated from *W. lamellata* by the wide, oval Gc and the three leaf like setae and two slenderer ones on the apex of the Gs in the latter species. Another similar species is *W. esmeraldasi* Levi-Castillo, 1955, which has its type locality in Ecuador, but whose holotype is apparently lost. According to the original description of *W. esmeraldasi* (Levi-Castillo 1955), the main characteristic separating it from *W. coenonus* is that each lobe of IX-Te has three separate, saber-like, curved, pointed setae at the apex in the former species. Also, *W. coenonus* can be differentiated from *W. negrensis* Gordon & Evans, 1922 by the presence of three setae on each lobe of IX-Te in the latter and by the overall shape of the Gc and Gs (Lane 1953). According to Lane (1953), *W. coenonus* is distinguished by the following characteristics: Gc thick at its base and narrow close to the apex, and setae on the sides and apex of the Gc (Fig. 2F); X-Te ending in a single tooth; IX-Te concave and with a wide interlobular space; and each ninth tergum lobe (IX-TL) with four short, slender setae curved outwards (Fig. 2E).

Females are characterized by having the basal section of the 4th vein of the wing with anterior and lateral ligulate scales; Scu. with dark scales; tarsi dark; and abdomen separated with a straight line (Fig. 3A–C). *Wyeomyia coenonus* can be confused with *W.* (*Exallomyia*) *tarsata* Lane & Cerqueira, 1942 because both species have a dark PrN whose base has white scales, or with *W. negrensis*, which has violaceus scales on the PrN. There are few characters described for adult males to distinguish *W. coenonus* from other similar species; adults have the femora whitish ventrally and tarsi from segments I to V in a continuous white line.

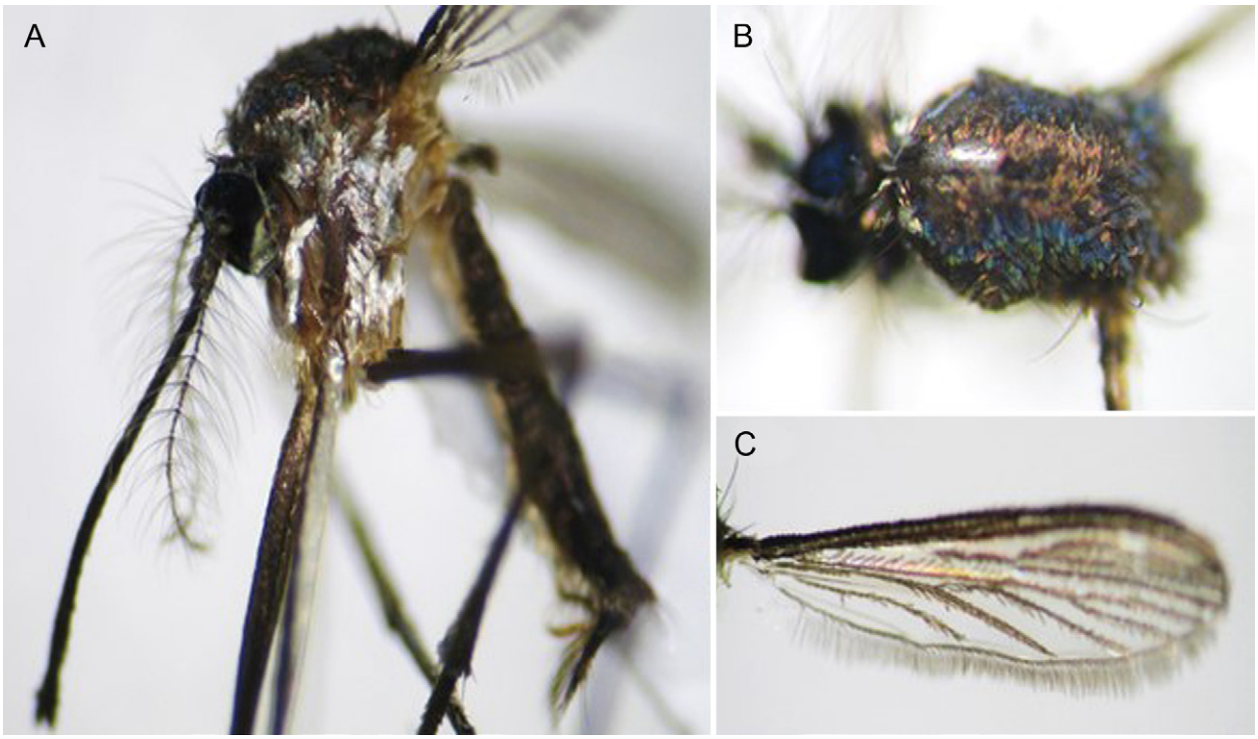

Figure 3. Adult male of *Wyeomyia coenonus*. **A.** Lateral view. **B.** Scutellum showing scales. **C.** Wing with veins and scales.

Discussion

Wyeomyia abebela was originally described from Cordoba, Veracruz, Mexico, and this species is also known from Belize, Costa Rica, Guatemala, Honduras, and Panama (Lane 1953; Heinemann and Belkin 1977; Knight and Stone 1977; Pecor et al. 2002; WRBU 2021a). Our new record is the first from South America and adds Ecuador as the seventh country in the Americas where this species has been found. *Wyeomyia abebela* is the third species of the subgenus *Wyeomyia* s.s. in Ecuador; the other species are *W. arthrostigma* and *W. celaenocephala* Dyar & Knab, 1906.

Wyeomyia coenonus was originally described from Tabernilla, Canal Zone, Panama (WRBU 2021b). Our new Ecuadorian record is the first from South America. *Wyeomyia coenonus* is the third species of the subgenus *Hystatomyia* in Ecuador; *W. autocratica* Dyar & Knab, 1906 and *W. esmeraldasi* are the other two species.

Immature stages of *W. abebela* prefer subtropical and tropical bromeliads (Lane 1953; Heinemann and Belkin 1977; Knight and Stone 1977; Pecor et al. 2002), but knowledge is more limited for *W. coenonus*. However, other members of the subgenus *Hystatomyia* are dominant in natural breeding sites such as bromeliads in forest habitats (Wolff 2004). Overall, little is known about the biology, relationship with other species, population dynamics, and these two species' natural breeding sites. There is a lack of evidence for the natural transmission of pathogens in sylvan areas. We found *W. abebela* and *W. coenonus* in a preserved cloud rainforest. We collected immature stages of these species on bromeliads at 1.8–2.5 m above the ground. In addition, *W. coenonus* was collected 30 m above the ground, this is the first time that immature mosquitoes were collected in natural breeding sites at this higher (30 m) stratification level in Ecuador.

We confirm that the immature stages of *W. abebela* and *W. coenonus* prefer species of Bromeliaceae. We also note the pivotal necessity of a continued and permanent effort aiming to increase knowledge on the behavior of the genus *Wyeomyia* in Ecuador, although in recent years studies on mosquitoes and specifically this genus have increased (Navarro et al. 2015; Navarro et al. 2016; Navarro et al. 2018; Duque et al. 2019). Overall, the immature stages and natural classification of *Wyeomyia* are still poorly studied due to the diversity of its behavior and the absence of discriminating characters in adult stages (Harbach and Peyton 1990; Judd, 1996, 1998; Motta et al. 2007).

In summary, we extend the geographical distribution of *W. abebela* and *W. coenonus* to Ecuador and report these species for the first time from South America. As a result, alpha diversity of mosquitoes in Ecuador has increased to around 253 species (Navarro et al. 2016, 2018).

Acknowledgements

Our work was supported by the Universidad Central del Ecuador, Project DI- CONV-2019-073. We extend our gratitude to Mashpi Lodge Reserve and Mateo Roldán, head of the Research and Biology Department of the Mashpi Lodge Reserve, Mashpi, Ecuador. Our research was performed under the Ministerio del Ambiente del Ecuador collection permit (MAE-DNB-CM-2015-0028). We acknowledge Juan Guevara and Roberto Alajo for their field assistance. We are thankful the support and comments of the Check List reviewers, the editor-in-chief, and the academic and copy editors.

Authors' Contributions

Conceptualization: FVM. Data curation: FVM, MSM, PLD. Formal analysis: VH, PLD, FVM, SE, MSM, ER, CAM. Funding acquisition: SE, CAM. Investigation: PLD, FVM. Methodology: SE, MSM, FVM, ER, PLD, VH, CAM. Project administration: CAM. Resources: SE. Writing – original draft: FVM, CAM, PLD, SE, VH, MSM, ER. Writing – review and editing: FVM, MSM, SE, ER, CAM, PLD, VH.

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