



## Biting midges (Diptera: Ceratopogonidae) from an urban forest fragment in Central Amazon (Brazil): Effects of opening areas on abundance, richness, and composition

EDUARDA D.R. DOURADO<sup>1</sup>, RUTH L. FERREIRA-KEPPLER<sup>1</sup>,  
RENATO T. MARTINS<sup>1,2</sup> and MARIA M. RONDEROS<sup>3,4</sup>

<sup>1</sup> Instituto Nacional de Pesquisas da Amazônia/ INPA, Coordenação de Biodiversidade/  
COBIO, Av. André Araújo, 2936, Campus II, 69067-375 Manaus, AM, Brazil

<sup>2</sup> Programa de Pós-Graduação em Ecologia e Evolução, Universidade Federal de Goiás/  
UFG, Av. Esperança, s/n, Campus Samambaia, 74690-900 Goiânia, GO, Brazil

<sup>3</sup> Centro de Estudios Parasitológicos y de Vectores/CEPAVE, Facultad de Ciencias Naturales y Museo (UNLP), Consejo Nacional  
de Investigaciones Científicas y Técnicas/ CONICET, Boulevard 120, s/n, e/61 y 62 La Plata, Buenos Aires, Argentina

<sup>4</sup> Facultad de Ciencias Naturales y Museo (UNLP), División Entomología, Museo  
de La Plata, Paseo del Bosque, s/n, 1900 La Plata, Argentina

*Manuscript received on May 17, 2017; accepted for publication on August 29, 2017*

### ABSTRACT

We assessed the immature stages of Ceratopogonidae (Diptera) in artificial containers in an urban forest fragment in Manaus (Brazil), including their behavioral, biological and ecological information. In addition, we evaluated the effects of deforestation in an open and forested area on Ceratopogonidae communities. Immatures were sampled between August 2012 and July 2014 in artificial containers installed in both habitat types. We collected 685 immatures of seven morpho-species of *Bezzia* Kieffer, *Culicoides* Latreille, *Dasyhelea* Kieffer, *Forcipomyia* Meigen, and *Palpomyia* Meigen. In the open area, we recorded higher temperature and electrical conductivity values than in the forested area; however, these variables did not differ between seasons. Water volume was higher in open area and in rainy season, while pH was similar in both areas and seasons. Species richness was higher in forested area, but did not differ between seasons. We did not record differences in abundance between areas or seasons. Community composition differed between areas, but not between seasons. We provide the first records of *Culicoides* (*Hoffmania*) *insignis* Lutz and *C. (Haematomyidium) quasiparaensis* Clastrier in artificial containers from the state of Amazonas. Our results suggest that the preservation of forested areas in Amazonas is fundamental for the maintenance of the life cycle of some species of Ceratopogonidae.

**Key words:** Amazonia, aquatic insects, artificial containers, deforestation.

### INTRODUCTION

Biting midges (Culicomorpha; Ceratopogonidae) are one of the most common and diverse families of Diptera. They are popularly known as “polvorines”,

“manta blanca”, “jejenes”, or “chaquistes” in Spanish-speaking countries, as biting midges, no-see-ums, or punkies in English-speaking countries, and as “maruim”, “mosquito-pólvora”, or “meruim” in Brazil (Spinelli and Wirth 1993). Currently, there are four extant subfamilies, 111 genera, and 6,267 species worldwide (Borkent 2016). The

---

Correspondence to: Renato Tavares Martins  
E-mail: [martinsrt@gmail.com](mailto:martinsrt@gmail.com)

subfamilies Leptoconopinae, Forcipomyiinae, Dasyheleinae and Ceratopogoninae include in 52 genera and nearly 1,225 species are known in or have been described from the Neotropical region. In Brazil, 483 species and 31 genera, and in the state of Amazon, 264 species and 28 genera are currently recognized (Santarém and Felipe-Bauer 2016).

These small flies can be vectors of several diseases such as mansonellosis and oncocercosis caused by nematodes, equine encephalitis and other viral diseases transmitted to humans and other vertebrates (Linley et al. 1983, Borkent 2004, Purse et al. 2015). *Culicoides* (*Hoffmania*) *insignis* Lutz is a vector of Bluetongue virus (BTV) of domestic and wild ruminants in the Neotropics (Tanya et al. 1992). *Culicoides* (*Haematomyidium*) *paraensis* (Goeldi) is incriminated as a vector of oropouche (ORO) virus (Bastos et al. 2012). In Brazil, ORO is the etiological agent of the second most frequent cause of febrile arboviroses (Bastos et al. 2012). Moreover, species in the genera *Atrichopogon* Kieffer and *Forcipomyia* Meigen are ectoparasites of adult and immature insects, for example Lepidoptera and Odonata (Kawahara et al. 2006, Koptur et al. 2013, Nielsen et al. 2014).

The density of Ceratopogonidae larvae can be negatively affected by land use. In general, higher larval abundance and richness have been recorded in forested areas compared to deforested and non-forested areas (Ngai et al. 2008). Moreover, the increase of distance of deforested areas habitats from forest areas also negatively affects Ceratopogonidae fauna (Zimmer et al. 2014). In non-forested and deforested areas, habitats are generally more exposed and may inhibit oviposition of most species (Yanoviak 1999a, 2001). However, some species of Ceratopogonidae have been recorded at different urbanization levels and may occur in higher abundance in some anthropogenic areas (Rubio et al. 2013, Silva and Carvalho 2013).

Ceratopogonidae species are also influenced by intensity and frequency of rainfall and periods of drought (Yee and Juliano 2012, Ptatscheck and Traunspurger 2015). Moreover, communities present in phytotelmata or artificial containers must be adapted to successive evaporation processes (Kitching 2001). Biotic (e.g., predators) and abiotic factors (electrical conductivity, oxygen concentration, water temperature, and pH) of microhabitats may also influence Ceratopogonidae fauna (Cribb et al. 2003, Yanoviak 1999a). For example, a positive relationship between water volume (or containers size) or detritus availability (food) with abundance and richness of this family has been reported (Yanoviak 1999a, b, Gossner et al. 2016).

Ceratopogonidae oviposition can occur in different natural or artificial semi-aquatic and aquatic environments. Their immature stages may occur in diverse habitats in terrestrial (e.g., on wet wood) and aquatic environments (e.g., lakes, rivers, filamentous algae, phytotelmata, and brackish water). Some species are semi-aquatic and can be collected from different substrates (e.g., algae, woods, rocks, decaying fruit), animal footprints, or plants with high moisture contents (e.g., cacti and banana trees, Mercer et al. 2003).

In Central Amazon, taxonomic studies observed Ceratopogonidae development in the laboratory, and described or redescribed species and their life cycle (Ferreira-Keppler et al. 2014). In other taxonomic studies, immatures were sampled in different habitats, for example phytotelmata (Spinelli et al. 2007), macrophytes (Cazorla et al. 2012, Torreias et al. 2013, Díaz et al. 2014), organic polluted streams (Ronderos et al. 2008), and artificial containers in urban areas (Díaz et al. 2013, Ronderos et al. 2013), that analyzed the following genera: *Bezzia* Kieffer, *Culicoides* Latreille, *Dasyhelea* Kieffer, *Forcipomyia*, *Stilobezzia* Kieffer. We studied immatures of Ceratopogonidae in artificial containers in an urban forest fragment

in Manaus (Brazil) that focused on behavioral, biological, and ecological information. In addition, we evaluated the effects of deforestation (opened and closed areas) on Ceratopogonidae communities. We hypothesized higher abundance and richness in closed areas in relation to open areas, mainly due to the higher availability of detritus and better protection for ovipositing females.

## MATERIALS AND METHODS

### STUDY AREA

We sampled for Ceratopogonidae immatures in a forest fragment near the Instituto Nacional de Pesquisas da Amazônia (INPA), located in the urban area of Manaus (Brazil). The climate is tropical equatorial humid with rainy (November to May) and dry (June to October) seasons. Mean annual rainfall and mean humidity is 2,286 mm<sup>3</sup> and 80%, respectively, with a mean annual temperature of 26°C (min = 23°C, max = 31°C, Costa et al. 2013).

### SAMPLING PROCEDURE

Ceratopogonidae immatures were sampled monthly between August 2012 and July 2014 in two areas (open and closed) in an urban forest fragment. The open area (03°05'40.4"S; 59°59'22.3"W) has buildings and lacks a vegetation cover; therefore, we installed artificial containers (plastic boxes, Styrofoam boxes, and bamboo internodes) containing water and organic matter (leaves and sticks). The closed area (03°05'44.3"S; 59°59'21.9"W) is a secondary forest with closed vegetation cover and several exotic species (e.g., *Artocarpus heterophyllus* Lam. [Moraceae], *Mangifera indica* L. [Anacardiaceae], *Musa* sp. [Musaceae], *Phenakospermum guyannense* [Strelitziaceae] and *Caladium* sp. [Araceae]). We installed artificial containers (plastic boxes, Styrofoam boxes, cut bamboo, aluminum buckets, toilets, asbestos tanks, and tires) in the area. We used pipettes and plastic trays to collect immatures,

which were transported to the laboratory in 80-mL plastic bottles.

In the laboratory, the immatures were placed in individual plastic containers (2.5 x 2.5 cm) and kept at a constant temperature (~24°C). Every other day, we vacuumed the containers and replenished water and organic matter. Organic matter was obtained from bryophytes to avoid fungal proliferation (Ferreira and Rafael 2006). After emergence, exuviae of adults, larvae, and pupae were preserved in 70% ethanol. For identification, larvae, pupae and adults then dissected and slide-mounted in Canada Balsam (Borkent and Spinelli 2007). Examined, identified specimens were deposited in the Invertebrate Collection of INPA and the Collection of the Entomology Division of the Museu de La Plata (Argentina).

### ABIOTIC VARIABLES

Water temperature, pH, and electrical conductivity were measured monthly using portable equipment (waterproof pH meter, conductivity meter, and thermometer; Oakton Instruments, Vernon Hills, Illinois, USA). Water volume was measured with a graduated cylinder (mm).

### STATISTICAL ANALYSIS

To evaluate differences in abiotic variables between areas (open and closed) and seasons (rainy and dry), we used Repeated Measure Analysis of Variance (RM-ANOVA). This analysis was also used to test differences of abundance and richness of Ceratopogonidae between areas and seasons. Non-metric multidimensional scaling (NMDS; Sørensen distance) was used to verify the separation of invertebrate communities in relation to areas and seasons. Posteriorly, we tested the significant differences of NMDS groups using Multivariate Analysis of Variance (MANOVA). To perform MANOVA, we used abundance data and Sørensen distance. Similarity percentage analysis

(SIMPER, Clarke 1993) was used to determine the percentage of dissimilarity between areas and seasons. Moreover, SIMPER was used to determine the taxa that most contributed to these differences in invertebrate communities. We used Bray-Curtis similarity matrix and non-transformed data to perform SIMPER.

We modeled the relationship between abundance, richness, and composition (NMDS axis 1) and independent variables (pH, water temperature, electrical conductivity, and water volume) using multiple regression (backward elimination). We used Linear regression (normal distribution) for abundance and composition (continuous data) and a generalized linear model (GLM, Poisson distribution) for richness (count data). Prior to analysis, we assessed the multicollinearity ( $r > 0.7$ ) among explanatory variables by pair-wise correlation analyses. Model selection was done using Akaike's Information Criterion. All statistical analyses were performed in program R (R Core Team 2014), using the package "vegan" (Oksanen et al. 2017).

## RESULTS

### ABIOTIC VARIABLES

During the experiment, mostly in the rainy season, we recorded a high quantity of plant organic matter (leaves, small fruits, sticks and inflorescences) from the allochthonous vegetation in the open area. Mean pH was similar between areas ( $F_{1,17} = 0.70$ ,  $p = 0.414$ ) and seasons ( $F_{1,17} = 3.25$ ,  $p = 0.293$ , Table I). Mean water volume was higher in containers located in the open area ( $F_{1,17} = 15.10$ ,  $p = 0.001$ ) and in the rainy season ( $F_{1,17} = 63.45$ ,  $p < 0.001$ ). Water temperature varied between 25.6 and 29°C, with higher values in the open area ( $F_{1,17} = 16.87$ ,  $p = 0.001$ ); however, temperature was similar between the studied seasons ( $F_{1,17} = 0.49$ ,  $p = 0.492$ ). We recorded a significant interaction effect between area and seasons for electrical conductivity ( $F_{1,17} =$

12.26,  $p = 0.003$ ). In the closed area, conductivity values were slightly lower in the rainy than in the dry season. On the other hand, in the open area, conductivity increased in the dry season.

### FAUNA

We collected 685 immatures from seven species morphotypes (Appendix 1). Immatures were kept in the laboratory and 385 reached the adult stage (open area = 78; closed area = 307). In the open area, Ceratopogonidae were associated with Culicidae (*Aedes aegypti* Linn., *Aedes albopictus* Skuse and *Toxorhynchites* Theobald) Ephydriidae, Psychodidae, Phoridae, Oligochaeta, and Copepoda. In the closed area, we recorded Ceratopogonidae associated with Culicidae, Scatopsidae, Phoridae, and Cecidomyiidae.

*Forcipomyia* sp., *Culicoides* (*Haematomyidium*) *crucifer* Clastrier and *Dasyhelea eloyi* Díaz & Ronderos were recorded in both areas and seasons (Appendix 1). We recorded *Culicoides insignis* only in the open area during the rainy season. *Culicoides quasiparensis* was recorded only in the rainy season in both areas, while *Bezzia* sp. and *Palpomyia* sp. were found only in the closed area during both seasons (Appendix 1).

### BIONOMIC COMMENTS

Larvae of *Bezzia* sp. are elongated with a thin, prognathous cephalic capsule; coloration is slightly yellowish with dark spots visible through their transparent cuticles. They are predators and are known to feed on larvae of Culicidae and Chironomidae (Mullen and Hribar 1988) as well as larvae of some Ceratopogonidae (Grogan and Messersmith 1976). Their locomotion is serpentine, varying from extremely rapid in most aquatic species to extremely slow in others, are phototropic positive and were collected in the deepest parts of the containers within the organic matter. Pupae are dark brown, elongated with pointed terminal

**TABLE I**  
**Results of Repeated Measure Analysis of Variance (RM-ANOVA) to test differences in abiotic variables between studied seasons and area; minimum (Min), maximum (Max), mean, and standard deviation (SD) values of abiotic variables in artificial containers in an urban forest fragment in Central Amazon.**

Water abiotic variable	Area	Seasons	Min - Max	Mean $\pm$ SD	RM-ANOVA					
					Seasons		Area		Seasons * Area	
					$F_{1,17}$	$p$	$F_{1,17}$	$p$	$F_{1,17}$	$p$
pH	Open	Rainy	7.25 – 8.17	7.73 $\pm$ 0.31	3.25	0.293	0.70	0.414	1.23	0.282
		Dry	6.57 – 8.50	7.67 $\pm$ 0.58						
	Closed	Rainy	7.67 – 8.21	8.03 $\pm$ 0.17						
		Dry	7.23 – 8.26	7.72 $\pm$ 0.40						
Volume (ml)	Open	Rainy	433.33 – 600.00	523.33 $\pm$ 59.81	63.45	< 0.001	15.10	0.001	0.48	0.496
		Dry	200.00 – 500.00	316.25 $\pm$ 97.60						
	Closed	Rainy	358.33 – 528.57	426.08 $\pm$ 62.22						
		Dry	176.67 – 344.29	249.55 $\pm$ 61.15						
Temperature (°C)	Open	Rainy	26.75 – 29.00	27.47 $\pm$ 0.70	0.49	0.492	16.87	0.001	0.06	0.813
		Dry	26.00 – 28.30	27.60 $\pm$ 0.77						
	Closed	Rainy	25.57 – 27.60	26.75 $\pm$ 0.57						
		Dry	26.29 – 27.50	26.96 $\pm$ 0.44						
Electrical conductivity ( $\mu$ S/m)	Open	Rainy	207.14 – 278.33	232.51 $\pm$ 23.34	1.59	0.225	18.66	< 0.001	12.26	0.003
		Dry	228.57 – 356.67	286.43 $\pm$ 53.21						
	Closed	Rainy	216.67 – 380.00	297.58 $\pm$ 47.91						
		Dry	230.00 – 366.67	288.33 $\pm$ 44.93						

processes. Pupae exhibit circular movements and were collected in the water slide. During adult emergence, pupae are free or adhere to vegetation and other substrates. The development time of 4<sup>th</sup> instar larvae to pupae was about 15 days, and, from pupa to adult was two to three days.

Larvae of *C. crucifer* are yellowish. They swim rapidly and exhibited greater activity when placed under incandescent light. Larvae swam serpent-like within organic substrate where they remained before pupation. Pupae were also yellowish and occurred alongside coarse particles of debris; they could be collected from margins of aquatic habitats. The development time of 4<sup>th</sup> instar larvae to pupae was about 25 days, and, from pupa to adult was two to three days.

Larvae of *D. eloyi* were actively creeping and moved towards the bottoms and sides of the

breeding grounds, which are usually shallow (~3 cm). They fed on loose or aggregated, coiled to organic fragments deposited in the bottom of the breeding places next to aquatic larvae of Psychodidae, Ephydriidae, and Phoridae. They exhibited a hypognathous cephalic capsule, pale brown in color and grayish at the dorsal part, with spots along the segments of the thorax and abdomen. At the last instar and in the pupa stage, they had a pale yellow coloration. The development time of the larvae (4<sup>th</sup> instar) to pupae was about 17 days and that from pupa to adult was one to three days.

Larvae of *Forcipomyia (Euprojoannisia)* sp. had a yellowish coloration. They exhibited slow movements and could be found on the water slide; in some cases, they were dragging themselves along the wall of the nursery or along leaves and



debris. The pupa as yellowish in color. During pupation, the exudation of the larva was retained to the last segments of the abdomen of the pupa. The cephalothorax as darker than the abdomen; segment 9 with elongated and pointed process. Pupae had little mobility, adhering to the side of the nursery often close to the water table until adult emergence. The development time of the larvae (4<sup>th</sup> instar) to pupae was five days and that from pupa to adult was two to three days.

Larvae of *Palpomyia* sp. were collected near the submerged leaf litter. These larvae were yellowish in color, with darker longitudinal bands visible through transparency. The cephalic capsule was elongated, light brown in color and the caudal segment with wide setae. Larvae swam rapidly, with fast serpentine wave movements. They are predators of small larvae of Culicidae, Chironomidae and other Ceratopogoninae that cohabit these environments. Pupae were dark brown in color, with an elongate, pointed terminal processes. They were found along water margins or adhered to substrates and exhibited lateral movements of their abdomens. The development time of 4<sup>th</sup> instar larvae to pupations, was about 20 days, and from pupa to adult in 2–3 days.

#### FAUNA × ABIOTIC VARIABLES

In the open area, we recorded Ceratopogonidae during only 9 (37.5%) of 24 studied months; however, during the dry season of 2013, no individuals were collected (Figure 1). Conversely, we collected immatures during 18 months (75%) in the closed area. *Forcipomyia* sp. and *D. eloyi* were the most abundant taxa. *Forcipomyia* sp. was recorded in higher abundance during the dry season, while *D. eloyi* occurred in higher abundance during the rainy season (Appendix 1). Mean abundance was similar between seasons (rainy =  $3.07 \pm 5.35$ , dry =  $1.85 \pm 3.73$ ,  $F_{1,22} = 0.74$ ,  $p = 0.398$ ) and areas (open =  $1.51 \pm 3.70$ , closed =  $3.40 \pm 5.26$ ,  $F_{1,22} =$

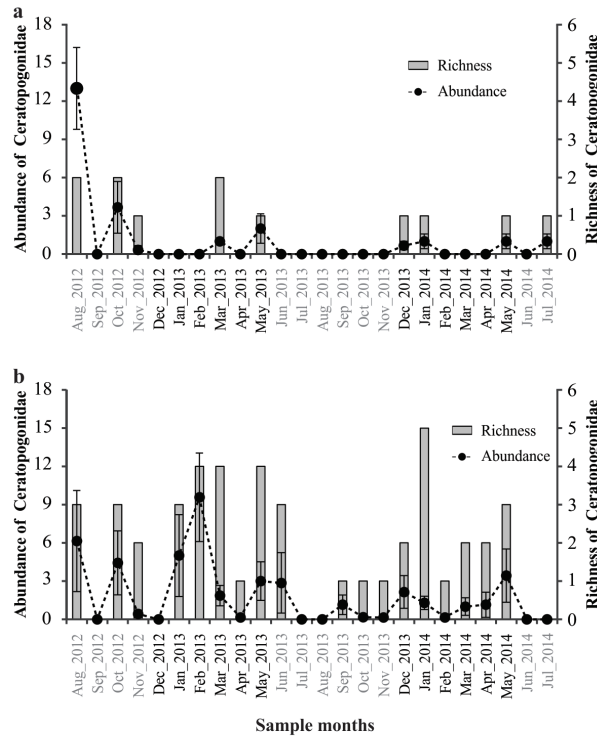
$2.44$ ,  $p = 0.132$ ). The interaction effect between area and seasons on abundance was not significant ( $F_{1,22} = 0.80$ ,  $p = 0.380$ ). Ceratopogonidae richness was high in the closed area ( $2.00 \pm 1.38$ ) in relation to the open area ( $0.75 \pm 1.03$ ,  $F_{1,22} = 24.03$ ,  $p < 0.001$ ); however, richness was similar between seasons (rainy =  $1.50 \pm 1.25$ , dry =  $1.25 \pm 1.48$ ,  $F_{1,22} = 0.33$ ,  $p = 0.573$ ). The interaction effect between area and seasons on richness was not significant ( $F_{1,22} = 0.11$ ,  $p = 0.747$ ).

According to NMDS, it was possible to separate the Ceratopogonidae community into open and closed areas along the first axis; however, there were no differences between the two seasons (Figure 2). Community composition differed between areas (MANOVA:  $F_{1,27} = 7.58$ ,  $p = 0.001$ ), but not between studied seasons (MANOVA:  $F_{1,27} = 3.08$ ,  $p = 0.155$ ). The dissimilarity between open and closed areas was 82.31%. *Forcipomyia* sp., and *Culicoides crucifer*, *D. eloyi*, and *Bezzia* sp. (Table II) were the taxa that most contributed to this dissimilarity.

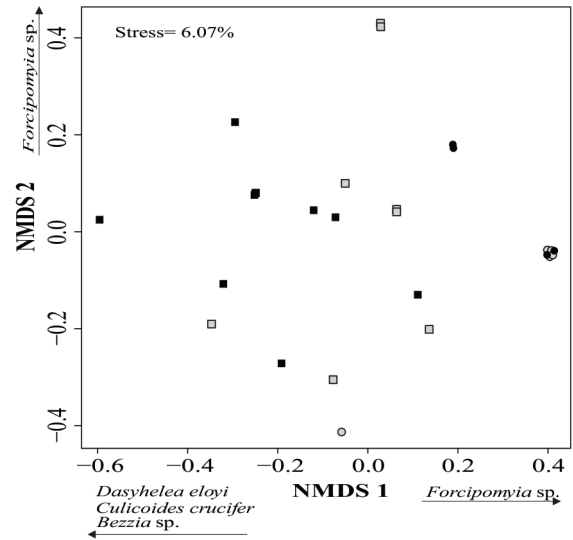
The selected multiple regression model for abundance explained 8% of the data variation. Ceratopogonidae abundance was negatively related to container water volume (Table III). For richness and composition, the selected multiple regression models explained 29 and 36% of the variation, respectively. Both metrics were positively related to electrical conductivity and negatively to water temperature (Table III).

#### DISCUSSION

*Forcipomyia* (*Euprojoannisia*) is a cosmopolitan subgenus with five records for Brazil in the states of Rio de Janeiro, Bahia, Santa Catarina, and São Paulo (Borkent and Spinelli 2007). Their larvae are semi-aquatic and have been found in floating vegetation, plant concavities (phytotelmata), tree holes, mosses, rock algae, dams with shallow marshes, mud, river margins, macrophytes, and



**Figure 1** - Mean abundance and richness of Ceratopogonidae sampled between August 2012 and July 2014 in artificial containers located in (A) open area and (B) closed area in an urban forest fragment in Central Amazon. Sample months in black = rainy season; Sample months in gray = dry season.



**Figure 2** - Non-Metric Multidimensional Scaling (NMDS) and taxa of Ceratopogonidae selected by Similarity percentage tests (SIMPER) to explain the dissimilarity between open and closed areas in terms of Ceratopogonidae abundances in artificial containers located in an urban forest fragment in Central Amazon. Gray square = open area in rainy season; Black square = open area in dry season; Gray circle = closed area in rainy season; Black circle = closed area in dry season.

**TABLE II**  
Mean abundance (SD) and results of the Similarity Percentage Tests (SIMPER) indicating the contribution of Ceratopogonidae taxa to the dissimilarity between open and closed areas in an urban forest fragment in Central Amazon. In bold, values of dissimilarity (%) between groups. Standard deviation values are given in parentheses.

Taxa	Mean abundance		Contribution to dissimilarity (%)
	Open	Closed	<b>82.30</b>
<i>Forcipomyia</i> sp.	1.04 (2.88)	0.58 (1.90)	27.03
<i>Culicoides crucifer</i>	0.29 (0.57)	0.64 (1.03)	21.74
<i>Dasyhelea eloyi</i>	0.13 (0.43)	0.94 (2.09)	15.91
<i>Bezzia</i> sp.	0.00 (0.00)	0.77 (1.94)	11.50

**TABLE III**  
Results of the multiple regression modeling the abundance, richness, and composition (Non-Metric Multidimensional Scaling – Axis 1) of Ceratopogonidae from an urban forest fragment in Central Amazon. In bold, significant *p*-values.

	Abundance			Richness			Composition		
	Coefficients	t value	<i>p</i>	Coefficients	t value	<i>p</i>	Coefficients	t value	<i>p</i>
Intercept	4.627	3.191	0.003	10.037	2.33	0.061	3.914	2.42	<b>0.023</b>
Volume (ml)	-0.007	-2.056	<b>0.047</b>	-	-	-	-	-	-
Temperature (°C)	-	-	-	-0.421	-2.214	<b>0.027</b>	-0.151	2.168	<b>0.040</b>
Electrical conductivity (µS/m)	-	-	-	0.006	2.416	<b>0.016</b>	0.002	2.510	<b>0.019</b>

epiphytic bromeliads (Winder and Silva 1972, Winder 1977, Debenham and Wirth 1984, Torreias et al. 2013). We recorded a higher abundance of larvae of *Forcipomyia* sp. in containers in the open area. Larvae of this genus utilize algae as an important food source (Yeh and Chuang 1996, Wen et al. 2011, Chen et al. 2016). Containers in the open area received a higher incidence of light, which can stimulate algae growth in debris, thereby providing more available food for species of *Forcipomyia*. However, in rainforest areas with low density, primary productivity in aquatic systems is much lower (Vannote et al. 1980).

We recorded a greater abundance of *C. crucifer* and *D. eloyi* in the closed area. In previous studies in tropical regions, larvae of *D. eloyi* were associated with leaves of aquatic macrophytes (Díaz et al. 2013, Torreias et al. 2013). *Culicoides* larvae occur in a wide variety of aquatic and semi-aquatic habitats and feed on debris with abundant organic matter (Silva and Carvalho 2013). Thus, the higher abundance of these two species in the closed area may be associated with higher inputs of detritus (mainly leaves) in the containers. In taxonomic studies of *D. eloyi* and *C. crucifer* with Amazonian material, both species were recorded in artificial containers only in closed areas (Díaz et al. 2013, Ronderos et al. 2013).

The larvae of *C. insignis* were recorded only in the open area during a single sampling event in the rainy season. In Brazil, this species has previously been found in street ditches with still water, small water ponds, and holes dug by crustaceans, *Oedipleura cordata* (Forattini et al. 1956). For adults of *C. insignis*, higher abundance was recorded in anthropic environments (forest edges) compared to forested areas (Veggiani Aybar et al. 2011). The abundance of adults of *C. insignis* is positively correlated with high temperature, precipitation, and humidity (Veggiani Aybar et al. 2012).

Immatures of *Bezzia* sp. and *Palpomyia* sp. were recorded only in the closed area. The larvae of these genera are predators (Mullen and Hribar 1988) and feed on other insects (e.g., Chironomidae and Ephydriidae, Collins 1975) or other invertebrates (e.g. Copepoda, Ronderos et al. 2004). Thus, the presence of *Bezzia* sp. and *Palpomyia* sp. may be related to the greater abundance of non-Ceratopogonidae invertebrates in containers in the closed area as well as the presence of Ephydriidae and Copepoda only in this area.

We recorded higher richness in the closed area compared to the open area; however, abundance was similar in both areas. In general, higher invertebrate richness and abundance are commonly observed in forested rather than non-forested areas, and these areas also differ in species composition (Blaustein et al. 1999, Ngai et al. 2008, Seger et al. 2012). These differences can be attributed mainly to the greater input of detritus in containers in the closed area (Kling et al. 2007, Yee and Juliano 2007). Thus, closed areas provide higher availability of food and habitats for different species, which can stimulate oviposition (Reiskind and Wilson 2004). Moreover, other factors may influence the community of Ceratopogonidae, such as predation or abiotic variables (Seger et al. 2012, Rubio et al. 2013).

We observed a difference in the community of Ceratopogonidae (richness, abundance or composition) between the rainy and the dry season. In general, temperature and precipitation significantly impact insects of phytotelmata and affects their development time as well as the availability of habitats for these organisms (Armbruster et al. 2002, Dézerald et al. 2015, Gossner et al. 2016). However, in our study, water temperature did not differ between the two seasons. In addition, we did not record any dried containers (minimum volume = 176.67 mL), which may have reduced the effects of precipitation on Ceratopogonidae (Rangel et al. 2017). In addition, in the Amazon region, the



higher leaf inputs occur in the dry season (Tonin et al. 2017), which may have attenuated the lower water volume in containers in this season.

In general, the phytotelmata invertebrate fauna is influenced by food availability, water parameters, and seasonality (Torreias and Ferreira-Keppler 2011, Dézerald et al. 2016). In our study, pH was the only variable that did not significantly affect the Ceratopogonidae fauna. In general, when extreme pH values are not recorded, it is expected that this variable does not strongly affect the community of invertebrates in phytotelmata (Yanoviak 1999b).

In our study, richness and composition were positively associated with electrical conductivity in water containers. Higher values of electrical conductivity result from increased ion concentrations and indicate increased nutrient concentrations in the water (Leite et al. 2016). Thus, higher nutrient values may be associated with the decomposition of allochthonous organic matter (mainly leaves) in containers (Gessner et al. 1999). These higher amounts of nutrients can be used by micro-organisms (e.g., fungi, bacteria, or algae), which are then consumed by invertebrates. Thus, increased food availability results in a more diverse community. In addition, electrical conductivity may be related to the leaching of the detritus (e.g., leaves, branches, and flowers) present in the containers (Yanoviak 1999b).

We recorded a decrease in the abundance of Ceratopogonidae with an increase of the water volume in their containers. In general, there is a positive relationship between invertebrate fauna (abundance and richness) and volume in phytotelmata (Schmidl et al. 2008, Jabiol et al. 2009, Campos 2013, Dézerald et al. 2014). However, water volume has a higher influence on the abundance of invertebrates in environments with frequent droughts, mainly due to egg dehydration and larval mortality (Yanoviak 1999a, Paradise 2004). We did not observe completely dried containers, even in the dry season. In

addition, the number of competitors and predators may be another factor that contributed to the lower abundance of Ceratopogonidae in containers with high water volumes. In previous studies, these organisms have been frequently recorded in higher abundance in larger phytotelmata (Srivastava and Lawton 1998, Yanoviak 1999a).

We observed a negative relation between temperature and Ceratopogonidae richness and composition. Temperature is one of the main abiotic variables that influence the development of immature insects (Spinelli et al. 2005, Bernotiene and Bartkeviciene 2013, Martins et al. 2017). In general, higher temperature values tend to be related to higher abundance of invertebrates (Mayo et al. 2014). However, this increase is not linear and is generally associated with the thermal tolerance limit of each species. In addition, higher temperature values may be associated with lower oxygen values and, consequently, less diverse fauna (Ngai et al. 2008, Fulan et al. 2009).

In conclusion, high conductivity values resulted in high species richness. On the other hand, high water temperature and volume resulted in low richness and abundance levels, respectively. Ceratopogonidae communities did not significantly differ between the two seasons; however, we recorded a higher richness in the closed area. According to our results, the preservation of forested areas is fundamental for the maintenance of the life cycle of some species of Ceratopogonidae.

#### ACKNOWLEDGMENTS

We thank to Daniel de Paula and Laboratório de Insetos Aquáticos (INPA/ COBIO) for help on fieldwork. We are especially grateful to Dr. L. William Grogan for critical reading of the manuscript, which provided a much appreciated and detailed review of our manuscript. EDRD received the fellowships by PAIC/FAPEAM. RLFK received financial support by CNPq

(processes 306081/2013-0; 483130/2013-3). RTM received a fellowship from the Programa de Apoio à Fixação de Doutores no Amazonas—FIXAM/AM, Fundação de Amparo à Pesquisa do Estado do Amazonas – (FAPEAM) and Programa Nacional de Pós-Doutorado – (PNPD)/ Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES) Programa Nacional de Pós-Doutorado – (PNPD)/Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES)/Programa de Pós-Graduação em Ecologia e Evolução (PPGEE)/UFG fellowships. We thanks Programa de Apoio a Núcleos de Excelência (Pronex)/ Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq)/FAPEAM—Aquatic insects and INCT ADAPTA-II (CNPq/FAPEAM) supported the invertebrate sampling.

## REFERENCES

- ARMBRUSTER P, HUTCHINSON RA AND COTGREAVE P. 2002. Factors influencing community structure in a South American tank bromeliad fauna. *Oikos* 96: 225-234.
- BASTOS MS, FIGUEIREDO LTM, NAVECA FG, MONTE RL, LESSA N, FIGUEIREDO RMP, GIMAQUE JBL, JOÃO GP, RAMAZAWMY R AND MOURÃO MPG. 2012. Identification of Oropouche *Orthobunyavirus* in the cerebrospinal fluid of three patients in the Amazonas, Brazil. *Am J Trop Med Hyg* 86: 732-735.
- BERNOTIENE R AND BARTKEVICIENE G. 2013. The relationship between water temperature and the development cycle beginning and duration in three black fly species. *J Insect Sci* 13: 1-15.
- BLAUSTEIN L, GARBJE, SHEBITZ D AND NEVO E. 1999. Microclimate, developmental plasticity and community structure in artificial temporary pools. *Hydrobiologia* 392: 187-196.
- BORKENT A. 2004. Ceratopogonidae. In: Marquardt WC (Ed), *Biology of Disease Vectors*, Elsevier Press, Amsterdam, Netherlands, p. 113-126.
- BORKENT A. 2016. World species of Biting Midges (Diptera: Ceratopogonidae). Available from: <http://www.inhs.illinois.edu/research/flytree/borkent>. Accessed: 13/01/2017.
- BORKENT A AND SPINELLI GR. 2007. Neotropical Ceratopogonidae (Diptera: Insecta). In: Adis J et al. (Eds), *Aquatic Biodiversity in Latin America (ABLA)*. v. 4. Sofia – Moscow, Russian, p. 198.
- CAMPOS RE. 2013. The aquatic communities inhabiting internodes of two sympatric bamboos in Argentinean subtropical forest. *J Insect Sci* 13: 93-110.
- CAZORLA CG, RONDEROS MM, SPINELLI GR, TORREIAS SRS AND FERREIRA-KEPPLER RL. 2012. A new species of *Stilobezzia* Kieffer from the Neotropical Region (Diptera, Ceratopogonidae). *Rev Bras Entomol* 56: 399-404.
- CHEN HW, CHOU JY, LIN CC, WEN YD AND WANG WL. 2016. Seasonal yeast compositions in *Forcipomyia taiwana* (Diptera: Ceratopogonidae). *J Asia Pac Entomol* 19: 509-514.
- CLARKE KR. 1993. Non-parametric multivariate analyses of changes in community structure. *Aust J Ecol* 18: 117-143.
- COLLINS NC. 1975. Population biology of a brine fly (Diptera: Ephydriidae) in the presence of abundant algal food. *Ecology* 56: 1139-1148.
- COSTA ACL, SILVA-JUNIOR JA, CUNHA AC, FEITOSA JRP, PORTELA BTT, SILVA GGC AND COSTA RF. 2013. Índices de conforto térmico e suas variações sazonais em cidades de diferentes dimensões na Região Amazônica. *Rev Bras Geogr Fis* 6: 478-487.
- CRIBB BW, BREI B, RIDLEY AW AND MERRITT DJ. 2003. Occurrence of immature *Culicoides molestus* (Skuse) (Diptera: Ceratopogonidae) in relation to habitat characteristics. *Aust J Entomol* 42: 114-118.
- DEBENHAM ML AND WIRTH WW. 1984. Australian and New Guinea Species of the *Forcipomyia* Subgenus *Euprojoannisia* (Diptera: Ceratopogonidae). *Aust J Zool* 32: 851-889.
- DÉZÉRALD O, CÉRÉGHINO R, CORBARA B, DEJEAN A AND LEROY C. 2015. Temperature: diet interactions affect survival through foraging behavior in a bromeliad-dwelling predator. *Biotropica* 47: 569-578.
- DÉZÉRALD O, LEROY C, CORBARA B, DEJEAN A, TALAGA S AND CÉRÉGHINO R. 2016. Environmental drivers of invertebrate population dynamics in Neotropical tank bromeliads. *Freshw Biol* 62: 229-242.
- DÉZÉRALD O, STANISLAS T, LEROY C, CARRIAS J, CORBARA B, DEJEAN A AND CÉRÉGHINO R. 2014. Environmental determinants of macroinvertebrate diversity in small water bodies: insights from tank-bromeliads. *Hydrobiologia* 723: 77-86.
- DÍAZ F, RONDEROS MM, SPINELLI GR, FERREIRA-KEPPLER RL AND TORREIAS SRS. 2013. A new species of *Dasyhelea* Kieffer (Diptera: Ceratopogonidae) from Brazilian Amazonia. *Zootaxa* 3686: 85-93.
- DÍAZ F, TORREIAS SRS, SPINELLI GR AND RONDEROS MM. 2014. A new species of *Dasyhelea* from Brazilian Amazonas and description of male of *D. paulistana* (Diptera: Ceratopogonidae). *Acta Entomol Mus Natl Pragae* 54: 715-728.

- FERREIRA RLM AND RAFAEL JA. 2006. Criação de imaturos de mutuca (Tabanidae: Diptera) utilizando briófitas e areia como substrato. *Neotrop Entomol* 35: 141-144.
- FERREIRA-KEPPLER RL, RONDEROS MM, DÍAZ F, SPINELLI GR AND TORREIAS SRS. 2014. Ordem Diptera. Família Ceratopogonidae. In: Hamada N et al. (Eds), *Insetos Aquáticos na Amazônia brasileira: taxonomia, biologia e ecologia*. Editora INPA. Manaus, Brasil, p. 711-723.
- FORATTINI OP, RABELLO EX AND PATTOLI D. 1956. Nota sobre a larva e pupa de *Culicoides insignis* Lutz, 1913 (Diptera: Ceratopogonidae). *Rev Bras Entomol* 4: 195-198.
- FULAN JA, DAVANSO R AND HENRY R. 2009. A variação nictemeral das variáveis físicas e químicas da água influencia a abundância dos macroinvertebrados aquáticos. *Rev Bras Biosc* 7: 150-154.
- GESSNER MO, CHAUVET E AND DOBSON M. 1999. A perspective on leaf litter breakdown in streams. *Oikos* 85: 377-384.
- GOSSNER MM, LADE P, ROHLAND A, SICHARDT N, KAHL T, BAUHUS J, WEISSER WW AND PETERMANN JS. 2016. Effects of management on aquatic tree-hole communities in temperate forests are mediated by detritus amount and water chemistry. *J Anim Ecol* 85: 213-226.
- GROGAN WL AND MESSERSMITH DH. 1976. The immature stages of *Alluaudomyia paraspina* (Diptera: Ceratopogonidae) Wirth with notes on its biology. *Ann Entomol Soc Am* 69: 687-690.
- JABIOL J, CORBARA B, DEJEAN A AND CÉRÉGHINO R. 2009. Structure of aquatic insect communities in tank-bromeliads in an East-Amazonian rainforest in French Guiana. *Forest Ecol Manag* 257: 351-360.
- KAWAHARA AY, WINKLER IS AND HSU WW. 2006. New host records of the ectoparasitic biting midge *Forcipomyia (Trichohalea) pectinunguis* (Diptera: Ceratopogonidae) on adult geometrid moths (Lepidoptera: Geometridae). *J Kans Entomol Soc* 79: 297-300.
- KITCHING RL. 2001. Food webs in phytotelmata: "bottom-up" and "top-down" explanations for community structure. *Annu Rev Entomol* 46: 729-760.
- KLING LJ, JULIANO SA AND YEE DA. 2007. Larval mosquito communities in discarded vehicle tires in a forested and unforested site: detritus type, amount, and water nutrient differences. *J Vector Ecol* 32: 207-217.
- KOPTUR S, PENA JE AND GROGAN JR WL. 2013. The biting midge, *Forcipomyia (Microhelea) eriophora* (Williston) (Diptera: Ceratopogonidae), an ectoparasite of larval *Phoebis sennae* (Pieridae) in South Florida. *J Lepid Soc* 67: 128-130.
- LEITE GFM, SILVA FTC, NAVARRO FKSP, REZENDE RDS AND GONÇALVES JÚNIOR JF. 2016. Leaf litter input and electrical conductivity may change density of *Phylloicus* sp. (Trichoptera: Calamoceratidae) in a Brazilian savannah stream. *Acta Limnol Bras* 28: 12-21.
- LINLEY JR, HOCH AL AND PINHEIROS FP. 1983. Biting midges (Diptera: Ceratopogonidae) and human health. *J Med Entomol* 20: 347-364.
- MARTINS RT, MELO AS, GONÇALVES JF, CAMPOS CM AND HAMADA N. 2017. Effects of climate change on leaf breakdown by microorganisms and the shredder *Phylloicus elektoros* (Trichoptera: Calamoceratidae). *Hydrobiologia* 789: 31-44.
- MAYO CE, OSBORNE CJ, MULLENS BA, GERRY AC, GARDNER IA, REISEN WK, BARKER CM AND MACLACHLAN NJ. 2014. Seasonal variation and impact of waste-water lagoons as larval habitat on the population dynamics of *Culicoides sonorensis* (Diptera: Ceratopogonidae) at two dairy farms in northern California. *PLoS ONE* 9: e89633.
- MERCER DR, SPINELLI GR, WATTS DM AND TESH RB. 2003. Biting rates and developmental substrates for biting midges (Diptera: Ceratopogonidae) in Iquitos, Peru. *J Med Entomol* 40: 807-812.
- MULLEN GR AND HRIBAR LJ. 1988. Biology and feeding behavior of ceratopogonid larvae (Diptera: Ceratopogonidae) in North America. *Bull Soc Vector Ecol* 13: 60-81.
- NGAI JT, KIRBY KR, GILBERT B, STARZOMSKI BM, PELLETIER AJ AND CONNER JR. 2008. The impact of land-use change on larval insect communities: Testing the role of habitat elements in conservation. *EcoScience* 15: 160-168.
- NIELSEN ER, MANGER R AND MARTENS A. 2014. First records of *Forcipomyia paludis* (Diptera: Ceratopogonidae), a midge parasitising dragonfly adults (Odonata: Libellulidae), for the Balearic Islands, Spain. *Not Odonatol* 8: 77-116.
- OKSANEN J ET AL. 2017. Vegan: Community ecology package. R package version 2.4-3. <https://CRAN.R-project.org/package=vegan>.
- PARADISE CJ. 2004. Relationship of water and leaf litter variability to insects inhabiting treeholes. *J N Am Benthol Soc* 23: 793-805.
- PTATSCHECK C AND TRAUNSPURGER W. 2015. Meio- and Macrofaunal communities in artificial water-filled tree holes: Effects of seasonality, physical and chemical parameters, and availability of food resources. *PLoS ONE* 10: e0133447.
- PURSE BV, CARPENTER S, VENTER GJ, BELLIS G AND MULLENS BA. 2015. Bionomics of temperate and tropical *Culicoides* midges: knowledge gaps and consequences

- for transmission of *Culicoides*-borne viruses. *Annu Rev Entomol* 60: 373-392.
- R CORE TEAM. 2014. R: a language and environment for statistical computing. R Foundation for Statistical Computing, Vienna.
- RANGEL JV, ARAÚJO RE, CASOTTI CG, COSTA LC, KIFFER JR WP AND MORETTI MS. 2017. Assessing the role of canopy cover on the colonization of phytotelmata by aquatic invertebrates: an experiment with the tank-bromeliad *Aechmea lingulata*. *J Limnol* 76: 230-239.
- REISKIND MH AND WILSON ML. 2004. *Culex restuans* (Diptera: Culicidae) oviposition behavior determined by larval habitat quality and quantity in Southeastern Michigan. *J Med Entomol* 41: 179-186.
- RONDEROS MM, CAZORLA CG, SPINELLI GR AND CARRASCO DS. 2008. Description of immature stages and adult diagnosis of *Stilobezzia coquillettii* Kieffer 1917 (Diptera: Ceratopogonidae). *Zootaxa* 1958: 31-40.
- RONDEROS MM, SPINELLI GR AND DÍAZ F. 2004. Description of larva and redescription of pupa and adults of *Palpomyia guarani* (Diptera: Ceratopogonidae). *Rev Soc Entomol Argent* 63: 45-54.
- RONDEROS MM, SPINELLI GR AND FERREIRA-KEPPLER RL. 2013. Description of the Pupa of *Culicoides crucifer* Clastrier (Diptera: Ceratopogonidae). *Neotrop Entomol* 42: 492-497.
- RUBIO A, BELLOCQ IM AND VEZZANI D. 2013. Macro and microenvironmental factors affecting tyre-breeding flies (Insecta: Diptera) in urbanised areas. *Ecol Entomol* 38: 303-314.
- SANTARÉM MCA AND FELIPPE-BAUER ML. 2016. *Espécies de Maruins do Brasil* (Diptera: Ceratopogonidae). FIOCRUZ, Rio de Janeiro, Brasil, 67 p.
- SCHMIDL J, SULZER P AND KITCHING RL. 2008. The insect assemblage in water filled tree-holes in a European temperate deciduous forest: community composition reflects structural, trophic and physicochemical factors. *Hydrobiologia* 598: 285-303.
- SEGER KR, SMILEY JR PC, KING KW AND FAUSEY NR. 2012. Influence of riparian habitat on aquatic macroinvertebrate community colonization within riparian zones of agricultural headwater streams. *J Freshw Ecol* 27: 393-407.
- SILVA FS AND CARVALHO LPC. 2013. A population study of the *Culicoides* Biting midges (Diptera: Ceratopogonidae) in urban, rural, and forested sites in a cerrado area of northeastern Brazil. *Ann Entomol Soc Am* 106: 463-470.
- SPINELLI GR, RONDEROS M AND DÍAZ F. 2005. The bloodsucking biting midges of Argentina (Diptera: Ceratopogonidae). *Mem Inst Oswaldo Cruz* 100: 137-150.
- SPINELLI GR, RONDEROS MM, MARINO PI, CARRASCO DS AND FERREIRA RLM. 2007. Description of *Culicoides (Mataemyia) felippebaueri* sp. n., *Forcipomyia musae* immatures, and occurrence of *F. genualis*, breeding in banana stems in Brazilian Amazonia (Diptera: Ceratopogonidae). *Mem Inst Oswaldo Cruz* 102: 659-669.
- SPINELLI GR AND WIRTH WW. 1993. Los Ceratopogonidae de la Argentina (Insecta: Diptera). In: Castellanos ZA (Ed), *Fauna de agua dulce de la República Argentina*, Profadu (Conicet), La Plata, Argentina, p. 1-121.
- SRIVASTAVA DS AND LAWTON JH. 1998. Why more productive sites have more species: an experimental test of theory using tree-hole communities. *Am Nat* 52: 510-529.
- TANYAVN, GREINER EC AND GIBBS EPJ. 1992. Evaluation of *Culicoides insignis* (Diptera: Ceratopogonidae) as a vector of bluetongue virus. *Vet Microbiol* 31: 1-14.
- TONIN AM ET AL. 2017. Plant litter dynamics in the forest-stream interface: precipitation is a major control across tropical biomes. *Scientific Reports* 7: 10799.
- TORREIAS SRDS AND FERREIRA-KEPPLER RL. 2011. Macroinvertebrates inhabiting the tank leaf terrestrial and epiphyte bromeliads at Reserva Adolpho Ducke, Manaus, Amazonas. *Braz Arch Biol Technol* 54: 1193-1202.
- TORREIAS SRS, FERREIRA-KEPPLER RL AND RONDEROS MM. 2013. Biting midges (Ceratopogonidae: Diptera) present in aquatic macrophytes from wetlands of Marchantaria Island, Iranduba, Central Amazonia, Brazil. *J Nat Hist* 48: 109-122.
- VANNOTE RL, MINSHALL GW, CUMMINS KW, SEDELL JR AND CUSHING CE. 1980. The river continuum concept. *Can J Fish Aquat Sci* 37: 130-137.
- VEGGIANI AYBAR CA, JURI MJD DE GROSSO MSL AND SPINELLI GR. 2011. Spatial and temporal distribution of *Culicoides insignis* and *Culicoides paraensis* in the subtropical mountain forest of Tucumán, northwestern Argentina. *Fla Entomol* 94: 1018-1025.
- VEGGIANI AYBAR CA, JURI MJD, SANTANA M, DE GROSSO MSL AND SPINELLI GR. 2012. The spatio-temporal distribution patterns of biting midges of the genus *Culicoides* in Salta province, Argentina. *J Insect Sci* 12: 145-154.
- WEN YD, LIN CC, LIOU WM AND WANG WL. 2011. *Wolbachia* in field populations of *Forcipomyia taiwana* (Diptera: Ceratopogonidae) in Taiwan. *J Asia Pac Entomol* 14: 341-348.
- WINDER JA. 1977. Field observations on Ceratopogonidae and other Diptera: Nematocera associated with cacao flowers in Brazil. *Bull Entomol Res* 67: 57-63.
- WINDER JA AND SILVA P. 1972. Cacao pollination: Microdiptera of cacao plantations and some of their breeding places. *Bull Entomol Res* 67: 651-655.
- YANOVIK SP. 1999a. Community structure in water-filled tree holes of Panama: effects of hole height and size. *Selbyana* 20: 106-115.



- YANOVIK SP. 1999b. Effects of leaf litter species on macroinvertebrate community properties and mosquito yield in Neotropical tree hole microcosms. *Oecologia* 120: 147-155.
- YANOVIK SP. 2001. The macrofauna of water-filled tree holes on Barro Colorado Island, Panama. *Biotropica* 33: 110-120.
- YEE DA AND JULIANO SA. 2007 Abundance matters. A field experiment testing the More Individuals Hypothesis for richness-productivity relationships. *Oecologia* 153: 153-162
- YEE DA AND JULIANO SA. 2012. Concurrent effects of resource pulse amount, type, and frequency on community and population properties of consumers in detritus-based systems. *Oecologia* 169: 511-522.
- YEH CC AND CHUANG YY. 1996. Colonization and bionomics of *Forcipomyia taiwana* (Diptera: Ceratopogonidae) in the laboratory. *J Med Entomol* 33: 445-448.
- ZIMMER JY, BROSTAUZ Y, HAUBRUGE E AND FRANCIS F. 2014. Larval development sites of the main *Culicoides* species (Diptera: Ceratopogonidae) in northern Europe and distribution of coprophilic species larvae in Belgian pastures. *Vet Parasitol* 205: 676-686.

### Appendix 1

Mean and standard deviation of Ceratopogonidae sampled between August 2012 and July 2014 in artificial containers in an urban forest fragment in Central Amazon.

Month	Seasons	Area	<i>Forcipomyia</i> sp.	<i>Dasyhelea</i> <i>eloyi</i>	<i>Culicoides</i> <i>crucifer</i>	<i>Culicoides</i> <i>quasiparaensis</i>	<i>Culicoides</i> <i>insignis</i>	<i>Bezzia</i> sp.	<i>Palpomyia</i> sp.
AUG/12	Dry	Open	10.33 ± 6.03	2 ± 2.65	2.33 ± 4.04	1 ± 1.73	-	-	-
SEP/12	Dry	Open	-	-	-	-	-	-	-
OCT/12	Dry	Open	-	-	0.33 ± 0.58	-	-	-	-
NOV/12	Dry	Open	-	-	-	-	-	-	-
DEC/12	Rainy	Open	-	-	-	-	-	-	-
JAN/13	Rainy	Open	0.33 ± 0.58	-	-	-	-	-	-
FEB/13	Rainy	Open	0.67 ± 1.15	-	0.33 ± 0.58	-	-	-	-
MAR/13	Rainy	Open	-	-	1.33 ± 2.31	-	0.33 ± 0.58	-	-
APR/13	Rainy	Open	-	-	-	-	-	-	-
MAY/13	Rainy	Open	4 ± 6.93	-	1 ± 1.73	-	-	-	-
JUN/13	Dry	Open	-	-	1 ± 1.73	-	-	-	-
JUL/13	Dry	Open	-	0.33 ± 0.58	-	-	-	-	-
AUG/13	Dry	Open	-	-	-	-	-	-	-
SEP/13	Dry	Open	-	-	-	-	-	-	-
OCT/13	Dry	Open	-	-	-	-	-	-	-
NOV/13	Dry	Open	-	-	-	-	-	-	-
DEC/13	Rainy	Open	-	-	-	-	-	-	-
JAN/14	Rainy	Open	-	-	0.33 ± 0.58	-	-	-	-
FEB/14	Rainy	Open	-	-	-	-	-	-	-
MAR/14	Rainy	Open g	-	0.67 ± 1.15	0.33 ± 0.58	-	-	-	-
APR/14	Rainy	Open g	-	-	-	-	-	-	-
MAY/14	Rainy	Open g	9.67 ± 16.74	-	-	-	-	-	-
JUN/14	Dry	Open	-	-	-	-	-	-	-
JUL/14	Dry	Open g	-	-	-	-	-	-	-
AUG/12	Dry	Closed	7.29 ± 18.41	-	0.71 ± 1.5	0.29 ± 0.76	-	-	-
SEP/12	Dry	Closed	-	-	-	-	-	-	-
OCT/12	Dry	Closed	6.14 ± 13.07	0.29 ± 0.76	0.43 ± 0.79	-	-	-	-



Month	Seasons	Area	<i>Forcipomyia</i> sp.	<i>Dasyhelea</i> <i>eloyi</i>	<i>Culicoides</i> <i>crucifer</i>	<i>Culicoides</i> <i>quasiparaensis</i>	<i>Culicoides</i> <i>insignis</i>	<i>Bezzia</i> sp.	<i>Palpomyia</i> sp.
NOV/12	Dry	Closed	0.57 ± 1.51	0.14 ± 0.38	1.29 ± 3.4	-	-	-	-
DEC/12	Rainy	Closed	-	-	0.29 ± 0.76	-	-	-	-
JAN/13	Rainy	Closed	-	7.29 ± 19.28	-	-	-	4.71 ± 12.03	7.14 ± 18.9
FEB/13	Rainy	Closed	-	7.57 ± 13.19	-	-	-	8.43 ± 18.27	1.86 ± 4.91
MAR/13	Rainy	Closed	-	1.71 ± 4.54	2.71 ± 6.75	-	-	0.71 ± 1.89	0.14 ± 0.38
APR/13	Rainy	Closed	-	1.86 ± 3.76	1.57 ± 3.04	-	-	0.29 ± 0.49	-
MAY/13	Rainy	Closed	-	1 ± 2.65	3.43 ± 9.07	-	-	0.14 ± 0.38	-
JUN/13	Dry	Closed	-	0.14 ± 0.38	0.86 ± 1.46	0.57 ± 1.51	-	2.57 ± 6.8	0.71 ± 1.89
JUL/13	Dry	Closed	-	-	0.57 ± 1.51	-	-	0.39 ± 0.76	-
AUG/13	Dry	Closed	-	-	-	-	-	-	-
SEP/13	Dry	Closed	-	-	3 ± 7.94	0.29 ± 0.76	-	-	-
OCT/13	Dry	Closed	-	0.14 ± 0.38	0.29 ± 0.49	-	-	-	-
NOV/13	Dry	Closed	-	0.14 ± 0.38	0.14 ± 0.38	-	-	-	-
DEC/13	Rainy	Closed	-	-	-	-	-	-	-
JAN/14	Rainy	Closed	-	0.86 ± 1.86	-	-	-	0.29 ± 0.76	-
FEB/14	Rainy	Closed	-	-	-	-	-	0.29 ± 0.76	0.14 ± 0.38
MAR/14	Rainy	Closed	-	0.29 ± 0.76	-	-	-	0.14 ± 0.38	0.14 ± 0.38
APR/14	Rainy	Closed	-	-	-	-	-	0.71 ± 1.89	-
MAY/14	Rainy	Closed	-	-	-	-	-	-	-
JUN/14	Dry	Closed	-	-	-	-	-	-	-
JUL/14	Dry	Closed	-	0.14 ± 0.38	-	-	-	-	-