www.biotaxa.org/rce. ISSN 0718-8994 (online) Revista Chilena de Entomología (2024) 50 (3): 459-471.

Research Article / Articulo de Investigación

Is the composition of the dipteran (Insecta) assemblage altered in mangrove environments by the presence of anthropogenic disturbances? A preliminary study on the coast of Rio Grande do Norte, Brazil

¿Se altera la composición del ensamble de dípteros (Insecta) en ambientes de manglar por la presencia de perturbaciones antropogénicas? Un estudio preliminar en la costa de Rio Grande do Norte, Brasil

Alessandra de Campos[1], Taciano Moura Barbosa[1], Ricardo José de Paula Souza e Guimarães[2], Jéssica Teixeira Jales[1], Tainá Costa Baia[1], Fernanda Vanessa Pires da Fonseca[1], Renata Antonaci Gama[1]

[1]*Universidade Federal do Rio Grande do Norte, Departamento de Microbiologia e Parasitologia, Laboratório de Insetos e Vetores-Live, Natal, Rio Grande do Norte, Brazil.* [2]*Instituto Evandro Chagas, Seção de Epidemiologia, Ananindeua, Pará, Brazil.* *renata.antonaci@ufrn.br**

ZooBank: urn:lsid:zoobank.org:pub:74B50EA7-2263-4F63-9775-9025190AE0D2
https://doi.org/10.35249/rche.50.3.24.06

Abstract. Sarcosaprophagous dipterans are well known due to their forensic and medical-sanitary importance, as they are possible vectors of pathogens and cause myiasis. However, their potential as bioindicators of anthropogenic actions is still little explored, especially in environments with high human interference (*e.g.*, coastal environments). Thus, this work aims to compare the abundance and frequency of dipteran of forensic importance between two mangrove landscapes with different human impact levels: i) Baía Formosa, characterized as less anthropized, and ii) Extremoz, as a more anthropized mangrove. Adult flies were collected by using suspended traps containing decomposing animal tissue (chicken liver) as baits. A diverse assemblage of sarcosaprophagous Diptera was registered in the two mangroves, consisting of 18 species from four families: Calliphoridae (n = 1,738; S = 4), Sarcophagidae (n= 214; S = 9), Muscidae (n = 46; S = 3) and Fanniidae (n = 26; S = 2). *Chrysomya megacephala* was the most abundant species in Baía Formosa and *Oxysarcodexia intona* in Extremoz. In addition, this study reveals that human impact levels can modulate the structure and composition of flies' assemblages in mangrove environments.

Key words: Calliphoridae; *Chrysomya*; human impact; Sarcophagidae.

Resumen. Los dípteros sarcosaprófagos son conocidos por su importancia forense y médico-sanitaria, ya que son posibles vectores de patógenos y pueden causar miasis. Sin embargo, su potencial como bioindicadores de acciones antropogénicas ha sido poco explorado, especialmente en entornos con alta interferencia humana (*e.g.*, entornos costeros). Por lo tanto, este trabajo tiene como objetivo comparar la abundancia y frecuencia de dípteros de importancia forense entre dos paisajes de manglares con diferentes niveles de impacto humano: i) Baía Formosa, caracterizada como menos antropizada y ii) Extremoz, como manglar más antropizado. Las moscas adultas fueron recolectadas utilizando trampas suspendidas que contenían tejido animal en descomposición (hígado de pollo) como cebo. Se registró una diversa comunidad de dípteros sarcosaprófagos en los dos manglares,

Received 28 May 2024 / Accepted 5 August 2024 / Published online 30 August 2024
Responsible Editor: José Mondaca E.

compuesta por 18 especies de cuatro familias: Calliphoridae (n = 1.738; S = 4), Sarcophagidae (n = 214; S = 9), Muscidae (n = 46; S = 3) y Fanniidae (n = 26; S = 2). *Chrysomya megacephala* fue la especie más abundante en Baía Formosa y *Oxysarcodexia intona* en Extremoz. Además, este estudio revela que los niveles de impacto humano pueden modular la estructura y composición de las comunidades de moscas en entornos de manglar.

Palabras clave: Calliphoridae; *Chrysomya*; impacto humano; Sarcophagidae.

Introduction

Forensic entomology has over a century of history in Brazil, presenting various applications in the field of criminal investigation (Pujol-Luz *et al*. 2008). However, one of the main challenges for the consolidation of environmental forensic entomology is to understand how insect distribution patterns can be used to assess or infer environmental crimes, such as those related to deforestation, negligence, or inadequate management of conservation units (Nakaza *et al*. 2009; Carmo and Vasconcelos 2016). Despite being a recent area, environmental forensic entomology has already produced relevant evidence on the potential of sarcosaprophagous dipterans as indicators of environmental conservation and anthropization (Carmo and Vasconcelos 2014; Maramat and Nor Aliza 2015; Carmo and Vasconcelos 2016; Barbosa *et al*. 2017; Dufek *et al*. 2020), with promising results for mangrove ecosystems (Azmi and Lim 2013; Maramat and Nor Aliza 2015), which have suffered significant natural losses due to anthropogenic activities (Diniz *et al*. 2019).

Mangroves are characterized by taxonomically diverse, salt-tolerant, and primarily woody vegetation, commonly found in the intertidal zone of tropical and subtropical coastlines (Polidoro *et al*. 2010). This vegetation is typically associated with a wide variety of fauna that spans various interconnected terrestrial and marine habitats, and they are linked to adjacent ecosystems through physical, biochemical, and biological interactions (Nagelkerken *et al*. 2008; Rog *et al*. 2016).

Mangroves play an important role in the sustainability of the planet, providing a wide range of environmental services such as nutrient cycling, soil formation, and wood production, in addition of being considered the most productive and complex ecosystems on Earth (Donato *et al*. 2011; Diniz *et al*. 2019). Despite their importance, mangrove vegetation is suffering from anthropogenic action, and significant losses of their vegetative mass has been identified over the years (Giri *et al*. 2011; Hamilton and Casey 2016; Diniz *et al*. 2019). In Brazil, more than 2% of mangrove area disappeared between 1998 and 2018 (Diniz *et al*. 2019), with a loss rate between 1999 and 2012 of approximately 0.41% per year (Hamilton and Casey 2016).

Among the inhabitants of this ecosystem, dipterans from the families Calliphoridae and Sarcophagidae stand out for their high forensic potential in cases of investigations against environmental crimes (Nakaza *et al*. 2009; Botteon *et al*. 2016), where they can be associated with the presence of human activity. This association, according to Linhares (1981), is known as synanthropy and can vary with species, geographical and climatic characteristics, as well as population concentrations. Some studies have highlighted the preference of calliphorids, especially for *Chrysomya* Robineau-Desvoidy, 1830, for anthropized environments (Linhares 1981; Montoya *et al*. 2009; Carmo and Vasconcelos 2016; Barbosa *et al*. 2017).

These individuals, especially *Chrysomya megacephala* (Fabricius, 1794) and *Chrysomya albiceps* (Widemann, 1819), were introduced in Brazil in the 1970s (Guimarães *et al*. 1978), and their invasive characteristic may be favored by urbanization processes, leading to alteration of the composition and structure of local necrophagous assemblages due to anthropic activities (Kavazos and Wallman 2012). Some studies have been conducted highlighting the association of species from the Sarcophagidae family with conserved or modified environments due to anthropic action (Yepes-Gaurisas *et al*. 2013; Barbosa *et al*. 2017; Dufek *et al*. 2020), but a strong relation between species of this family and clearings and conserved environments may characterize them as efficient bioindicators of forest conservation and regeneration (Sousa *et al*. 2014).

Therefore, considering the importance of mangrove vegetation, the need to assess the dipteran fauna present in these vegetations, as well as the impact of human activities in natural conservation environments, this study aimed to compare the composition and structure of sarcosaprophagous dipteran assemblages in two mangrove landscapes with different levels of human impact. We tested the hypothesis that human activity in mangrove environments acts as a biological modulator in the composition and structure of assemblages, with a higher number of synanthropic species in more anthropogenic environments.

Materials and Methods

Study area

The study was conducted in two mangrove landscapes located in the state of Rio Grande do Norte, Brazil (Fig. 1). To assess the influence of anthropogenic impact on the composition and structure of sarcosaprophagous Diptera assemblages, both mangrove landscapes were classified based on the levels of human impact according to Barbosa *et al*. (2017). This classification considered several characteristics, including the presence of 1) human-made landscape modifications such as roads and buildings, 2) pollution, with a focus on solid waste production and management, 3) adjacent economic activities (fishing), and constant circulation of people and vehicles (tourism).

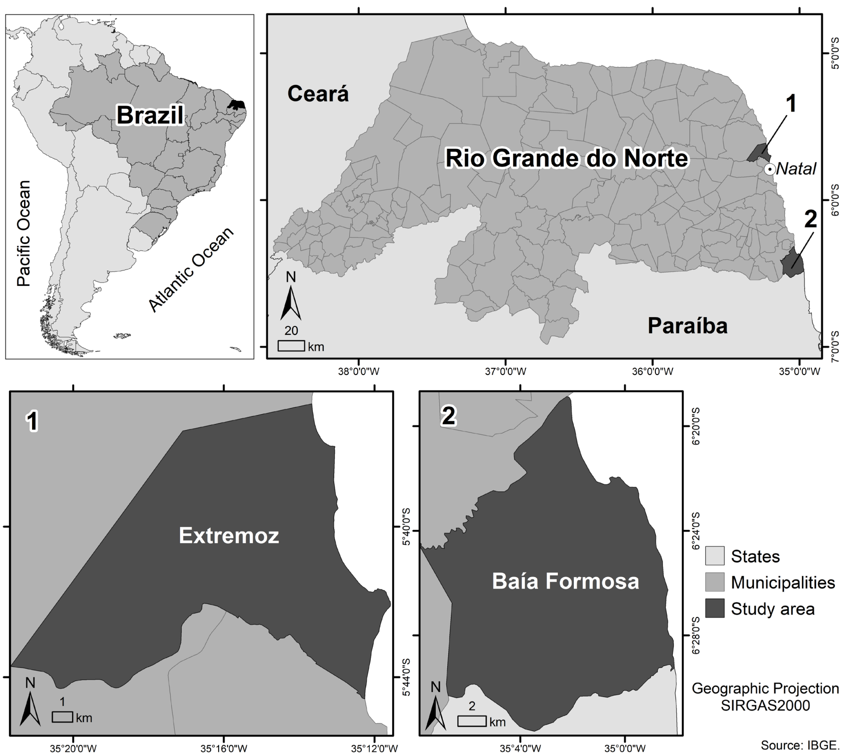

Figure 1. Localization of the municipalities of Baía Formosa and Extremoz in the state of Rio Grande do Norte, Brazil. / **Figura 1.** Localización de los municipios de Baía Formosa y Extremoz en el estado de Rio Grande do Norte, Brasil.

According to the methodology, we selected two mangrove vegetations located in different municipalities, but at the same state. Baía Formosa mangrove (Fig. 2) is located in the municipality of Baía Formosa, 90 km from Natal city (6°22′10″ S; 35°0′28″ W). The rainy season of the municipality comprises the months of January to August and its maximum and minimum temperatures are on average 30.0 °C and 21.0 °C, respectively. Its climate is considered rainy tropical, and the annual average relative humidity is 79% (Idema 2008). The mangrove vegetation is located next to a conservational unit, a particular reservation of the natural patrimony – PRNP – Mata Estrela, with a total area of 2,365 ha (Idema 2008). In the area, there are no roads or buildings nearby; the waste production and tourist attractiveness are controlled and managed to conserve the unit. Thus, the mangrove of Baía Formosa was characterized as having low anthropization (LA).

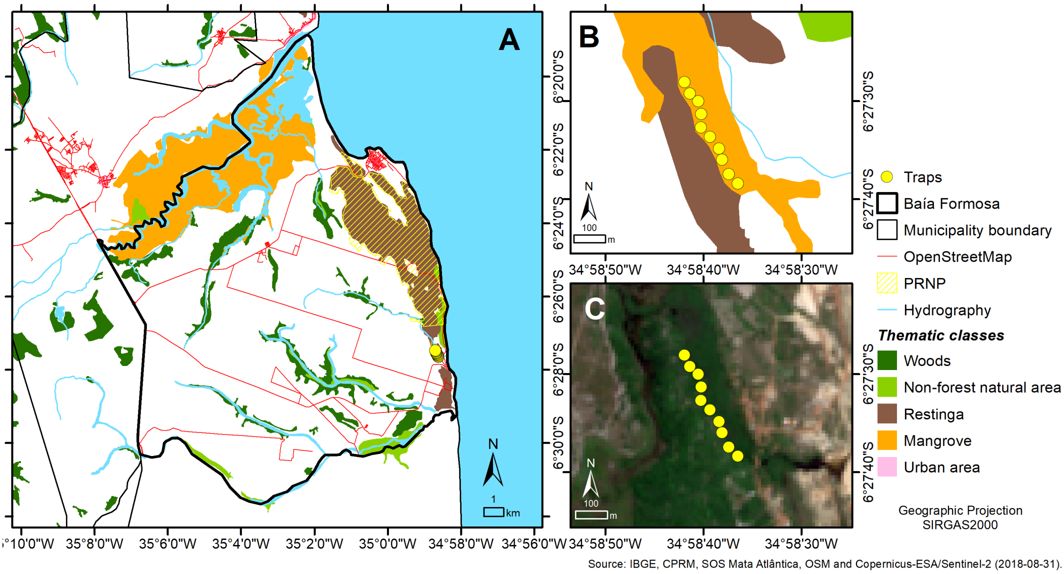

Figure 2. Images from Sentinel-2 and thematic classes from SOS Mata Atlântica of the mangrove landscape located in Baía Formosa, in the state of Rio Grande do Norte, Brazil. **A:** Overview thematic image of Baía Formosa; **B:** Detailed thematic image with collection points; **C:** Detailed satellite image of the area with collection points. / **Figura 2.** Imágenes de Sentinel-2 y clases temáticas de SOS Mata Atlântica del paisaje de manglares ubicado en Baía Formosa, estado de Rio Grande do Norte, Brasil. **A:** Imagen temática general de Baía Formosa; **B:** Imagen temática detallada con puntos de muestreo; **C:** Imagen satelital detallada del área con puntos de muestreo.

The second mangrove is located in the municipality of Extremoz (Fig. 3), 16 km from the capital (05°42′ 20″ S; 35°18′26″ W). Its rainy season begins in February and extends to September, with maximum and minimum temperatures averaging 30 °C and 21 °C, respectively. Its climate is also considered rainy tropical, and the average annual relative humidity is 77% (Idema 2013). Unlike the Baía Formosa mangrove, the Extremoz mangrove is located adjacent to the RN-304 highway and therefore it presents a high intensity of car flow and often waste disposal. The Extremoz mangrove has a significantly lower amount of vegetation coverage, even though it is part of the Jenipabu Environmental Protection Area (JEPA) (Idema 2013). Considering changes in vegetative cover, as well as human-driven exploitation of mangrove natural resources, Extremoz has been classified as a highly anthropized mangrove (HA).

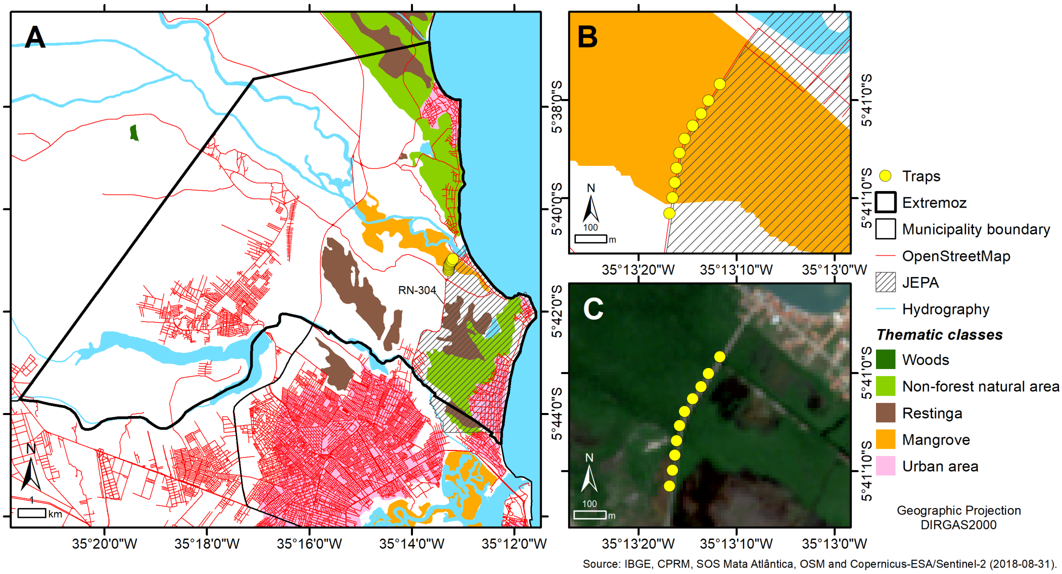

Figure 3. Images from Sentinel-2 and thematic classes from SOS Mata Atlântica of the mangrove landscape located in Extremoz in the state of Rio Grande do Norte, Brazil. **A:** Overview thematic image of Extremoz; **B:** Detailed thematic image with collection points; **C:** Detailed satellite image of the area with collection points. / **Figura 3.** Imágenes de Sentinel-2 y clases temáticas de SOS Mata Atlântica del paisaje de manglares ubicado en Extremoz, estado de Rio Grande do Norte, Brasil. **A:** Imagen temática general de Extremoz; **B:** Imagen temática detallada con puntos de muestreo; **C:** Imagen satelital detallada del área con los puntos de muestreo.

Collection and identification of specimens

Adult insects were collected using suspended traps adapted from Ferreira (1978), constructed from PET bottles and baited with approximately 300 grams of chicken meat previously decomposed for 24 hours. Ten traps were distributed in each environment, with a distance of 60 meters among each trap, and a height of 1.5 meters above the ground.

Insects were sampled in two field expeditions, totaling 20 independent sampling units per mangrove type. To maximize the representativeness, sampling was performed in two seasons: rainy and dry. Environmental data such as temperature, humidity, and precipitation were obtained at the meteorological station closest to the collection site. The collected insects were stored in a freezer for later identification, which was performed with stereomicroscope and the entomological keys of Carvalho and Mello-Patiu (2008) in the Laboratory of Insects and Vectors of the Federal University of Rio Grande do Norte (UFRN), Brazil. After identification, some specimens were duly pinned and later deposited in the Entomological Collection Adalberto Varela Freire (UFRN).

Geoprocessing data

The geographic coordinates of the traps were obtained using a handheld Global Positioning Systems receiver (Garmin, GPSMap 62s). The data used in this study were obtained from the Brazilian Institute of Geography and Statistics (IBGE - https://www.ibge.gov.br/) - boundaries; Geological Survey of Brazil (CPRM - http://www.cprm.gov.br/) - Hydrography; OpenStreetMap (OSM - https://www.openstreetmap.org) - streets and roads; Brazilian Ministry of the Environment (MMA) - PRNP and GEPA; SOS Mata

Atlântica (https://www.sosma.org.br/) - thematic classes: woods, non-forest natural area, mangrove, restinga (sandy coastal plain vegetation) and urban area. The Sentinel-2 images by European Space Agency (ESA - https://sentinel.esa.int/web/sentinel/home) were obtained using Google Earth Engine script (https://code.earthengine.google.com /9104949f5ea80bbf3d437cb6ee5c99a9). A Geographical Information System (GIS) was used to visualize the obtained coordinates into a base map.

Data analysis

Data normality verification was performed by using the statistical test Kolmogorov-Smirnov, with a 5% probability. Even after the transformation of the data into Log 10, they were not normal and therefore were submitted to the Mann-Whitney non-parametric statistical test to evaluate if there was difference in the abundance of flies between the two mangrove landscapes and between the seasons: rainy and dry. A multiple linear regression was performed to evaluate the interference of abiotic variables (maximum and minimum temperature, humidity and precipitation) in the abundance of dipterans collected at both collection sites.

Subsequently, it was performed a non-metric multidimensional scaling (nMDS) using the numerical abundance matrix of fly species. According to Gotelli and Ellison (2011), the aim of this statistical technique is to generate a graph in which similar points are positioned close while the distinct ones are positioned far from the ordering space. Posteriorly, the log transformation (x + 1) was applied, and the Bray-Curtis similarity measure was used to compare the similarity between the two levels of human action (low and high).

Besides, the ANOSIM similarity test was conducted to ensure the reliability of nMDS results. The ecological analyzes were carried out in order to evaluate the diversity of the families found. Relative frequency and dominance were determined for each species, with species classified as dominant (D) and non-dominant (nD), according to the frequency values found (Silveira Neto *et al.* 1976). Shannon and Pielou indexes were calculated to obtain diversity and equitability, respectively. Kolmogorov-Smirnov normality test, Mann-Whitney, and regression were performed in the BioEstat® 5.3 statistical program. For the nMDS and the diversity indexes, the Primer® 6.0 program was used, always adopting the significance level of 5%.

Results

When all samples were combined, a total of 2,651 specimens were captured (Tab. 1) belonging to the families Calliphoridae, Fanniidae, Muscidae and Sarcophagidae. Calliphoridae was the most representative family, comprising 67.7% of all collected adults, followed by Sarcophagidae (29.0%), Muscidae (2.0%) and Fanniidae (1.3%). However, when considering richness, Sarcophagidae was the most diverse family, with 10 spp., followed by Calliphoridae (4 spp.), Muscidae (3 spp.), and Fanniidae (2 spp.). *Chrysomya megacephala* (Fabricius, 1794) was the most abundant species, with 60.4% of the collected specimens, followed by *Chrysomya albiceps* (Wiedemann, 1819) (7.0%), *Oxysarcodexia intona* (Curran & Walley, 1934) (3.1%) and *Atherigona orientalis* (Schiner, 1868) (1.4%).

Table 1. Abundance of families and species found in the two mangrove landscapes of Rio Grande do Norte, Brazil. / **Tabla 1.** Abundancia de familias y especies encontradas en los dos paisajes de manglar de Rio Grande do Norte, Brasil.

Family/Species	**N**
Calliphoridae	**1,795**
Chrysomya megacephala (Fabricius, 1794)	1,602
Chrysomya albiceps (Wiedemann, 1819)	185
Chrysomya putoria (Wiedemann, 1818)	7
Lulicia eximia (Wiedemann, 1819)	1
Fanniidae	**33**
Fannia pusio (Wiedemann, 1830)	5
Fannia sp.	28
Muscidae	**54**
Hydrotaea aenescens (Wiedemann, 1830)	11
Atherigona orientalis (Schiner, 1868)	36
Musca domestica (Linnaeus, 1758)	7
Sarcophagidae	**769**
Oxysarcodexia intona (Curran & Walley, 1934)	81
Oxysarcodexia timida (Aldrich, 1916)	25
Oxysarcodexia amorosa (Schiner, 1868)	9
Peckia (*Peckia*) *chrysostoma* (Wiedemann, 1830)	11
Peckia (*Peckia*) *pexata* (Wulp, 1895)	3
Peckia (*Peckia*) *vellegasi* (Dodge, 1966)	3
Peckia (*Sarcodexia*) *lambens* (Wiedemann, 1830)	6
Tricharaea (*Sarcophagula*) *occidua* (Fabricius, 1794)	12
Villegasia pernambucana (Tibana & Lopes, 1985)	2
Sarcofartiopsis cuneata (Townsend, 1935)	2
Sarcophagidae females	615
Total	**2,651**

Considering the level of human impact, LA areas presented 16 species, and 74.8% (n = 2,054) of all recorded specimens, while in HA areas, 15 species were recorded, representing 25.2% (n = 690) of the captured adults. The abundance differed between the landscapes, and LA had a greater number of collected specimens (Mann-Whitney $P = 0.005$). When families were considered, Calliphoridae was the most representative family in LA mangrove, while Sarcophagidae was the most representative family in HA mangrove (Tab. 2). *Chrysomya albiceps* and *C. megacephala* were the only dominant species in LA mangroves, while in HA mangroves there were four dominant species, *C. albiceps*, *C. megacephala*, *O. intona* and *O. timida* (Tab. 2).

For richness, there was no variation in the number of species recorded in both mangroves ($P > 0.05$), with 16 species and 15 species registered for LA and HA, respectively. However, the assembly compositions were divergent. Regarding the species, *C. megacephala* was the most representative species in LA mangrove, comprising 84.1% of the individuals collected in this area, followed by *C. albiceps* (9.2%), *Atherigona orientalis* (Schiner, 1868) (1.6%) and *Oxysarcodexia intona* (Curran & Walley, 1934) (1%). Besides, *Chrysomya putoria* (Wiedemann, 1818), *Lucilia eximia* (Wiedemann, 1819) and *Peckia* (*Sarcodexia*) *lambens* (Wiedemann, 1830) were unique in the LA mangrove (Tab. 2).

Table 2. Abundance, relative frequency and dominance of the species found in the mangrove with high and low anthropization (HA and LA), respectively. D = dominant; nD = non-dominant. / **Tabla 2.** Abundancia, frecuencia relativa y dominancia de las especies encontradas en el manglar con alta y baja antropización, HA y LA, respectivamente. D = dominante; nD = no dominante.

	Baía Formosa mangrove (LA)			**Extremoz mangrove (HA)**		
Family/Species	**Nº**	**%**	**D**	**Nº**	**%**	**D**
Calliphoridae	**1,739**			**56**		
Chrysomya megacephala (Fabricius, 1794)	1,559	84.18	D	43	27.56	D
Chrysomya albiceps (Wiedemann, 1819)	172	9.29	D	13	8.33	D
Chrysomya putoria (Wiedemann, 1818)	7	0.38	Nd	0	0	
Lulicia eximia (Wiedemann, 1819)	1	0.05	Nd	0	0	
Fanniidae	**3**			**2**		
Fannia pusio (Wiedemann, 1830)	3	0.16	Nd	2	1.28	Nd
Muscidae	**46**			**8**		
Hydrotaea aenescens (Wiedemann, 1830)	10	0.54	Nd	1	0.64	Nd
Atherigona orientalis (Schiner, 1868)	30	1.62	Nd	6	3.85	Nd
Musca domestica (Linnaeus, 1758)	6	0.32	Nd	1	0.64	Nd
Sarcophagidae	**64**			**90**		
Oxysarcodexia intona (Curran & Walley, 1934)	20	1.08	Nd	61	39.10	D
Oxysarcodexia timida (Aldrich, 1916)	11	0.59	Nd	14	8.97	D
Oxysarcodexia amorosa (Schiner, 1868)	7	0.38	Nd	2	1.28	Nd
Peckia (*Peckia*) *chrysostoma* (Wiedemann, 1830)	6	0.32	Nd	5	3.20	Nd
Peckia (*Peckia*) *pexata* (Wulp, 1895)	2	0.11	Nd	1	0.64	Nd
Peckia (*Peckia*) *vellegasi* (Dodge, 1966)	0	0		3	1.92	Nd
Peckia (*Sarcodexia*) *lambens* (Wiedemann, 1830)	6	0.32	Nd	0	0	
Tricharaea (*Sarcophagula*) *occidua* (Fabricius, 1794)	11	0.59	Nd	1	0.64	Nd
Villegasia pernambucana (Tibana & Lopes, 1985)	1	0.05	Nd	1	0.64	Nd
Sarcofartiopsis cuneata (Townsend, 1935)	0	0		2	1.28	Nd
Total	**1,852**	**100**		**156**	**100**	

On the other hand, the most representative species in HA mangrove was *Oxysarcodexia intona* (Curran & Walley, 1934), comprising 39.1% of all collected specimens in this vegetation, followed by *C. megacephala* (27.5%), *Oxysarcodexia timida* (Aldrich, 1916) (8.9%) and *C. albiceps* (8.3%). In addition, *Peckia* (*Peckia*) *vellegasi* (Dodge, 1966) and *Sarcofartiopsis cuneata* (Townsend, 1935) were found exclusively in HA mangrove. These data indicate that the less anthropized mangrove presented a higher abundance of specimens and dominance of species from the Calliphoridae family, while in the highly anthropized mangrove, the Sarcophagidae family dominated the assemblage. These entomological differences led to a more in-depth analysis of the area, resulting in the discovery of an illegal landfill located 4.5 km from the LA mangrove.

No significant difference was observed between the rainy season and dry season in Baía Formosa (*P* = 0.0653). However, there was a strong trend towards a differentiation, with the rainy season presenting more abundant assemblages in relation to the dry season. In the HA mangrove, the results were not significant (*P* = 0.4251). Multiple linear regression did not show interference of abiotic variables (maximum and minimum humidity and precipitation) in the abundance of dipterans collected at both collection sites (*P* = 0.7859, LA and *P* = 0.9999, in HA).

The analysis of nMDS from the numerical abundance data of the fly species evidenced a pattern of dipterofauna separation according to the degree of human impact (Fig. 4). The difference between the environments in relation to the level of human interference was still confirmed by ANOSIM (r = 0.64, P = 0.01). The values of the Equitability Indices J' were 0.2553 and 0.6598 and Shannon H' diversity indices were 0.7078 and 1.787 for the mangrove with LA (Baía Formosa) and with HA (Extremoz), respectively.

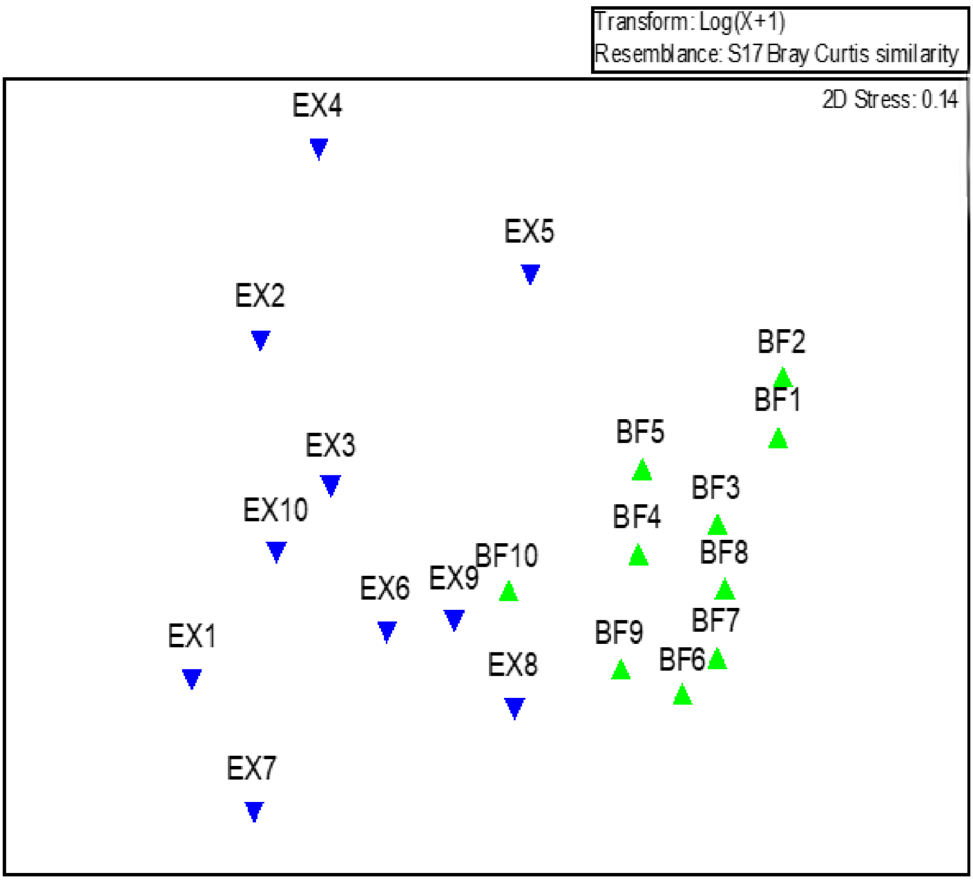

Figure 4. NMDS from both studied landscapes using the Bray-Curtis distance according to the human impact level with Stress value of 0.14. BF = Baía Formosa; EX = Extremoz. / **Figura 4.** NMDS de ambas paisajes estudiadas utilizando la distancia de Bray-Curtis según el nivel de impacto humano con un valor de estrés de 0,14. BF = Baía Formosa; EX = Extremoz.

Discussion

Mangrove ecosystems play a crucial role in human sustainability by providing a wide range of ecosystem services (Diniz *et al*. 2019). However, they have been consistently impacted by human activities. In Brazil, over 2% of mangrove landscapes have disappeared in the last two decades (Diniz *et al*. 2019), and sarcosaprophagous dipterans can be a useful tool in assessing the environmental impact of human activities on natural environments (Nakaza *et al*. 2009; Carmo and Vasconcelos 2016; Botteon 2016; Barbosa *et al*. 2017), including trophic turnover, such as in mangroves. This study demonstrates the impact of anthropization on the composition and structure of sarcosaprophagous dipteran assemblages where areas with a high level of anthropization are negatively affected. This fact is evidenced by the presence of more synanthropic species and the absence of native taxa (*e.g*., *L. eximia*) in HA mangroves.

Our results depict a direct application of environmental forensic entomology. Due to the contradiction in the entomological data found in our study and considering that *C. megacephala* and *C. albiceps* are exotic species and have been recorded as dominant in mangroves with high levels of anthropization and landfills (Gonçalves *et al*. 2010; Seolin *et al*. 2009; Fraga and D'Almeida 2005), the conservation status of the area was questioned. New visits were made to the mangrove landscapes, and with the assistance of residents and city officers, the surroundings of the mangrove were explored. After an active search, we found an illegal dumping ground located 4.5 km from the mangrove. Thus, the data reveal that sarcosaprophagous dipterans of Calliphoridae family, in addition to their sanitary, medical, and forensic relevance (Amendt 2004), are important as bioindicators of anthropogenic disturbances.

In mangrove ecosystems, the Calliphoridae and Sarcophagidae families are commonly found as representatives of the decomposing fauna (Gonçalves *et al*. 2010; Azmi *et al*. 2013; Maramat and Nor Aliza 2015), and in this study, these families dominated the visiting assemblages in both mangrove areas, representing approximately 93% of all specimens collected. However, the LA mangrove presented a higher number of species and numerical abundance when compared to the HA mangrove, confirming our initial hypothesis. Additionally, we expanded the distribution of *Peckia* (*Peckia*) *vellegasi* (Dodge, 1966) in Brazilian territory, as this species was recently recorded in Brazil, with occurrences on the coast of Pernambuco (Barbosa *et al*. 2015).

Dias *et al*. (2009) experimentally demonstrated that the Calliphoridae family exhibits synanthropic behavior and can thus be found in environments with varying degrees of anthropogenic impact and nutrient availability (*e.g*., landfills). This may help explaining the high abundance of *Chrysomya* species in the study, including in LA areas, since there was an illegal dumping ground in the vicinity of this environment. On the other hand, species of Sarcophagidae family have been associated with environments exposed to human activity (*e.g*., *Peckia* and *Oxysarcodexia* species) (Yepes-Gaurisas *et al*. 2013), which corroborates our data, as *O. intona* was the dominant species in the HA area. Barbosa *et al.* (2017) also showed that the level of anthropization significantly altered the composition of assemblages in coastal areas in the state of Pernambuco, Brazil, where the higher the level of anthropization, the greater the dominance of Calliphoridae.

Besides C. *megacephala*, *C. albiceps*, *A. orientalis* and *O. intona* stood out in our study, they were recorded in both mangrove ecosystems. These species have been recorded in coastal environments with different environmental impacts (Barbosa *et al*. 2017; Carmo and Vasconcelos 2016) and are closely associated with the decomposition of organic matter, regardless of the level of anthropization. Some species were recorded only in mangroves under LA, such as *C. putoria*, *L. eximia*, and *P.* (*S.*) *lambens*, while *P.* (*P.*) *vellegasi* and *S. cuneata* were exclusive to the HA mangrove.

These results show that, despite being mangrove vegetation and located in the same state, the assembly composition varies according to the presence of human impacts, thereby strengthening the theoretical framework for environmental forensic entomology. It is important to highlight that we attributed all changes in the composition and structure of sarcosaprophagous dipteran assemblages to the level of human impact in the landscapes, as in this study, the abiotic variables did not influence dipteran abundance.

Conclusion

This study presents, for the first time, a direct application of environmental forensic entomology in Brazil, where flies were used as a key tool to identify environmental impact in a mangrove area in the state of Rio Grande do Norte. The mangroves located in Baía Formosa and Extremoz are suffering human impact, and the authorities must make efforts to identify those responsible for the impact. Furthermore, being located near or within a conservation unit, the mangroves must be efficiently protected to minimize negative impacts and preserve native species.

Acknowledgment

We thank the Chico Mendes Institute for Biodiversity Conservation (ICMBio) for permission and license to collect insects.

Author Contributions

AC, RAG: Conceived and designed the research. **AC, TCB, FVPF, TMB, RAG:** Conducted fieldwork and identification. **AC, TMB, RJPSG, RAG:** Analyzed the data. **RJPSG:** Created the maps. **AC, JTJ, TMB, RAG:** Wrote the manuscript. All authors read, contributed to, and approved the final version of the manuscript.

Literature Cited

Amendt, J., Krettek, R. and Zehner, R. (2004) Forensic Entomology. *Naturwissenschaften*, *91*: 51-65.

Azmi, W.A. and Lim, S.P. (2013) Comparative study of dipteran species diversity and their succession on rabbit carrion in two different mangrove areas of Peninsular Malaysia. *Journal of Insects, 2013*(398159): 1-9. http://dx.doi.org/10.1155/2013/398159

Barbosa, T.M., Mello-Patiu, C.A. and Vasconcelos, S.D. (2015) Flesh fly (Diptera: Sarcophagidae) survey on coastal environments in northeastern Brazil: New records and notes on the expanded geographical distribution. *Entomotropica*, *30*(12): 112-117.

Barbosa, T.M., Carmo, R.F.R., Silva, L.P., Sales, R.G. and Vasconcelos, S.D. (2017) Diversity of sarcosaprophagous calyptratae (Diptera) on sandy beaches exposed to increasing levels of urbanization in Brazil. *Environmental Entomology*, *46*: 460-469.

Botteon, V.W. (2016) Perspectivas de uso de insetos bioindicadores ambientais em trabalhos periciais. *Brazilian Journal of Forensic Sciences, Medical Law and Bioethics, 5*(4): 383-401.

Carmo, R.F.R. and Vasconcelos, S.D. (2014) First record of the blow fly *Chrysomya megacephala* (Diptera: Calliphoridae) on a southern Atlantic island: implications for disease transmission in a protected environment. *Journal of Vector Ecology*, *39*: 228-230.

Carmo, R.F.R. and Vasconcelos, S.D. (2016) Assemblage of necrophagous Diptera in Atlantic insular environments and response to different levels of human presence. *Neotropical Entomology*, *45*: 471-481.

Carvalho, C.J.B. and Mello-Patiu, C.A. (2008) Key to the adults of the most common forensic species of Diptera in South America. *Revista Brasileira de Entomologia*, *52*: 390-406.

Dias, L.S., Santarem, V.A., Almeida, M.S.R., Medina, A.O. and Silva, A.V. (2009) Biodiversidade de moscas Calliphoridae no lixão urbano de Presidente Prudente, São Paulo, Brasil. *Arquivos do Instituto de Biologia*, *76*(4): 659-663.

Diniz, C., Cortinhas, L., Nerino, G., Rodrigues, J., Sadeck, L., Adami, M. and Souza-Filho, P.W.M. (2019) Brazilian mangrove status: Three decades of satellite data analysis. *Remote Sensing*, *11*(7): 808.

Donato, D.C., Kauffman, J.B., Murdiyarso, D., Kurnianto, S., Stidham, M. and Kanninen, M. (2011) Mangroves among the most carbon-rich forests in the tropics. *Nature Geoscience*, *4*: 293-297.

Dufek, M.I., Larrea, D.D., Damborsky, M.P. and Mulieri, P.R. (2020) The effect of anthropization on Sarcophagidae (Diptera: Calyptratae) community structure: An assessment on different types of habitats in the humid Chaco Ecoregion of Argentina. *Journal of Medical Entomology*, *5*(1): 1468-1479.

Ferreira, M.J.M. (1978) Sinantropia de dípteros muscoides de Curitiba, Paraná. I: Calliphoridae. *Revista Brasileira de Biologia*, *38*(2): 445-454.

Fraga, M.B. and D'Almeida, J.M. (2005) Observações preliminares sobre a atratividade por diferentes cores em Calliphoridae (Diptera), Niterói, RJ, Brasil. *Entomología y Vectores*, *12*(1): 141-147.

Giri, C., Ochieng, E., Tieszen, L.L., Zhu, Z., Singh, A., Loveland, T., Masek, J. and Duke, N. (2011) Status and distribution of mangrove forests of the world using earth observation satellite data. *Global Ecology and Biogeography*, *20*: 154-159.

Gonçalves, L., Dias, A., Espindola, C.B. and Almeida, F.S. (2010) Inventário de Calliphoridae (Diptera) em manguezal e fragmento de Mata Atlântica na região de Barra de Guaratiba, Rio de Janeiro, Brasil. *Revista Brasileira de Biociência*, *9*(1): 50-55.

Gotelli, N.J. and Ellison, A.M. (2011) Princípios de estatística em ecologia. Artmed, Porto Alegre, 1ª ed. 528 pp.

Guimarães, J.H., Prado, A.P. and Linhares, A.X. (1978) Tree newly introduced blowfly species in southern Brazil (Diptera, Calliphoridae). *Revista Brasileira de Entomologia*, *22*: 53-60.

Hamilton, S.E. and Casey, D. (2016) Creation of a high spatio-temporal resolution global database of continuous mangrove forest cover for the 21st century (CGMFC-21). *Global Ecology and Biogeography*, *25*: 729-738.

IDEMA [Instituto de Desenvolvimento Sustentável e Meio Ambiente do Rio Grande do Norte] (2008) Perfil do Seu Município Baía Formosa V.10: 1-21. Available at: http://www.idema.rn.gov.br. (Accessed at June 04, 2020).

IDEMA [Instituto de Desenvolvimento Sustentável e Meio Ambiente do Rio Grande do Norte] (2013) Perfil do Seu Município Extremoz. Available at: http//www.idema.rn.gov.br. (Accessed at June 04, 2020).

Kavazos, C.R.J. and Wallman, J.F. (2012) Community composition of carrion-breeding blowflies (Diptera: Calliphoridae) along an urban gradient in south-eastern Australia. *Landscape and Urban Planning*, *106*: 183-190.

Linhares, A.X. (1981) Synanthropy of Calliphoridae and Sarcophagidae (Diptera) in the city of Campinas, São Paulo, Brazil. *Revista Brasileira de Entomologia*, *25*: 189-215.

Maramat, R. and Nor Aliza, A.R. (2015) Forensically important flies associated with decomposing rabbit carcasses in mangrove forests in Kuching, Sarawak, Malaysia. *Malaysian Journal of Forensic Sciences*, *6*(1): 79-83.

Montoya, A.L.G., Sánchez, J.D.R. and Wolff, M.E. (2009) Sinantropía de Calliphoridae (Diptera) del municipio La Pintada, Antioquia – Colombia. *Revista Colombiana Entomología*, *35*: 73-82.

Nagelkerken, I., Blamar, S.J.M., Bouillon, S., Green, P., Haywood, M.D.E., Kirton, L.G., Meynecke, J-O., Pawlik, J.R., Penrose, H., Sasekumar, A. and Somerfield, P. (2008) The habitat function of mangroves for terrestrial and marine fauna: A review. *Aquatic Botany*, *89*: 155-185.

Nakaza, E., Dias Filhos, C.R., Fontes, L.R. and Silva, U.F.S.M. (2009) Aplicação da entomologia forense nos crimes contra o meio ambiente: Perspectivas para o Brasil. *Arquivos da Policia Civil, 51*: 11-24.

Polidoro, B.A., Carpenter, K.E., Collins, L., Duke, N.C., Ellison, A.M., Ellison, J.C. Farnsworth, E.J., Fernando, E.S., Kathiresan, K., Koedam, N.E., Livingstone, S.R., Miyagi, T., Moore, G.E., Ngoc Nam, V., Ong, J.E., Primavera, J.H., Salmo, S.G., Sanciangco, J.C., Sukardjo, S., Wang, Y. and Yong, J.W. (2010) The loss of species: Mangrove extinction risk and geographic areas of global concern. *PLoS ONE*, *5*(4): e10095. https://doi.org/10.1371/journal.pone.0010095

Pujol-Luz, J.R., Arantes, L.C. and Constantino, R. (2008) Cem anos da entomologia forense no Brasil (1908-2008). *Revista Brasileira de Entomologia*, *52*(4): 485-492.

Rog, S.M., Clarke, R.H. and Cook, C.N. (2016) More than marine: Revealing the critical importance of mangrove ecosystems for terrestrial vertebrates. *Diversity and Distributions*, *23*: 221-230.

Seolin Dias, L., Santarém, V.A., Almeida, M.S.R., Medina, A.O. and Silva, A.V. (2009) Biodiversidade de moscas Calliphoridae no lixão urbano de Presidente Prudente, São Paulo, Brasil. *Arquivos do Instituto de Biologia, 76*(4): 659-663.

Silveira Neto, S., Nakano, O., Barbin, D. and Villa Nova, N.A. (1976) Manual de ecologia dos insetos. Piracicaba: Ceres, 419 pp.

Sousa, J.R., Esposito, M.C., Filho, F.S.C. and Juen, L. (2014) The potential use of sarcosaprophagous flesh flies and blowflies for the evaluation of the regeneration and conservation of forest clearings: a case study in the Amazon forest. *Journal of Insect Science, 14*: 1-5.

Yepes-Gaurisas, D., Sánchez-Rodríguez, J.D., Mello-Patiu, C.A. and Wolff, M.E. (2013) Synanthropy of Sarcophagidae (Diptera) in La Pintada, Antioquia – Colombia. *Revista de Biología Tropical*, *61*: 1275-1287.