

Richness and composition of Calliphoridae in an Atlantic Forest fragment: implication for the use of dipteran species as bioindicators

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Received: 16 April 2013 / Accepted: 7 August 2013 / Published online: 11 August 2013
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Abstract Studies measuring the damage in degraded environments have increased, and it is necessary to obtain reliable biological and ecological indicators for the recovery of such degraded environments and subsequent preservation. This study aimed to conduct a survey of local insects and evaluate their utility in monitoring forest restoration in a degraded area in the Atlantic Forest biome in Ribeirão Grande, State of São Paulo, Brazil. Collections with Shuey traps were conducted from 2006 to 2011 in four distinct forest fragments characterized by environmental impacts, human actions, and phytophysiognomic profiles: Forest (FO), a preserved area with native plant species; “Capoeira” (CA), an area in the natural regeneration process; Planting (PL), an area reforested with native plants; and Pasture (PA), an area with only shrubs. A total of 2,456 specimens of Calliphoridae (Diptera) were collected. *Mesembrinella bellardiana* was the most abundant ($n = 1,884$) and dominant species in all environments sampled. The relative abundance of *M. bellardiana* in the most preserved environment (60.2 % FO, 24.4 % CA, 8.6 % PL, and 6.7 % PA) and other ecological parameters showed that it could be a bioindicator species, i.e., data on its presence or absence directly reflected the status of local preservation. Information on Neotropical dipterans associated with the forested environment is very scarce in the literature. To our knowledge, this is the first study describing the occurrence of *Calliphora lopesi* (Calliphoridae) in the State of São Paulo.

Keywords Blowflies · Diversity · Rainforest · South America

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Introduction

The Atlantic Rainforest biome, one of the most important in the world, extends over a large part of Brazil and, in a lesser extent, into Paraguay and Argentina. It is a mosaic of diverse ecosystems, with structures and floristic compositions varying according to soil type, topography, and climatic characteristics (IBAMA 2012). It is one of the most threatened tropical forests in the world, especially by human actions associated with agricultural activities and rearing of livestock (Dean 2004). It is highly fragmented and only 7.3 % of the original forest cover remains (IBAMA 2012).

Ecological disequilibrium caused by fragmentation can substantially influence diversity. In environments in competitive balance, the total diversity of species is normally low because certain species that coexist avoid direct competition for limited resources (Huston 1995). As a result, measuring the effect of fragmentation in some communities, particularly in invertebrate communities, is not a simple task (Offerman et al. 1995; Didham 1997).

Many invertebrate taxa, including insects, are biological and ecological indicators of degradation because of their low resistance to environmental imbalances (Margalef 1951; Kremen et al. 1993; Brown and Hutchings 1997). They are excellent for qualitative measures in short-term because they have high reproductive capacity, which ensures rapid reestablishment of original populations (Brown 1997; Holloway et al. 1987), while vertebrates can take decades to restructure population equilibrium (Brown 1997).

Diptera may be used to quantify the degree of human interference on natural environments (Gadelha et al. 2009; Bizzo et al. 2010; Mata et al. 2010) because of their rapid populational response capacity. Some insects are well adapted to live in inhospitable and highly modified environments, which is often evidenced by their proximity to humans and their domestic animals (i.e., synanthropic classification) (Greenberg 1973; Linhares 1981). However, other insects are associated only with undisturbed environments (D'Almeida and Lopes 1983; Mariluis et al. 1990; Doge et al. 2008; Ferraz et al. 2010; Sousa et al. 2011a, b).

Analyses of species richness, faunistic composition, and other ecological parameters associated with different levels of impact on the environment are used to indicate if a given group of organisms could be useful for monitoring local recovery. Thus, this study aimed to survey local insects and to evaluate the utility of this information in monitoring forest restoration in an area of the Atlantic Forest biome degraded by mineral extraction and cement manufacturing.

Materials and methods

Area of study

The site, Intermontes Farm, comprised 343 ha and is located near the State Park Intervales in the county of Ribeirão Grande, State of São Paulo, Brazil. It is triangulated among the coordinates 24°0'5.74''S:48°20'21.81''W, 24°0'16.43''S:48°20'23.96''W, and 24°0'10.34''S:48°20'29.07''W. This area is included in the Geomorphological Atlantic Plateau Province, Plateau Guapiara zone, which comprises the elevated region of Paranapiacaba Serra and extends up to the sedimentary cover of the Paraná basin, which includes the effluents of the upper course of the left margin of Paranapanema (Nave 2005).

The collections were conducted in a 215.4 ha area. Four distinct fragments were chosen based on environmental impact, human action, and phytophysiognomic profiles (Nave 2005): Forest (FO), with 74.49 ha, is a well-preserved area with native plant species whose

heights range from 12 to 20 m; “Capoeira” (CA), with 51.09 ha, is an area in the process of natural restoration and has predominantly shorter (2–6 m high) forest formations and few tree species. It is generally in the early stages of succession, and has low infestation rates of *Brachiaria decumbens*; Planting (PL), with 55.43 ha, was reforested with native plants planted from 2001 to 2004; and Pasture (PA), with 34.39 ha, has only shrubs, mainly *B. decumbens*.

Sampling

Two Shuey traps (Shuey 1997) using fermented sugarcane juice and banana as bait, were placed approximately 50 m apart from each other for 24 h once a month in each fragment in January to April, June, August, November, and December from 2006 to 2011. Insects were removed, killed by transferring to a killing jar, and transferred to appropriately labeled containers. Insects were mounted and identified using dichotomous keys (Guimarães 1977; Carvalho and Ribeiro 2000; Bonatto 2001; Mello 2003) in the Laboratory of Entomology of the Department of Animal Biology, IB, UNICAMP.

Monthly of temperature and precipitation means for the study period were obtained from the meteorological database of the National Institute of Meteorology (INMET) (BDMEP 2012).

Data analysis

Abundance, dominance, diversity, equitability, and similarity of species were compared. The comparison among the abundance of fragments was performed using the *t* test and values were considered significant at $P < 0.05$. Analyses of correlation between fragment type and species abundance, and between diversity and climatic conditions were performed using the correlation coefficient nonparametric Spearman.

The Berger–Parker index (*d*), which expresses the proportional importance of the most abundant species in a given area or sample (Magurran 1988), was used to measure the degree of dominance among the studied areas. The value of this index is obtained by expression $d = N_{\max}/N$, where “*d*” is the degree of dominance, N_{\max} is the number of individuals of the most abundant species, and *N* is the total number of individuals sampled in the area. The Shannon-Wiener index (*H'*) was used to calculate the species diversity. The Shannon’s equitability index (*J'*) was calculated according to the formula: $J' = H'/H_{\max}$, where *H'* is the Shannon-Wiener’s index and H_{\max} is the logarithm of the total number of species in the sample. Equitability assumes a value between 0 and 1, with 1 being complete evenness. The similarity analysis, calculated using the Jaccard’s coefficient $I_j = c/a + b - c$, allows evaluation of significant differences in species compositions among different environments. The values range between -1 and 1 , with values close to 1 being larger differences (Moreno 2001).

Statistical analyzes were performed using BioestatTM software (Ayres et al. 2003) and all analyzes of ecological parameters were done with the programs PastTM (Hammer et al. 2001) and DivesTM (Rodrigues 2007).

Results

A total of 2,456 specimens of Calliphoridae (Diptera), distributed in 11 genera and 13 species (Table 1), were collected from 2006 to 2011. The highest abundance was found in

Table 1 Abundance (n); relative abundance (p); dominance (d); total diversity (H'), equitability (J'), and similarity (I_j) of species (Diptera: Calliphoridae) collected from different fragments in Interfontes Farm

Species	Fragments												Total (n)	Total (p)	t test
	Forest			"Capoeira"			Planting			Pasture					
	n	p	d	n	p	d	n	p	d	n	p	d			
<i>Mesembrinella bellardiana</i>	1,140	76.9	0.77	521	86.8	0.87	140	65.7	0.66	83	50.0	0.50	1,884	76.6	*
<i>Mesembrinella peregrina</i>	275	18.6	0.19	48	8.0	0.08	24	11.3	0.11	26	15.7	0.16	373	15.2	*
<i>Calliphora lopezi</i>	16	1.1	0.01	0	0.0	0	10	4.7	0.05	12	7.2	0.07	38	1.5	N/A
<i>Lucilia eximia</i>	13	0.9	0.01	12	2.0	0.02	4	1.9	0.02	4	2.4	0.02	33	1.3	**
<i>Laneella nigripes</i>	25	1.7	0.02	4	1.3	0.01	24	11.3	0.11	2	1.8	0.01	60	2.4	*
<i>Huascaromusca purpurata</i>	10	0.7	0.01	4	0.7	0.01	4	1.9	0.02	0	0.0	0	18	0.7	N/A
<i>Eumecesbrinella quadrlinnea</i>	1	0.1	0	1	0.2	0	0	0.0	0	0	0.0	0	2	0.1	N/A
<i>Chrysomya putoria</i>	0	0.0	0	1	0.2	0	3	1.4	0.01	4	2.4	0.02	8	0.3	N/A
<i>Chrysomya albiceps</i>	0	0.0	0	4	0.7	0.01	1	0.5	0	16	9.6	0.09	21	0.9	N/A
<i>Paralucilia pseudolyrcea</i>	1	0.1	0	0	0.0	0	0	0.0	0	4	2.4	0.02	5	0.2	N/A
<i>Cochliomyia macellaria</i>	1	0.1	0	1	0.2	0	2	0.9	0.01	12	7.2	0.07	16	0.7	**
<i>Hemilucilia segmentaria</i>	0	0.0	0	0	0.0	0	1	0.5	0	1	0.6	0.01	2	0.1	N/A
<i>Chrysomya megacephala</i>	0	0.0	0	0	0.0	0	0	0.0	0	1	0.6	0.01	1	0.0	N/A
Total	1,482	60.3	0.77	596	24.2	0.87	213	8.7	0.66	165	6.8	0.50	2,456	100	
Total of species	9			9			10			11			13		
H'	0.3136			0.2310			0.5276			0.708					
J'	0.3287			0.2421			0.5276			0.6799					
I _j	4			3			1			2					

N/A not analyzed, * $P < 0.05$, ** $P > 0.05$

the more preserved fragment (FO $n = 1482$). However, the greatest number of species ($S = 11$) occurred in the more impacted environment (PA). The lowest values of diversity were found in most preserved environments (FO $H' = 0.31$, and CA $H' = 0.23$) due to high dominance, as shown by the Berger–Parker index (d) values (Table 1). This was expected because the dominance of one or more species in a given environment tends to reduce local diversity. The lowest values of equitability in fragments CA and FO (Table 1) shows that individuals are not distributed equally among these species, which is also occasioned by dominance.

In all fragments, *Mesembrinella bellardiana* was the most abundant species ($n = 1,884$) and represented more than 50 % of all individuals collected. This species also showed significant preference ($P < 0.05$) for more preserved environments, as indicated by their abundance (FO $n = 1140$, CA $n = 521$, PL $n = 140$, and PA $n = 83$). *Mesembrinella peregrina* was the second most abundant species (FO $n = 275$, CA $n = 48$, PL $n = 24$, and PA $n = 26$) and was more closely associated with more preserved environments ($P < 0.05$). It was the second most dominant species in all fragments except for PL, where it was co-dominant with *Laneella nigripes*.

The abundance of all other species was low, though there is a variation in the order of abundance in different fragments. In general, the relative abundance did not exceed 10 % (Table 1), except for *L. nigripes* (11.3 %) in PL.

Faunistic composition between FO and CA was more similar than that between PL and PA (Fig. 1) showing that the pairs of fragments shared distinct vegetation and therefore provide diversified niches. Although *M. bellardiana*, *M. peregrina*, *L. nigripes* and *Lucilia eximia* were present in all fragments, the abundances were slightly higher when considering the sum of the number of individuals in the pairs FO and CA, except for *Cochliomyia macellaria*, where the abundance was higher in pairs PL and PA.

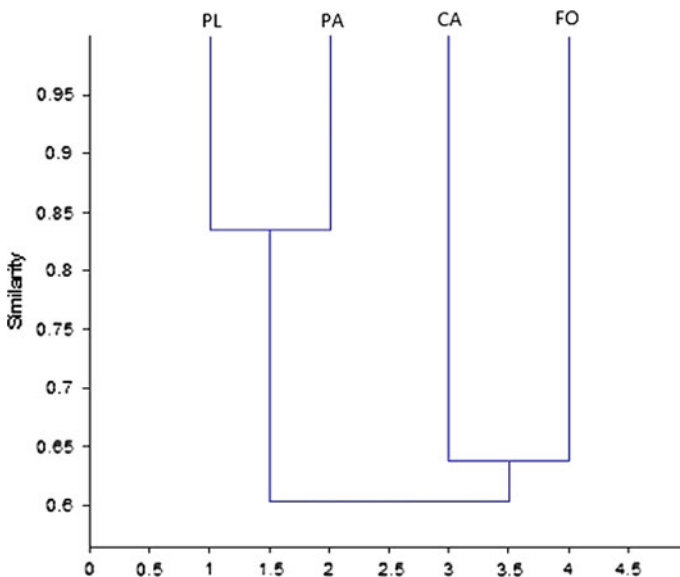


Fig. 1 Cluster of similarity (I_j) of species (Diptera: Calliphoridae) in different fragments at Intermones Farm from 2006 to 2011

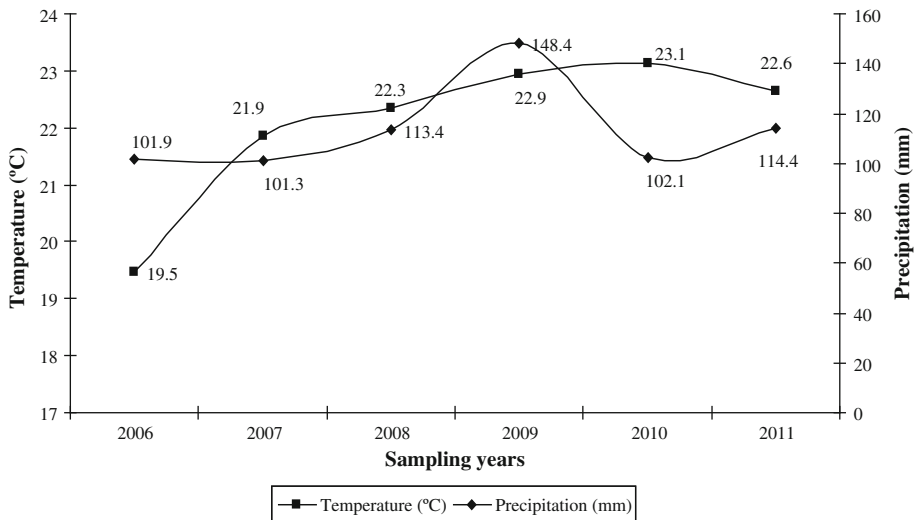


Fig. 2 Mean annual temperature (°C) and precipitation (mm) at Intermones Farm from 2006 to 2011

All fragments contained equal numbers of species considered rare ($n < 2$) varying only the faunistic composition (Table 1). *Hemilucilia segmentaria* is found only at PL and PA and *Eumesebrinella quadrilineata* is found at FO and CA. *Chrysomya megacephala*, species classified as exotic or adapted to anthropogenic environments (Linhares 1981), is present in a single habitat (PA), the most degraded environment.

Average annual temperatures ranged from 19.4 to 23.1 °C during the study period and were the lowest and highest temperatures in 2006 and 2010, respectively (Fig. 2). The highest levels of precipitation, 113.4 and 148.4 mm, were recorded during 2008 and 2009, while the lowest level, ~101 mm, was recorded between 2006 and 2007 (Fig. 2). Thus, the climate can be classified as humid temperate without a dry season, according to Köppen (1948). The values obtained from Spearman's test showed a significant positive correlation between temperature and the annual diversity indices (Fig. 3) ($r = 0.84$; $P < 0.05$), but there was no significant correlation with rainfall ($r = 0.35$; $P > 0.05$).

Discussion

Sampling was originally proposed for Lepidoptera. However, given the considerable abundance of dipterans (Calliphoridae) collected, we decided to analyze some ecological parameters that could be useful to assess the impacts of different levels of disturbance on biological diversity in the area of our study and to determine the possible use of these insects as bioindicators of local diversity.

A factor that may be useful in validating the use of our data is the long period of collection (~36 months), which increases the species inventory and the probability of detecting rare species (Summerville et al. 2003). Most notable, we recorded the first occurrence of *Calliphora loyesi* in the State of São Paulo, which had only been reported in Southern Brazil and the highlands of the state of Rio de Janeiro.

The biology and ecology of blowflies is widely documented in the literature because of their importance in the medical, veterinary, and forensic fields (Linhares and Thyssen

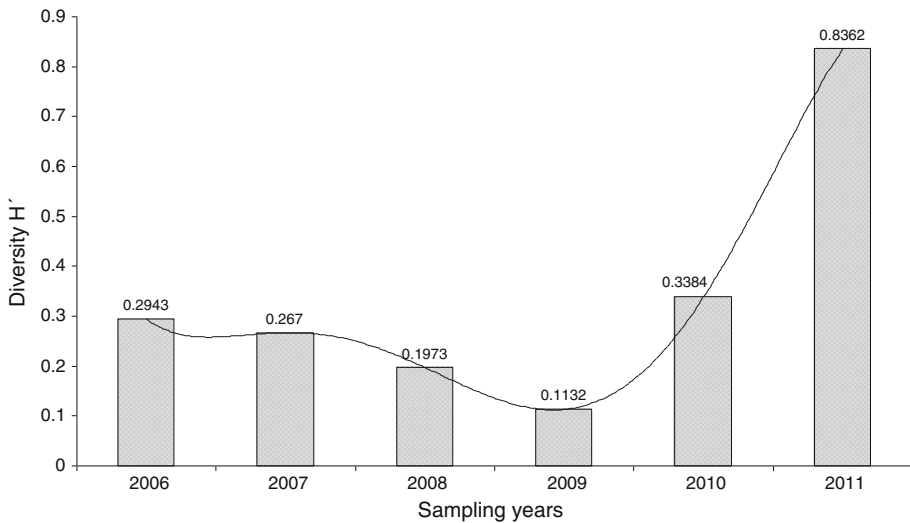


Fig. 3 Total diversity of species (H') by year of collection, at Intermontes Farm from 2006 to 2011

2007). In large part, this importance is due to the synanthropic behavior exhibited by most of the species (Greenberg 1971). However, with regard to species belonging to the sub-family Mesembrinellinae, mostly from wildlife, the habits of adults and the niche exploited by immatures are almost unknown (Guimarães 1977), which makes the necessity of studies on these native dipterans even more relevant.

The highest abundance of Mesembrinellinae species has been associated with the most preserved areas of the Amazon rainforest (Esposito and Carvalho 2006) and Atlantic Rainforest (Gadelha et al. 2009; Ferraz et al. 2010). In our study, a progressive increase in abundance of these species, in particular of *M. bellardiana*, as the area become less impacted, corroborates the existence of adaptations of these flies for this type of environment and ecosystem. Mata et al. (2008) obtained satisfactory results in a study of Drosophilidae (Diptera) in intact and disturbed areas of “cerrado”, the Brazilian savanna-like biome, and demonstrated that native species may become promising monitoring tools in these environments.

In vegetation recovery projects, given the monitoring conditions, is notorious observe that as they occur the rescue of diversity and the direction to the equilibrium environmental, insects respond in terms of density and diversity (McGeoch 1998). In our study, we observed that density (or abundance) seemed to follow this path, but not diversity, probably due to the dominance of *M. bellardiana*. Since there are no studies on the biology of this species in the literature we don't know whether it might have a greater preference for bait used in our traps, when compared to the other species.

Because of its abundance, *M. peregrina* also seems to fulfill the requirements as characteristic species of preserved environments, represented in our case by fragment FO. As expected, the synanthropic species are not characteristic of preserved environments, although there were few records of *Chrysomya* spp. rarely associated with the forest environment (D'Almeida and Lopes 1983; Leandro and D'Almeida 2005). Regarding to the other species of Calliphoridae found, the lack of more data on biology and ecology affect the understanding of how they could be appropriately used for monitoring and preservation of natural environments.

According to Ferreira and Lacerda (1993), the distribution of Calliphoridae is strongly influenced by variations in climatic conditions and each species may respond with different seasonal patterns. Even in this unpretentious study, we observed that diversity was influenced by temperature.

From the present study, we concluded that: (1) the trap Shuey described for Lepidoptera collected a representative sample of muscomorpha fauna in both preserved and less preserved areas, especially in the most preserved areas; (2) the area in environmental recuperation (PL) in Interfontes Farm has a high diversity of Calliphoridae, but with marked dominance of few species; (3) planting native seedlings for restoration of original conditions has not had a significant effect on increasing local Calliphoridae diversity; and (4) the presence of *Calliphora lopesi* suggests that the studied area is part of an important ecological corridor of the Atlantic Forest biome and that there may be other species of calliphorids not yet recorded or known at the State of São Paulo or even the rest of the country.

Acknowledgments The Department of Environment Company “Cimento Ribeirão Grande” by financial support and permission to access the collection area. To Dr. Paulo R. S. Bunde (UFPEl) for critical reading and suggestions.

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