

RESEARCH

The Potential Uses of Sarcosaprophagous Flesh Flies and Blowflies for the Evaluation of the Regeneration and Conservation of Forest Clearings: A Case Study in the Amazon Forest

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ABSTRACT. The level of association between dipterans of the families Calliphoridae and Sarcophagidae and habitats with different levels of vegetation cover was analyzed at Porto Urucu in Coari, Amazonas, Brazil, with the aim of identifying the potential of these taxa as bioindicators for the assessment of forest regeneration and conservation. The flies were collected in 16 sample areas, 12 of which were clearings at different stages of regeneration (C1—early regeneration; C2—moderate regeneration; and C3—advanced regeneration) and 4 in continuous forest (F). According to the IndVal analysis, nine sarcophagid species—*Peckia* (*Sarcodexia*) *lambens* (Wiedemann), *Peckia* (*Peckia*) *chrystostoma* (Wiedemann), *Peckia* (*Squamatodes*) *ingens* (Walker), *Sarcofahrtiopsis* *cuneata* (Townsend), *Oxysarcodexia* *thornax* (Walker), *Peckia* (*Euboettcheria*) *collusor* (Curran & Walley), *Oxysarcodexia* *fringidea* (Curran & Walley), *Oxysarcodexia* *amorosa* (Schiner), and *Helicobia* *pilifera* (Lopes)—were associated indiscriminately with clearings (C1 + C2 + C3). In contrast, only one calliphorid species *Chrysomya albiceps* (Wiedemann) was associated with clearings in the early moderate regeneration (C1 + C2) phases, and four calliphorids were associated with continuous forest or mature clearings (C3 + F): *Mesembrinella bicolor* (F.), *Eumesebrinella randa* (Walker), *Mesembrinella bellardiana* (Aldrich), and *Lucilia eximia* (Wiedemann). These results indicate that sarcophagids may be useful for evaluating the degree of anthropogenic impact but are not suitable for the detection of minor variations in forest cover. In contrast, calliphorids may be appropriate for the evaluation of both anthropogenic impacts and the degree of forest regeneration and conservation.

Key Words: conservation, Diptera, environmental impact, indicator species, neotropical region

Dipteran insects (flies) are potentially useful bioindicators for the evaluation of impacts and the monitoring of forest recuperation (Majer 1987). Their suitability is based on the fact that they tend to be very numerous, which facilitates the collection of large quantities of specimens, and also taxonomically diverse, which permits the analysis of community-level effects, based on the varying contribution of different species. These insects also occupy a wide variety of niches and several different trophic levels (Majer 1987). Given their ecological diversity, dipterans—larvae and adults—can be found in a wide range of habitats, both in the wild and in anthropogenic environments in rural or urban contexts (D’Almeida and Lopes 1983, Paraluppi and Castellon 1994, Triplehorn and Johnson 2011). Because individual dipteran species are often restricted to a narrow range of environmental conditions (Nuorteva 1963, Dias et al. 1984), analyses of dipteran community composition can provide a sensitive barometer for detecting even subtle changes in the environment.

Species of the families Calliphoridae and Sarcophagidae are found in both natural and anthropogenic environments. They differ substantially, however, in their sensitivity to man-made impacts. Some calliphorid species, such as *Eumesebrinella randa* (Walker) and *Hemilucilia semidiaphana* (Rondani), are more abundant in well-preserved forest habitats (Sousa et al. 2010), which makes them especially useful for the detection of anthropogenic impacts on these environments.

In environments in the Amazon basin, a reduction in the species richness and diversity of calliphorids has been recorded in anthropogenic environments (Esposito et al. 2009). Sousa et al. (2010, 2011b) suggested that this finding reflects the fact that some species occur primarily in densely forested habitats, whereas other species tend to be

found in more open areas. Studies of the Sarcophagidae, in contrast, have shown that most species are well-adapted to anthropogenic impacts and that habitat disturbance may yield an increase in both the abundance and species richness of this taxon (Sousa et al. 2011a,b).

These previous studies suggest that the sarcophagids are less harmed by the impacts associated with the clearance of forest vegetation and may in fact prefer more open habitats. This preference may be explained by sarcophagid traits that are advantageous for the utilization of open habitats (Sousa et al. 2011a). Unlike other ecologically similar taxa, sarcophagids are viviparous or ovoviviparous (Shewell 1987); this increases their development rate and makes them more efficient at exploiting the relatively ephemeral resources characteristic of open habitats.

Given these contrasts, this study aimed to evaluate whether calliphorids and sarcophagids were effective bioindicators by testing how strongly flies in these families were associated with environments with different degrees of vegetation cover at Porto Urucu in the Brazilian state of Amazonas. In particular, it was predicted that although different calliphorid species would be associated with different levels of forest cover, most sarcophagid species would be associated with more open areas with reduced regrowth. The confirmation of this prediction might allow individual calliphorid species to be used as indicators of relatively well-preserved habitats and individual sarcophagid species as indicators of impacted environments.

Materials and Methods

Study Area. The study area is located in the Geólogo Pedro de Moura operations base), the only active Petrobrás oil and gas field in

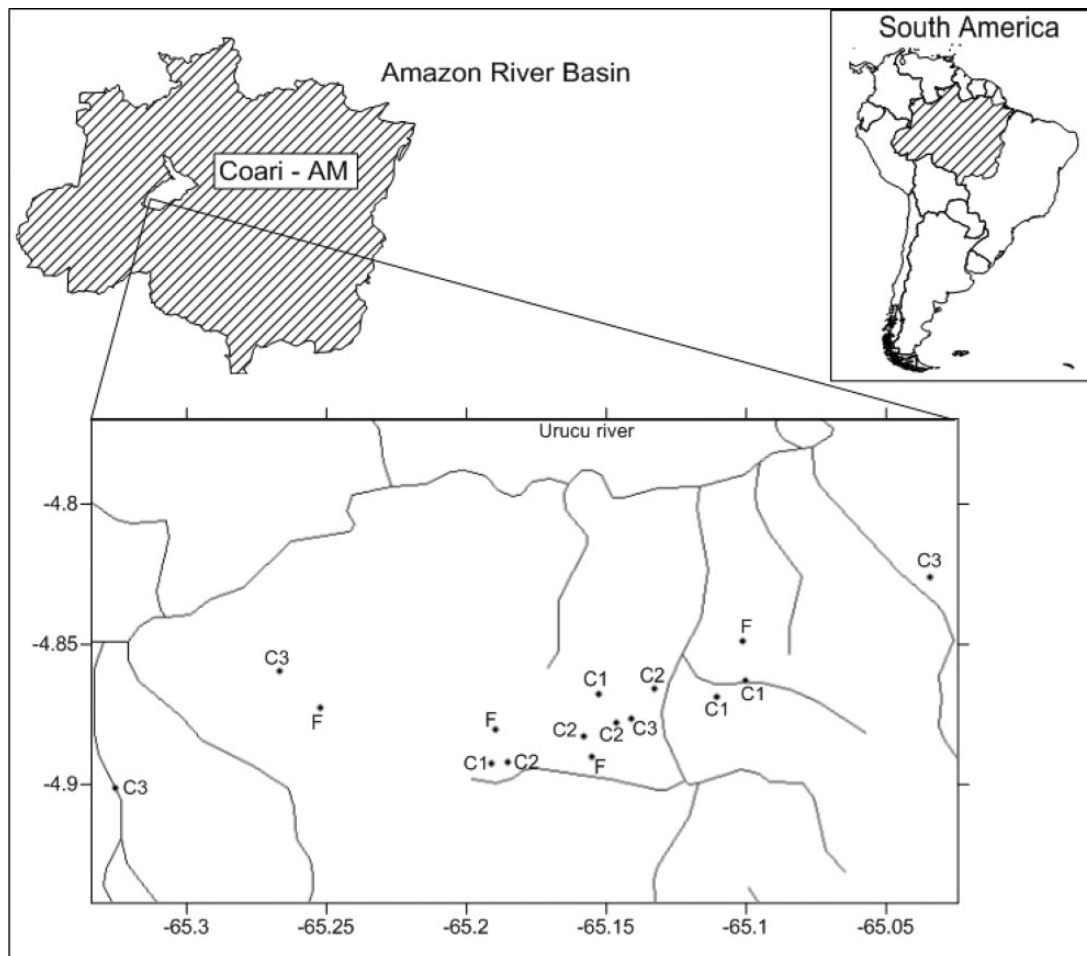


Fig. 1. Map showing the 16 areas studied in the Base Operacional Geólogo Pedro de Moura, Urucu River Basin, Coari, Amazonas, Brazil.

the Amazon basin. This base is located in an isolated area of primary forest, 680 km southwest of the city of Manaus, in the municipality of Coari, in the Brazilian state of Amazonas (Fig. 1). In addition to extensive areas of primary forest, a number of artificial clearings have been created to support oil prospecting exploration activities, and road maintenance. These clearings vary in their age and degree of regeneration (Lima and Rosário 2008).

Sampling and Analysis. In total, 16 different areas were sampled, of which 12 were clearings in different stages of regeneration (differentiated from each other on the basis of vegetation height; Pilar 1996). The other four sites were covered with primary forest, as described by Sousa et al. (2010, 2011a). The environments were classified in four categories—C1 (clearings with little regeneration, a predominance of herbaceous plants and vegetation 30–50 cm in height); C2 (clearings with moderate regeneration, a predominance of shrubs and vegetation 0.5–2 m in height); C3 (clearings at an advanced stage of regeneration, with a predominance of trees 2–5 m in height), and F (primary forest). Four flytraps were used in each area, for a total of 16 traps per environmental category and 64 total traps per sampling date. Each flytrap (see Almeida et al. 2003 for a detailed description) were baited with 50 g of rotting beef lung and exposed in the field for 48 h. This protocol was repeated three times, in April, June, and October 2007, for a total of 192 traps. In this study, the areas were considered as sampling units. Further details on the study area and procedures can be found in Sousa et al. (2011a).

The keys of Guimarães (1977), Ribeiro and de Carvalho (1998), de Carvalho and Ribeiro (2000), and Mello (2003) were used for the identification of blowflies, whereas Lopes (1939, 1946, 1958, 1976, 1989), Lopes and Tibana (1982, 1987), and Pape and Mello-Patiu (2006) were

used to identify the flesh flies. For the identification of sarcophagids, the terminalia of the male specimens were exposed using the techniques described by Lopes (1973) and Dahlem and Naczi (2006). The genital structure was exposed by pinning back the surface of the cerci (Mulieri et al. 2010).

In some species, the females were identified based on known diagnostic traits (Tibana and Mello 1985, Pape 1996), whereas in others, they were determined by the assignment of the males. The association of the sexes was evaluated based on the presence of males and females collected simultaneously in the same trap.

The degree of association of each species with a given type of habitat and the potential of the species analyzed for use as indicators of a given environmental trait were evaluated using the Indicator Value Method, IndVal (Dufrene and Legendre 1997, De Cáceres and Legendre 2009, De Cáceres et al. 2010). This approach considers two criteria—specificity and fidelity. A perfect indicator species will be present in every one of the samples (i.e., perfect fidelity) taken within a given habitat category and absent in every other habitat category (i.e., perfect specificity). This method does not consider rare species (presents in only one two places) as indicators because they have the same value as the species found at all sites in the same habitat (species actually considered as an indicator). An improved method of indicator species analysis, multilevel pattern analysis (De Cáceres et al. 2010), was applied in this study. This method permits the combination of groups of sites and identifies species that indicate either a single group or a combination of several groups. This function considers all possible combinations of groups of sites and selects the combination for which the species can be best used as indicators (De Cáceres et al. 2010).

This analysis was carried out with an abundance matrix. The significance of the indicator value was tested statistically using a Monte Carlo test with 10,000 randomizations. The indices were calculated using the Indicspecies package (De Cáceres and Legendre 2009, De Cáceres et al. 2010) of the R program (R Development Core Team 2011).

Results

Association of the Calliphorids With Different Environments. In total, 7,215 calliphorid specimens were collected, representing 16 species (Sousa et al. 2010). The IndVal approach detected associations with the four different habitat categories in the eight most abundant calliphorid species (Table 1). *Mesembrinella bicolor* (F.) was associated with forest (F), whereas *E. randa* (Walker), *Mesembrinella bellardiana* (Aldrich), and *Lucilia eximia* (Wiedemann) were associated with both forest and mature clearings (F + C3). In contrast, *Paralucilia adespota* (Dear) and *Cochliomyia macellaria* (F.) were associated with intermediate levels of regeneration (C2 + C3) and *Chrysomya albiceps* (Wiedemann) with more open habitats (C1 + C2). The species *H. semi-diaphana* (Rondani) was not a good indicator, given that it was associated with both well-preserved and impacted environments (C2 + C3 + F) (Table 1).

Association of the Sarcophagids With Different Environments. In total, 3,547 sarcophagid specimens were collected, belonging to 23 species (Sousa et al. 2011a). The IndVal approach detected associations involving the 10 most abundant species (Table 2). Nine of these species were associated indiscriminately with the three types of regeneration (C1 + C2 + C3). The species *Peckia (Pattonella) intermutans* was not a good indicator because it was associated with both well-preserved and impacted environments (C2 + C3 + F) (Table 2).

Discussion

Although different calliphorid species were associated with distinct habitat categories, the sarcophagids were almost invariably associated

with clearings. This indicates that they are less sensitive to minor alterations in the vegetation cover and may in fact benefit from the removal of the original cover. Some of the species of the family Calliphoridae, which have more restricted habitat preferences, may be appropriate as indicators of forest regeneration and conservation. In contrast, although sarcophagids may prove useful to evaluate the incidence of anthropogenic impacts, they are not appropriate for the assessment of minor variations in the cover.

The results support our hypothesis that individual calliphorid species are indicative of different levels of forest regeneration, whereas sarcophagid species are generally indicative of impacted environments. This suggests that species in these two families may prove a useful tool for biomonitoring habitats, especially for detecting anthropogenic impacts and evaluating forest conservation and regeneration.

Four calliphorid species are clearly associated with well-preserved forests or clearings at an advanced stage of regeneration. Three of these species are mesembrinellines, which have a strong ecological fidelity with the tropical rainforests of the Guianan-Brazilian subregion and have been classified as asynanthropic by Ferreira (1978) and Nuorteva (1963), i.e., species that do not adapt well to anthropogenic environments (Mello et al. 2007).

The phylogeography of the mesembrinellines indicates isolation from other groups and suggests that it originated in Gondwana. The type of vegetation appears to be the primary factor determining the distribution of these flies. The adults were regularly collected within areas of humid forest and were only observed in open habitats in the early morning and late afternoon, or on cloudy days, always near the edge of the forest, where they were attracted to decaying fallen fruits (Guimarães 1977).

The species *M. bellardiana* is not adapted to anthropogenic habitats (D'Almeida and Lopes 1983) and tends to be more abundant in better-preserved environments (Ferraz et al. 2010a), reflecting its sensitivity to anthropogenic impacts. Esposito et al. (2009) also recorded that *E. randa* and *M. bicolor* prefer preserved forest habitats. Given this, it seems likely that mesembrinelline species diversity will increase with the degree of forest preservation and that this group may be efficient indicators of preserved forest while also providing insights into ecological impacts in the study area (Gadelha et al. 2009).

L. eximia, which is also associated with forests and mature clearings, is common in forests and rural areas in Brazil (Ferreira 1978, Linhares 1981, Madeira et al. 1982), Peru (Baumgartner and Greenberg 1984), and Argentina (Mariluis and Schnack 1989). Studying synanthropic flies, Rodrigues-Guimaraes et al. (2008) and De Souza and Zuben (2012) recorded a preference of *L. eximia* for undisturbed forest. In the Brazilian municipalities of Campinas, São Paulo (Linhares 1981), Belo Horizonte, Minas Gerais (Madeira et al. 1982), and Goiânia, Goiás (Ferreira 1983), however, this species presented a preference for inhabited rural areas.

Table 1. Preferences for different habitat categories (well-preserved forest and clearings at different stages of regeneration) in calliphorid species (Diptera) sampled at the oil exploration base in the Urucu River basin in Coari, Amazonas, Brazil

Species	IndVal	P	Association
<i>M. bicolor</i> (F., 1805)	0.447	0.047	F
<i>Ch. albiceps</i> (Wiedemann, 1819)	0.59	0.018	C1 + C2
<i>P. adespota</i> (Dear, 1985)	0.884	0.001	C2 + C3
<i>C. macellaria</i> (F.)	0.673	0.001	C2 + C3
<i>E. randa</i> (Walker, 1849)	0.884	0.001	C3 + F
<i>M. bellardiana</i> (Aldrich, 1922)	0.637	0.011	C3 + F
<i>L. eximia</i> (Wiedemann, 1819)	0.545	0.019	C3 + F
<i>H. semi-diaphana</i> (Rondani, 1850)	0.834	0.001	C2 + C3 + F

Associations were identified based on IndVal scores.

Table 2. Preferences for different habitat categories (well-preserved forest and clearings at different stages of regeneration) in sarcophagid species (Diptera) sampled at the oil exploration base in the Urucu River basin in Coari, Amazonas, Brazil

Species	IndVal	P	Association
<i>Pe. (Sarcodexia) lambens</i> (Wiedemann, 1830)	0.998	0.001	C1 + C2 + C3
<i>Pe. (Peckia) Chrysostoma</i> (Wiedemann, 1830)	0.981	0.001	C1 + C2 + C3
<i>Pe. (Squamotodes) ingens</i> (Walker, 1849)	0.957	0.001	C1 + C2 + C3
<i>Sarcophahrtiopsis cuneata</i> Townsend, 1935	0.935	0.001	C1 + C2 + C3
<i>O. thornax</i> (Walker, 1849)	0.816	0.001	C1 + C2 + C3
<i>Pe. (Euboettcheria) collusor</i> (Curran & Walley, 1934)	0.811	0.008	C1 + C2 + C3
<i>O. fringidae</i> (Curran & Walley, 1934)	0.75	0.001	C1 + C2 + C3
<i>O. amorosa</i> (Schiner, 1868)	0.75	0.001	C1 + C2 + C3
<i>Helicobia pilifera</i> Lopes, 1939	0.629	0.005	C1 + C2 + C3
<i>Pe. (Pattonella) intermutans</i> (Walker, 1861)	0.825	0.003	C2 + C3 + F

Associations were identified based on IndVal scores.

The species *C. macellaria* has been collected in both natural habitats and anthropogenic areas at a number of different sites (Esposito et al. 2009, Ferraz et al. 2010a, Sousa et al. 2010). In some of these studies, however, the species was more closely associated with habitats that had suffered some degree of alteration, such as forest edges (Ferraz et al. 2010b) and pastures (Gomes et al. 2000, Koller et al. 2011).

Esposito et al. (2009) found that *P. adespota* was more abundant in open areas adjacent to urban environments than in primary forest, indicating that this species tends to be associated with habitats that have suffered some degree of anthropogenic impact, as observed in this study. One other species, *Ch. albiceps*, is also considered characteristic of man-made environments, in particular urban centers (Ferreira 1983, Baumgartner and Greenberg 1984, Mendes and Linhares 1993). This species has been recorded in anthropogenic environments (urban centers and forests) in southeastern Brazil (Leandro and D'Almeida 2005; Furusawa and Cassino 2006; Ferraz et al. 2009, 2010a) and the Amazon basin (Paraluppi and Castellón 1994, Esposito and Linhares 2002). These findings indicate that the calliphorid species associated with forest habitats and mature clearings may be used as bioindicators of environmental quality.

In the case of the sarcophagids, the fact that all but one of the species collected in this study occurred in all three types of clearing (C1, C2, and C3) suggests that they could be used as indicators of degraded environments. The ability of many sarcophagid species to rapidly exploit the ephemeral resources typical of degraded environments may lead to these species benefitting from the removal of forest cover (Sousa et al. 2011a).

In a number of studies, larger numbers of sarcophagid flies were collected on bait exposed in sunny areas than those in the shade (Mulieri et al. 2011), whereas adults were observed flying or visiting flowers during the sunniest hours of the day (Willmer and Unwin 1981, Willmer 1982). The coloration of the abdominal cuticle of the sarcophagids has a high capacity for thermal reflectance (Willmer 1982), which would help avoid overheating. These flies also appear to be able to alternate the pumping of the hemolymph from the thorax to the abdomen, and vice versa. These two mechanisms may combine to improve thermoregulation in open environments, allowing these flies to feed during the parts of the day when other flies (potential competitors) are obliged to seek refuge in the shade.

In addition, the larvae of some sarcophagid species are substantially resistant to desiccation. Lopes (1973) compared emergence rates in *Oxysacodexia angrensis*, which is found in humid habitats, with those of *Oxysacodexia amorosa*, which is typical of open areas, under the same conditions in the laboratory. Although only a few specimens of *O. angrensis* were able to emerge, all the *O. amorosa* emerged normally.

Given these differences, clearings, which are more open and exposed to solar radiation than closed forest habitats, should be occupied preferentially by sarcophagid species with desiccation-tolerant larvae (Lopes 1973) and adults that possess the adaptations necessary to avoid overheating (Willmer 1982). The clear association of certain species with impacted environments indicates that this taxon may accurately indicate the removal of original forest vegetation.

Determining the occurrence or abundance of a small set of indicator species, rather than the entire community, has been a particularly useful strategy for long-term environmental monitoring in conservation or ecological management programs (De Cáceres et al. 2010). The Indval approach was chosen for this study because it assesses the relationship between the occurrence and abundance of species and a set of environmental features, permitting the evaluation of the association of species and thus their usefulness as bioindicators. Although this method is widely used for this purpose, it may not be useful for evaluating rare (singleton or doubleton) species due to their low occurrence rates. However, the list of species considered to be closely associated allows a recently surveyed area to be classified as impacted or preserved on the basis of species detections. The predictive value of many indicator

species makes them valuable to conservationists and land managers because they provide a cost- and time-efficient approach for the assessment of habitat characteristics (Hilty and Merenlender 2000, Carignan and Villard 2002, McGeoch et al. 2002).

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