

Environmental Variation in Parasitism of Synanthropic Diptera: Host Use, Habitat Effects, and Biological Control Implications

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Abstract: Parasitoids are key regulators of insect populations, particularly those associated with decomposing organic matter and synanthropic environments. This study analyzed parasitoid–Diptera interactions across different environments and substrates, with emphasis on parasitism patterns, species diversity, host associations, and ecological relationships. Data were compiled from experimental studies conducted in Brazil, including forest, urban, pasture, livestock, and agricultural systems. Sampling methods included baited traps, exposure of organic substrates, recovery of pupae, and laboratory emergence of Diptera and parasitoids. Parasitism rates were standardized to allow comparisons among studies. In addition, the quantitative data available in several articles enabled complementary chi-square analyses, although these tests were not applied or reported in many of the original studies. The results showed that parasitoid diversity, abundance, and parasitism varied according to substrate type, host availability, and environmental conditions. Parasitoids are key regulators of insect populations, particularly those associated with decomposing organic matter and synanthropic environments. This study analyzed parasitoid–Diptera interactions across different environments and substrates, with emphasis on parasitism patterns, species diversity, host associations, and ecological relationships. Data were compiled from experimental studies conducted in Brazil, including forest, urban, pasture, livestock, and agricultural systems. Sampling methods included baited traps, exposure of organic substrates, recovery of pupae, and laboratory emergence of Diptera and parasitoids. Parasitism rates were standardized to allow comparisons among studies. In addition, the quantitative data available in several articles enabled complementary chi-square analyses, although these tests were not applied or reported in many of the original studies. The results showed that parasitoid diversity, abundance, and parasitism varied according to substrate type, host availability, and environmental conditions. Parasitoids such as *Aphaereta* sp. (Hymenoptera: Braconidae), *Nasonia vitripennis* (Walker, 1836) (Hymenoptera: Pteromalidae), *Pachycrepoideus vindemmiae* (Rondani, 1875) (Hymenoptera: Pteromalidae), and *Spalangia* spp. (Hymenoptera: Pteromalidae) were frequently associated with synanthropic Diptera, including *Chrysomya albiceps* (Wiedemann, 1819) (Diptera: Calliphoridae), *Musca domestica* L., 1758 (Diptera: Muscidae), and *Peckia chrysostoma* (Wiedemann, 1830) (Diptera: Sarcophagidae). Higher parasitoid activity was observed in environments rich in decomposing organic matter, especially animal feces, carcasses, poultry manure, and organic waste. Both generalist and more specialized parasitoids were recorded, indicating different ecological strategies and levels of host specificity. Overall, the findings highlight the ecological importance of parasitoids as natural enemies of Diptera and support their potential application in biological control programs. The complementary statistical analyses strengthened the interpretation of patterns previously described mainly in descriptive terms.

Keywords: Biological Control, Decomposing Substrates, Diptera, Host–Parasitoid Interaction, Parasitism, Synanthropic Flies.

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1.0. INTRODUCTION

Diptera species are widely distributed across diverse environments and are commonly associated with decomposing organic matter, playing an important role in nutrient cycling and ecological balance. Some species, however, have sanitary, veterinary, and economic importance because they can transmit pathogens, cause myiasis, and develop in substrates closely associated with humans and animals. Species such as *Chrysomya albiceps* (Wiedemann, 1819) (Diptera: Calliphoridae) and *Musca domestica* L., 1758 (Diptera: Muscidae) are especially relevant due to their synanthropic behavior and their occurrence in animal carcasses, feces, poultry manure, organic waste, and other decomposing substrates (Marchiori, 2019a; Marchiori, 2021a; Guimarães *et al.*, 2026; Österman *et al.*, 2026).

Parasitoids are important natural enemies of Diptera and help regulate fly populations across different ecological systems. These insects develop in or on immature hosts, especially larvae and pupae, usually causing host death. Several parasitoid groups are associated with synanthropic flies, including species that occur in animal feces, carcasses, poultry manure, viscera, liver, fish, and other organic substrates. The diversity and occurrence of parasitoids may vary according to host availability, substrate type, environmental conditions, seasonality, and habitat structure (Marchiori, 2019b; Marchiori, 2019c; Marchiori, 2019d; Marchiori, 2021b; Cingolani *et al.*, 2025).

Studies on parasitoids associated with synanthropic and economically important Diptera are essential for understanding host–parasitoid interactions and for supporting biological control programs. The identification of parasitoid species, their hosts, substrates, and parasitism rates important information for the management of fly populations of medical, veterinary, agricultural, and sanitary relevance. In this context, parasitoids such as *Aphaereta* sp. (Hymenoptera: Braconidae); *Brachymeria podagrica* (Fabricius, 1789) (Hymenoptera: Chalcididae), *Hemencyrtus herbertii* Ashmead, 1900 (Hymenoptera: Encyrtidae) *Nasonia vitripennis* (Walker, 1836) (Hymenoptera: Pteromalidae); *Pachycrepoides vindemmiae* (Rondani, 1875) (Hymenoptera: Pteromalidae), species *Spalangia* Latreille, 1805 (Hymenoptera: Pteromalidae), and *Trichopria* sp. (Hymenoptera: Diapriidae) are relevant because they are frequently associated with Diptera developing in decomposing organic substrates (Marchiori, 2019c; Schuster and Sivakumar, 2024; Cingolani *et al.*, 2025).

Although many experimental studies have reported parasitoids associated with Diptera in Brazil, several of these works presented the data mainly in descriptive form. In many cases, the tables included quantitative information such as the number of pupae, number of parasitoids, the number of parasitized pupae, frequencies, substrates, hosts, and parasitism rates. These

data made it possible to perform complementary chi-square analyses, even when such statistical tests had not been applied or reported in the original articles. Therefore, the present review adds a statistical approach to previously published experimental information, providing stronger support for patterns of parasitoid abundance, host association, substrate use, habitat influence, and parasitism.

This study aimed to review experimental works on parasitoid species associated with dipteran larvae and pupae collected from different substrates and environments, to evaluate their occurrence in relation to host species, substrate type, and environmental conditions, and to use the available quantitative data to perform complementary chi-square analyses when possible. By combining biological information with statistical support, this review contributes to the knowledge of parasitoid diversity associated with synanthropic and pest Diptera and provides information that may support future biological control and integrated pest management programs.

2.0. MATERIALS AND METHODS

2.0.1. Study Design

This study consisted of an integrative analysis based on experimental investigations conducted under both field and laboratory conditions. The focus was on interactions between parasitoids and Diptera across a range of ecological settings. The compiled data originated from independent studies carried out in different regions of Brazil, including Goiás and Minas Gerais, encompassing forested, rural, urban, and agricultural environments.

2.0.2. Data Sources and Experimental Basis

The dataset used in this study was obtained from previously performed experimental research involving the collection of dipteran hosts and their associated parasitoids. These investigations applied comparable sampling procedures, such as the use of baited traps, exposure of organic substrates (e.g., feces, animal tissues, and fruits), and pitfall trapping techniques. Across the selected studies, similar methodological principles were followed, including:

- Exposure of substrates to enable colonization by dipteran species.
- Recovery of pupae from natural or experimentally exposed substrates.
- Maintenance of pupae under controlled laboratory conditions.
- Identification of emerged dipteran and parasitoid specimens.

2.0.3. Inclusion Criteria

Studies were considered eligible for inclusion when they satisfied the following conditions:

1. Conducted under field or semi-natural conditions.

2. Involved dipteran species associated with decomposing organic substrates.
3. Reported the emergence of parasitoids from host pupae.
4. Included quantitative data on parasitism rates, host identity, or parasitoid diversity.

Both published studies and unpublished datasets were included, provided that methodological consistency was ensured.

2.0.4. Data Compilation and Standardization

Information extracted from the selected studies was organized into a standardized database. The following variables were recorded:

- Dipteran host species.
- Parasitoid species.
- Type of substrate (e.g., feces, carcasses, and fruits).
- Environmental context (forest, rural, and urban).
- Number of collected pupae.
- Number of emerging parasitoids.
- Parasitism rate (%).

To allow comparisons among studies, parasitism rates were calculated using the following expression: $\text{Parasitism (\%)} = (\text{number of parasitized pupae} / \text{total number of pupae}) \times 100$.

2.0.5. Experimental Methods

1. Method 1

Adult flies were collected using traps constructed from matte black metal cans measuring approximately 19 cm in height and 9 cm in diameter. Each can have two slit-type openings located in the lower third to allow insect entry. Nylon funnels were attached to the upper portion of the cans, with the narrow ends directed downward. The wider upper ends were connected to plastic collection bags used to capture the flies. Baits consisting of human feces, fish, bovine liver, chicken, and fruit were placed on a layer of soil inside the traps. A total of 20 traps were used, with ten installed at 740 m altitude and ten at 1,000 m. Two traps were assigned to each bait type and were suspended from tree branches at a height of 1 m above the ground, spaced 2 m apart. Collected insects were transported to the laboratory, euthanized using ethyl ether, and preserved in 70% ethanol for subsequent identification. The trap contents were transferred to plastic containers containing a layer of sand to allow larval pupation. After 15 days, the sand was sifted, and pupae were collected and individually placed in gelatin capsules (size 00) until adult emergence. Fly preferences for altitude and substrates were tested using ANOVA, with data transformed to $\sqrt{x} + 0.5$, adopting a 5% significance level.

2. Method 2

The material used in this study was collected in a dry forest ecosystem located in northern Peru between June and August 2017. The average temperature during the study period was 25.94°C, with a relative humidity of 62.19%. Six guinea pigs, *Cavia porcellus* Linnaeus, 1758 (Mammalia: Rodentia: Caviidae) were euthanized using a supersaturated magnesium sulfate solution to prevent bleeding. The carcasses were placed inside metal cages covered with wire mesh to prevent disturbance by vertebrates while allowing free access for insects.

3. Method 3

Flies were attracted to traps constructed from matte black metal cans (19 cm height × 9 cm diameter) with two lateral openings located in the lower third to allow insect entry. Nylon funnels were attached to the upper part of each can, with the narrow ends facing downward and the upper ends connected to plastic bags for specimen collection. Baits consisting of fruits, bovine liver, fish, human feces, and chicken viscera were placed on a layer of soil inside each trap and replaced weekly. Five traps were randomly hung on *Eucalyptus* sp. trees at a height of 1 m above ground level, spaced 2 m apart, and located approximately 50 m from a domestic waste deposit. Collected specimens were transported to the laboratory, euthanized using ethyl ether, and preserved in 70% ethanol. The trap contents were placed in plastic containers with sand to allow larval development. After 15 days, pupae were extracted and individually placed in gelatin capsules (size 00) until emergence of Diptera and/or parasitoids.

4. Method 4

The experiment with *Tuta absoluta* (Meyrick, 1927) (Lepidoptera: Gelechiidae) was conducted using tomato plants grown in 20-L PVC pots under greenhouse conditions at 27°C and 60% relative humidity. When the plants reached 40 days of age, leaves containing pupae were collected and transferred to the Biological Control Laboratory. Pupae were removed using forceps, counted, and individually placed in glass containers until the emergence of Lepidoptera or parasitoid adults. Emerged individuals were identified under a stereomicroscope and preserved in 70% ethanol.

5. Method 5

Malaise traps were used to capture insects by flight interception. These traps were constructed using fabric panels that direct insects upward into two connected plastic collection bottles (200 mL each). The lower bottle contained a preservative solution (Dietrich's solution: 600 mL of 96% ethanol, 300 mL distilled water, 100 mL of 40% formaldehyde, and 20 mL acetic acid). The traps were oriented northward to enhance parasitoid attraction. Collected insects were removed every two weeks using a fine mesh sieve and preserved in 70% ethanol for later identification.

6. Method 6

Fresh feces were marked immediately after deposition in pastures using wooden stakes (30 cm in height $\times$ 5 cm in thickness) to determine their age accurately. Twenty fecal pats were left in the field for eight days and subsequently collected along with approximately 5 cm of the underlying and surrounding substrate. Pupae were extracted using the flotation method, separated with a sieve, counted, and individually placed in gelatin capsules (size 00) until emergence of flies and/or parasitoids. Emerged parasitoids were identified using a stereomicroscope and preserved in 70% ethanol.

7. Method 7

Parasitoids were surveyed in areas of native vegetation and pasture using ten yellow pan traps or a Moericke trap filled with water, placed at ground level. Five traps were positioned in pasture areas and five in forested areas. Each trap consisted of a yellow basin (approximately 30 cm in diameter) containing a mixture of 2 L of water, 2 mL of detergent, and 2 mL of formaldehyde. Insects attracted to the yellow color fell into the solution, were collected using a fine sieve, and preserved in 70% ethanol for identification under a stereomicroscope.

In addition, parasitoids associated with muscoid flies were collected from cattle corrals. Fecal material was accumulated in a manure pile to serve as a substrate for fly development. Pupae were collected biweekly from manure older than eight days. Samples consisted of five portions of bovine feces (40 cm diameter $\times$ 12 cm height). Pupae were obtained using the flotation method, individually placed in gelatin capsules, and maintained until the emergence of flies and parasitoids. Adults were counted and identified.

8. Method 8

Hemiptera egg collections were carried out in maize fields. At the Agronomy School Farm, a 44 $\times$ 20 m plot was used, while at Santa Maria Farm, a one-hectare area was divided into similar plots. Fifty maize ears were randomly collected weekly from each site, totaling 700 ears. The collected ears were placed

individually in plastic bags and transported to the laboratory. Each ear was examined for the presence of Hemiptera egg masses. Egg masses were transferred along with a small portion of plant tissue into glass containers and maintained under laboratory conditions until the emergence of parasitoids or host nymphs.

Note: Figures include both photographic records and schematic illustrations. Wing venation was omitted or simplified in schematic figures for clarity and does not represent diagnostic morphological variation among parasitoid taxa.

2.0.6. Data Analysis

Descriptive and comparative approaches were used to examine:

- Variation in parasitoid diversity across environments.
- Host–parasitoid relationships.
- Differences in parasitism rates among substrates and habitats.

Parasitism rates were calculated as the percentage of parasitized pupae relative to the total number of collected pupae. When available, statistical analyses reported in the original studies were considered to support interpretations regarding host preference and environmental effects. These included analysis of variance (ANOVA) for comparisons among means and chi-square tests (χ^2) for evaluating associations between parasitoid species and host occurrence.

Species were categorized according to their frequency and distribution across the analyzed studies. Faunistic indices, including constancy and dominance, were applied based on the proportion of occurrences relative to total sampling events. Because this study is based on compiled data from multiple independent investigations, differences in sampling effort, environmental conditions, and methodological procedures may influence the observed patterns. Nevertheless, the adoption of standardized criteria for data selection and analysis allowed consistent comparisons among studies (Table 1).

Table 1: Taxonomic table of the main species and taxa cited in the article

Species/taxon	Authority	Order	Family	First citation in the text	Subsequent citation
<i>Aphaereta</i> sp.	Förster, 1863	Hymenoptera	Braconidae	<i>Aphaereta</i> sp. (Hymenoptera: Braconidae)	<i>Aphaereta</i> sp.
<i>Brachymeria podagrica</i>	Fabricius, 1789	Hymenoptera	Chalcididae	<i>Brachymeria podagrica</i> Fabricius, 1789 (Hymenoptera: Chalcididae)	<i>B. podagrica</i>
<i>Chrysomya albiceps</i>	Wiedemann, 1819	Diptera	Calliphoridae	<i>Chrysomya albiceps</i> Wiedemann, 1819 (Diptera: Calliphoridae)	<i>C. albiceps</i>

Species/taxon	Authority	Order	Family	First citation in the text	Subsequent citation
<i>Chrysoperla externa</i>	Hagen	Neuroptera	Chrysopidae	<i>Chrysoperla externa</i> Hagen (Neuroptera: Chrysopidae)	<i>C. externa</i>
<i>Dermatobia hominis</i>	Linnaeus Jr., 1781	Diptera	Oestridae	<i>Dermatobia hominis</i> Linnaeus Jr., 1781 (Diptera: Oestridae)	<i>D. hominis</i>
<i>Fannia pusio</i>	Wiedemann, 1830	Diptera	Fanniidae	<i>Fannia pusio</i> Wiedemann, 1830 (Diptera: Fanniidae)	<i>F. pusio</i>
<i>Gryon gallardoi</i>	Brèthes, 1914	Hymenoptera	Scelionidae	<i>Gryon gallardoi</i> Brèthes, 1914 (Hymenoptera: Scelionidae)	<i>G. gallardoi</i>
<i>Hemencyrtus herbertii</i>	Ashmead, 1900	Hymenoptera	Encyrtidae	<i>Hemencyrtus herbertii</i> Ashmead, 1900 (Hymenoptera: Encyrtidae)	<i>H. herbertii</i>
<i>Leptoglossus zonatus</i>	Dallas, 1852	Heteroptera	Coreidae	<i>Leptoglossus zonatus</i> Dallas, 1852 (Heteroptera: Coreidae)	<i>L. zonatus</i>
<i>Leptopilina boulardi</i>	Barbotin, Carton & Kelner-Pillaut, 1979	Hymenoptera	Figitidae	<i>Leptopilina boulardi</i> Barbotin, Carton & Kelner-Pillaut, 1979 (Hymenoptera: Figitidae)	<i>L. boulardi</i>
<i>Musca domestica</i>	Linnaeus, 1758	Diptera	Muscidae	<i>Musca domestica</i> Linnaeus, 1758 (Diptera: Muscidae)	<i>M. domestica</i>
<i>Nasonia vitripennis</i>	Walker, 1836	Hymenoptera	Pteromalidae	<i>Nasonia vitripennis</i> Walker, 1836 (Hymenoptera: Pteromalidae)	<i>N. vitripennis</i>
<i>Oxysarcodexia thornax</i>	Walker, 1849	Diptera	Sarcophagidae	<i>Oxysarcodexia thornax</i> Walker, 1849 (Diptera: Sarcophagidae)	<i>O. thornax</i>
<i>Pachycrepoideus vindemmiae</i>	Rondani, 1875	Hymenoptera	Pteromalidae	<i>Pachycrepoideus vindemmiae</i> Rondani, 1875 (Hymenoptera: Pteromalidae)	<i>P. vindemmiae</i>
<i>Paraganaspis egeria</i>	Diaz, Gallardo & Walsh, 1996	Hymenoptera	Figitidae	<i>Paraganaspis egeria</i> Diaz, Gallardo & Walsh, 1996 (Hymenoptera: Figitidae)	<i>P. egeria</i>
<i>Peckia chrysostoma</i>	Wiedemann, 1830	Diptera	Sarcophagidae	<i>Peckia chrysostoma</i> Wiedemann, 1830 (Diptera: Sarcophagidae)	<i>P. chrysostoma</i>
<i>Sarcophagula occidua</i>	Fabricius, 1794	Diptera	Sarcophagidae	<i>Sarcophagula occidua</i> Fabricius, 1794 (Diptera: Sarcophagidae)	<i>S. occidua</i>
<i>Spalangia nigroaenea</i>	Curtis, 1839	Hymenoptera	Pteromalidae	<i>Spalangia nigroaenea</i> Curtis, 1839 (Hymenoptera: Pteromalidae)	<i>S. nigroaenea</i>
<i>Stomoxys calcitrans</i>	Linnaeus, 1758	Diptera	Muscidae	<i>Stomoxys calcitrans</i> Linnaeus, 1758 (Diptera: Muscidae)	<i>S. calcitrans</i>
<i>Trichopria</i> sp.		Hymenoptera	Pteromalidae	<i>Trichopria</i> sp. (Hymenoptera: Diapriidae).	<i>Trichopria</i> sp.
<i>Triplasta atrocoxalis</i>	Ashmead, 1865	Hymenoptera	Figitidae	<i>Triplasta atrocoxalis</i> (Ashmead, 1865)	<i>Tripl atrocoxalis</i>

Species/taxon	Authority	Order	Family	First citation in the text	Subsequent citation
				(Hymenoptera: Figitidae: Eucoilinae)	
<i>Tuta absoluta</i>	Meyrick, 1927	Lepidoptera	Gelechiidae	<i>Tuta absoluta</i> Meyrick, 1927 (Lepidoptera: Gelechiidae)	<i>T. absoluta</i>
<i>Zaprionus indianus</i>	Gupta, 1970	Diptera	Drosophilidae	<i>Zaprionus indianus</i> Gupta, 1970 (Diptera: Drosophilidae)	<i>Z. indianus</i>

3.0. RESULTS

The results obtained in this study demonstrated variations in parasitoid diversity, host associations, and parasitism rates across different environments and substrates. Two summary tables are presented below to synthesize the main findings. As shown in Table 2, parasitoid–host associations varied according to substrate type and environmental conditions. Species

such as *Aphaereta* sp. and *N. vitripennis* were frequently associated with *P. chrysostoma*, particularly in substrates such as fish and chicken. In contrast, *H. herbertii* was mainly associated with *M. domestica* and exhibited a larval parasitism strategy. In pasture environments, species such as *P. egeria* and *Triplasta* sp. were strongly associated with Diptera developing in bovine feces, exhibiting relatively high parasitism rates (Table 2).

Table 2: Summary of parasitoid–host associations and parasitism patterns across different environments and substrates

Environment	Substrate	Host species	Parasitoid species	Parasitism rate
Agricultural	Maize	Hemiptera eggs	<i>Gryon gallardoi</i>	High
Forest	Carcass	Calliphoridae	<i>Aphaereta</i> sp.	High
	Chicken	<i>Peckia chrysostoma</i>	<i>Brachymeria podagrica</i>	Moderate
	Fish	<i>Peckia chrysostoma</i>	<i>Nasonia vitripennis</i>	Moderate
Urban	Bovine liver	<i>Musca domestica</i>	<i>Hemencyrtus herbertii</i>	Low
	Human feces		<i>Nasonia vitripennis</i>	Moderate
	Chicken	<i>Peckia chrysostoma</i>	<i>Hemencyrtus herbertii</i>	High
	Fish	<i>Peckia chrysostoma</i>	<i>Nasonia vitripennis</i>	Moderate

Additionally, agricultural systems showed distinct parasitoid assemblages. Egg parasitoids such as *Gryon gallardoi* (Brèthes, 1914) (Hymenoptera: Scelionidae) demonstrated high parasitism rates in Hemiptera, while pupal parasitoids such as *P. vindemmiae* showed dominance in dipteran hosts, particularly *Zaprionus indianus* Gupta, 1970 (Diptera: Drosophilidae). These findings reinforce the importance of parasitoids across multiple trophic levels and host developmental stages.

The patterns observed in parasitoid diversity and ecological interactions are summarized in Table 3, which highlights the influence of environmental factors, substrate type, and host availability on parasitism dynamics. Higher parasitoid diversity was generally associated with environments containing abundant decomposing organic matter, such as urban areas and livestock systems. In these environments, increased host availability contributed to higher parasitoid occurrence and interaction frequency.

Table 3: Summary of parasitoid diversity and ecological patterns across environments, substrates, and host availability

Category	Variable	Observation	Interpretation
Behavior	Specialist species	<i>Aphaereta</i> sp.	Host-specific interaction
Behavior	Generalist species	<i>Pachycrepoideus vindemmiae</i> dominant	Adaptability to multiple hosts
Control	Biological control	Parasitoids present	Potential pest regulation
Diversity	Species richness	Higher in urban and forest areas	Environmental heterogeneity increases diversity
Environment	Altitude	No significant effect	Diptera show ecological plasticity
Hosts	Diptera abundance	High in decomposing substrates	Organic matter drives colonization
Parasitism	Rates	Variable (0.7–75%)	Depends on the host and environment
Substrate	Human feces	High diversity	Favorable for colonization
	Chicken manure	High abundance	Suitable for development

Furthermore, parasitism rates varied considerably among substrates, ranging from low values

in some systems (e.g., *M. domestica* in manure) to high values in others (e.g., egg parasitoids of Hemiptera and

pupal parasitoids of Diptera). These variations indicate that both biotic and abiotic factors play a key role in shaping parasitoid–host relationships. The results demonstrate consistent patterns of parasitoid diversity and host associations across different ecological conditions. The variability observed among substrates and environments highlights the influence of ecological factors on parasitism dynamics. These findings provide a foundation for discussing ecological interactions and the potential application of parasitoids in biological control programs.

3.0.1. Chi-Square Values by Article

Complementary chi-square analyses were applied to the quantitative datasets when the data allowed statistical comparison. Significant differences were observed in parasitoid abundance, host association, substrate use, and species distribution in several articles, whereas some comparisons, such as the abundance of Figitidae between pasture and forest areas, were not statistically significant.

Article 2: Gregarious parasitoids of muscoid Diptera collected in different substrates in Itumbiara, Goiás:

- Distribution of gregarious parasitoid individuals among species:
 $\chi^2 = 379.91$; $df = 2$; $p < 0.0001$.
- Distribution of parasitoid abundance among substrates:
 $\chi^2 = 733.43$; $df = 6$; $p < 0.0001$.

Article 5: Diptera collected in the mountain of Caldas Novas in Goiás, Brazil:

- Total abundance between altitudes:
 $\chi^2 = 64.53$; $df = 1$; $p < 0.0001$.
- Distribution of substrates between altitudes:
 $\chi^2 = 104.07$; $df = 4$; $p < 0.0001$.
- Distribution of species between altitudes:
 $\chi^2 = 1040.58$; $df = 12$; $p < 0.0001$.

Article 6: Parasitoids of the subfamily Eucoilinae collected in a remnant of Cerrado Forest in Itumbiara, Goiás:

- Abundance among Eucoilinae taxa:
 $\chi^2 = 26.40$; $df = 7$; $p = 0.0004$.

Article 7: Agricultural pests of some parasitoids collected in Brazil:

- Distribution of parasitoids among four pest hosts:
 $\chi^2 = 507.96$; $df = 3$; $p < 0.0001$.
- Abundance among parasitoid species:
 $\chi^2 = 1977.30$; $df = 10$; $p < 0.0001$.

Article 9: Eucoilinae collected in Lavras, Minas Gerais, Brazil:

- Abundance among Eucoilinae taxa:
 $\chi^2 = 26.53$; $df = 6$; $p = 0.0002$.

Article 10: Cynipoidea associated with bovine feces and collected in native forest and pasture areas in Goiás, Brazil:

- Parasitism among host species:
 $\chi^2 = 785.77$; $df = 3$; $p < 0.0001$.
- Abundance among parasitoid taxa:
 $\chi^2 = 72.40$; $df = 2$; $p < 0.0001$.
- Host–parasitoid association:
 $\chi^2 = 88.60$; $df = 6$; $p < 0.0001$.
- Abundance between pasture and native forest:
 $\chi^2 = 2.17$; $df = 1$; $p = 0.1407$.
- Distribution of Figitidae taxa between pasture and forest:
 $\chi^2 = 12.35$; $df = 9$; $p = 0.1945$.

Article 12: Microhymenopterans of *M. domestica* collected on different substrates in Brazil:

- Parasitism among substrates:
 $\chi^2 = 346.32$; $df = 5$; $p < 0.0001$.
- Abundance among parasitoid taxa:
 $\chi^2 = 80.00$; $df = 6$; $p < 0.0001$.
- Substrate–parasitoid association:
 $\chi^2 = 133.83$; $df = 30$; $p < 0.0001$.

4.0. EXPERIMENTAL MANUSCRIPTS

1. Traps for the Specimen Collection of Insects in Brazil

The different trap models used for insect collection showed clear variations in efficiency depending on their structural design and placement in the field. Overall, all traps were effective at attracting and capturing insects in the orders Diptera and Hymenoptera, particularly in environments rich in decomposing organic matter, which serves as a strong attractant. Among the evaluated methods, Moericke traps, Malaise traps, metal container traps, and pitfall traps were all considered useful tools for insect sampling, although each presented specific advantages and operational limitations. The results demonstrate that trap design plays a fundamental role in insect capture, directly influencing both the diversity and abundance of collected specimens. Features such as lateral openings, bait compartments, and the use of preservative solutions contributed to higher capture efficiency, while also ensuring better specimen preservation, which is essential for accurate taxonomic identification.

In addition to structural characteristics, environmental conditions such as temperature, rainfall, humidity, and the availability of organic substrates significantly influenced trap performance. Areas with higher accumulation of decomposing organic matter exhibited greater insect activity, resulting in increased attractiveness and higher capture rates. These findings reinforce the importance of considering environmental context when designing and implementing sampling strategies. Furthermore, the study highlights that the use of multiple trap models is essential to minimize the selectivity inherent to individual sampling methods,

thereby allowing a more comprehensive representation of the insect community. This approach is particularly

important in ecological studies aiming to assess biodiversity and species interactions (Figure 1).



Figure 1: Different types of traps used for insect sampling in field studies across diverse environments. These traps vary in design and function, allowing the capture of insects with different behaviors and ecological niches. The combined use of multiple trapping methods improves the efficiency and representativeness of entomological surveys

Another relevant aspect discussed is the importance of specimen preservation. Properly preserved insects enable more precise identification and facilitate subsequent ecological and taxonomic analyses, especially in studies involving parasitoids and their hosts. The ability to maintain specimen integrity is crucial for understanding parasitoid-host relationships and for evaluating their potential in biological control programs. In this context, efficient trapping methods contribute not only to data collection but also to the quality and reliability of the scientific results obtained.

Overall, the findings emphasize that standardized, well-designed, and properly deployed traps are indispensable tools in entomological research. They support population monitoring, biodiversity assessments, and studies of ecological interactions, including those involving parasitoids of economic and sanitary importance. Moreover, these methods provide a solid foundation for applied research, particularly for the development and implementation of biological control strategies to manage pest populations in agricultural and urban environments (Marchiori, 2021a).

2. Gregarious Parasitoids of Muscoid Diptera Collected in Different Substrates in Itumbiara, Goiás

A total of 499 specimens of gregarious parasitoids were collected, belonging to three taxa: *Aphaereta* sp. (68.9%), *H. herberti* (12.4%), and *N. vitripennis* (18.6%). These results demonstrate the predominance of *Aphaereta* sp. among parasitoids associated with muscoid Diptera in the studied area. A total of 2,050 dipteran pupae were obtained from the substrates used as bait, with an overall parasitism prevalence of 1.2% (24/2,050). The highest number of parasitoids emerging from a single pupa was recorded for *H. herberti*, using *Peckia chrysostoma* (Wiedemann, 1830) (Diptera: Sarcophagidae) as host, with up to 60 individuals emerging from a single pupa.

Peckia chrysostoma showed the highest diversity of associated parasitoids, probably due to its larger pupal size, which may allow the development of several gregarious parasitoid individuals. In addition, human feces exposed in urban areas exhibited the greatest host diversity, indicating that substrate type plays an important role in host availability and parasitoid diversity (Table 4).

Table 4: Distribution of gregarious parasitoids associated with muscoid Diptera collected from different substrates in forest and urban areas of Itumbiara, Goiás, Brazil. The table shows host species, parasitoid taxa, number of parasitized pupae, and number of parasitoid individuals emerged

Area	Substrate	Number of pupae	Host species	Parasitoid species	Frequency
Forest	Chicken	01	<i>Peckia chrysostoma</i>	<i>Nasonia vitripennis</i>	10
	Fish	11	<i>Peckia chrysostoma</i>	<i>Aphaereta</i> sp.	344
Urban	Bovine liver	04	<i>Musca domestica</i>	<i>Nasonia vitripennis</i>	34
	Bovine rumen	01	<i>Chrysomya albiceps</i>	<i>Nasonia vitripennis</i>	09
	Chicken	02	<i>Peckia chrysostoma</i>	<i>Hemencyrtus herberti</i>	62
	Fish	01	<i>Peckia chrysostoma</i>	<i>Nasonia vitripennis</i>	16
	Human feces	02	<i>Musca domestica</i>	<i>Hemencyrtus herberti</i>	24
Total		24	-	-	499

The distribution of parasitoid individuals differed significantly among parasitoid taxa ($\chi^2 = 287.55$; $df = 2$; $p < 0.0001$), with *Aphaereta* sp. being the dominant taxon. Parasitoid abundance also differed between the forest and urban areas ($\chi^2 = 87.54$; $df = 1$; $p < 0.0001$), with more individuals obtained in the forest area. In addition, parasitoid abundance varied significantly among substrates ($\chi^2 = 869.71$; $df = 4$; $p < 0.0001$), mainly due to the high number of *Aphaereta* sp. individuals obtained from fish bait.

Comparisons with previous studies revealed variability in parasitism rates depending on environmental conditions, host density, and substrate type. Among the substrates tested, human feces and chicken bait attracted a greater diversity of dipteran hosts and supported a wider range of parasitoid taxa. However, the available table provides the number of parasitized

pupae and emerged parasitoid individuals, not the total number of pupae collected by each substrate; therefore, parasitism rates by substrate were not recalculated.

From an applied perspective, the results highlight the importance of gregarious parasitoids as natural biological control agents of synanthropic flies of medical and veterinary importance. Species such as *C. albiceps* and *M. domestica* were identified as relevant hosts because of their epidemiological importance. Furthermore, *Aphaereta* sp. showed a high degree of host association with *P. chrysostoma*. Overall, the study demonstrates that parasitoid diversity and abundance are influenced by host species, substrate type, and environmental conditions, highlighting the ecological importance of these organisms in synanthropic environments (Figure 2) (Marchiori *et al.*, 2002a).

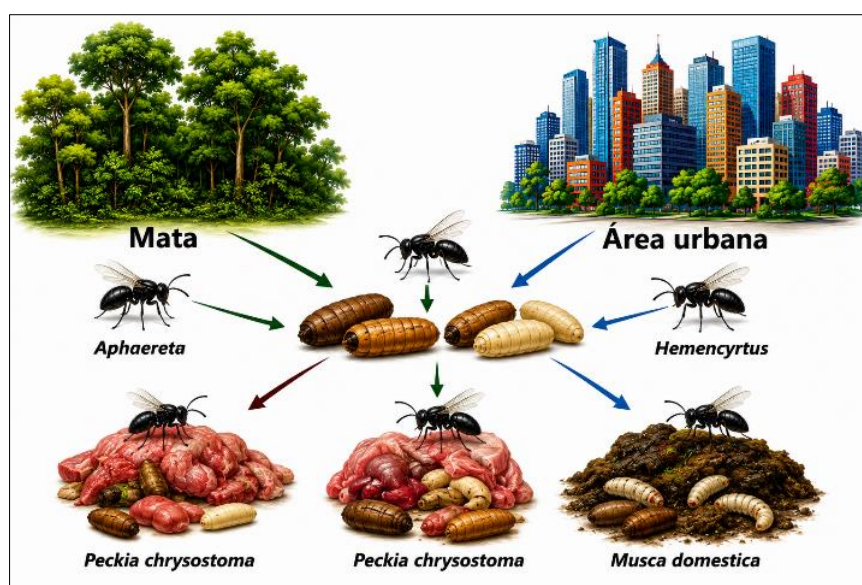


Figure 2: Gregarious parasitoids associated with muscoid Diptera in different substrates in Itumbiara, Goiás. Interactions between hosts and parasitoids vary according to environment and type of organic substrate. These relationships highlight the ecological importance of parasitoids in regulating synanthropic fly populations

3. *B. Podagrica*: First Record in Guinea Pig Carcasses in Northern Peru

A total of 91 specimens of *B. podagrica* were collected, in addition to 744 adults of Calliphoridae and 493 adults of Sarcophagidae associated with guinea pig

carcasses. As shown in Table 5, *B. podagrica* was present until the tenth day of the experiment, when the carcasses began the skeletonization stage. At this point, the Diptera present was mainly in larval and pupal stages, which persisted until the twelfth day. Therefore, the

presence of *B. podagrica* in guinea pig carcasses is attributed to its role as a parasitoid of Diptera larvae

belonging to the families Calliphoridae and Sarcophagidae (Table 5).

Table 5: Presence of *B. podagrica* and Diptera in guinea pig carcasses. Temporal distribution of taxa throughout decomposition stages. Occurrence of immature and adult stages during 12 days of observation

T Taxonomic group	1	2	3	4	5	6	7	8	9	10	11	12
<i>Brachymeria podagrica</i>	A	A	A	A	A	A	A	A	A	A	A	A
Calliphoridae	H-A	H-L-A	L-A	L-A	L-A	L-A	L-P-A	L-P-A	L-P	L-P	L-P	L-P
Sarcophagidae	L-A	L-A	L-A	L-A	L-A	L-A	L-P-A	L-P-A	L-P-A	L-P-A	L-P	L-P

Note: Presence of *B. podagrica* and Diptera in guinea pig carcasses in Piura, Peru. Decomposition stages: F=fresh, Cr=chromatic, Hi=swollen, D. Ac=active decomposition, D. Av=advanced decomposition, skeletonization. Insect stages: H=egg, L=larva, P=pupa, A=adult

Brachymeria podagrica was recorded in parts of the carcasses with a higher presence of Diptera, such as natural body orifices and areas where the fur was in contact with the soil. These sites are preferred by flies for oviposition and larviposition, as previously reported. This observation is consistent with other studies that

describe this hymenopteran species as a parasitoid of both Calliphoridae and Sarcophagidae. Females of *B. podagrica* oviposit on late-instar larvae that survive until pupation. Oviposition is stimulated by the presence of fluids released from decomposing tissues colonized by flies (Figure 3).

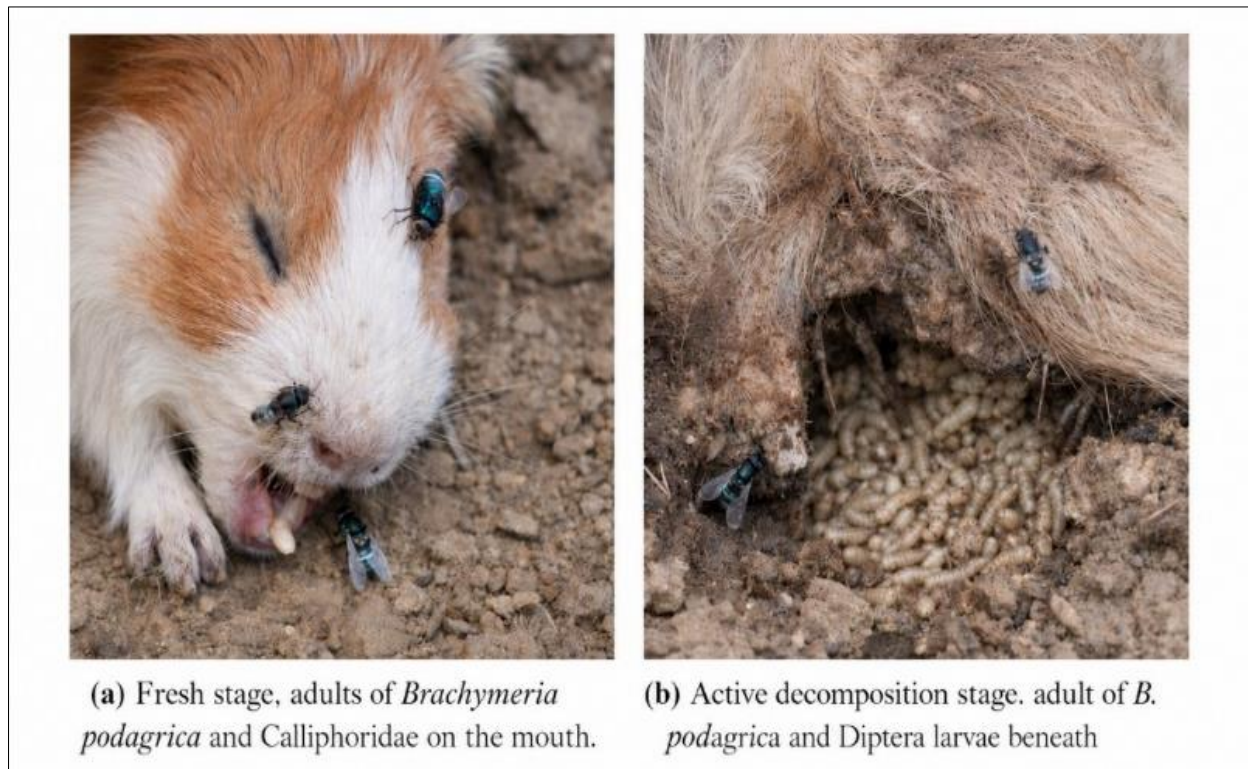


Figure 3: Guinea pig carcasses. (a). Fresh stage, adults of *B. podagrica* and Calliphoridae on the mouth. (b). Active decomposition stage, adult of *B. podagrica* and Diptera larvae beneath the carcass

Based on the collection of *B. podagrica* specimens parasitizing late-instar larvae of Calliphoridae and Sarcophagidae found in guinea pig carcasses in Piura, Peru, this represents the first record of this hymenopteran species in the northern region of the country. Previous records reported this species in baited traps with decomposing meat and in carcasses in other regions of South America. Regarding abundance, *B.*

podagrica was mainly observed during the active decomposition stage, when the highest number of Diptera larvae preparing for pupation was recorded. These larvae were distributed in the soil beneath and around the carcasses. During skeletonization, fly pupae with parasitoid emergence holes were observed, consistent with damage caused by species of *B. podagrica* (Figure 4).



Figure 4: Chalcididae parasitoid can be identified by their small body size, reduced wing venation, and enlarged hind femora. Although most species are minute, *B. podagrica* is relatively large for the group, reaching up to 5 mm in length

Studies on the biology, life cycles, and ecology of carrion-associated insects are still in early stages of development. Therefore, increasing taxonomic knowledge of insect groups involved in the decomposition of organic matter is essential, particularly for applications in forensic entomology (Andrade-Herrera and Marchiori, 2019).

4. Occurrence of *Stomoxys Calcitrans* L. 1758 (Diptera: Muscidae) in Cattle Feces Collected on Pasture in Goiânia, Goiás, Brazil

Dipterans are vectors of pathogens such as viruses, bacteria, protozoan cysts, and parasitic worms. They can cause diseases in animals and represent a nuisance to humans in both urban and rural environments. In confinement and semi-confinement systems, accumulated manure constitutes an excellent substrate for the development of several fly species in rural environments. Although excrement is periodically removed from breeding sites, its persistence in manure accumulation areas favors the development of a highly diverse entomofauna (Figure 5) (D'Amico *et al.*, 2026).



Figure 5: Life stages and clinical manifestations associated with *Dermatobia hominis* (Linnaeus Jr., 1781) (Diptera: Oestridae). Eggs attached to the abdomen of a carrier fly, furuncular skin lesions caused by larval infestation, and second-instar larva in dorsal and ventral views. Source: Adapted from Pereira (University of São Paulo), Ragi (2021), and original microscopic observations and <https://doi.org/10.3390/tropicalmed11050110>

The fly *S. calcitrans*, commonly known as the “stable fly,” is a hematophagous species that attacks several hosts, including cattle, goats, sheep, horses, dogs, and even humans. This species is one of the most

important dipterans affecting livestock due to the economic losses it causes and its role as a transmitter and potential vector of various diseases in domestic animals (Figure 6).

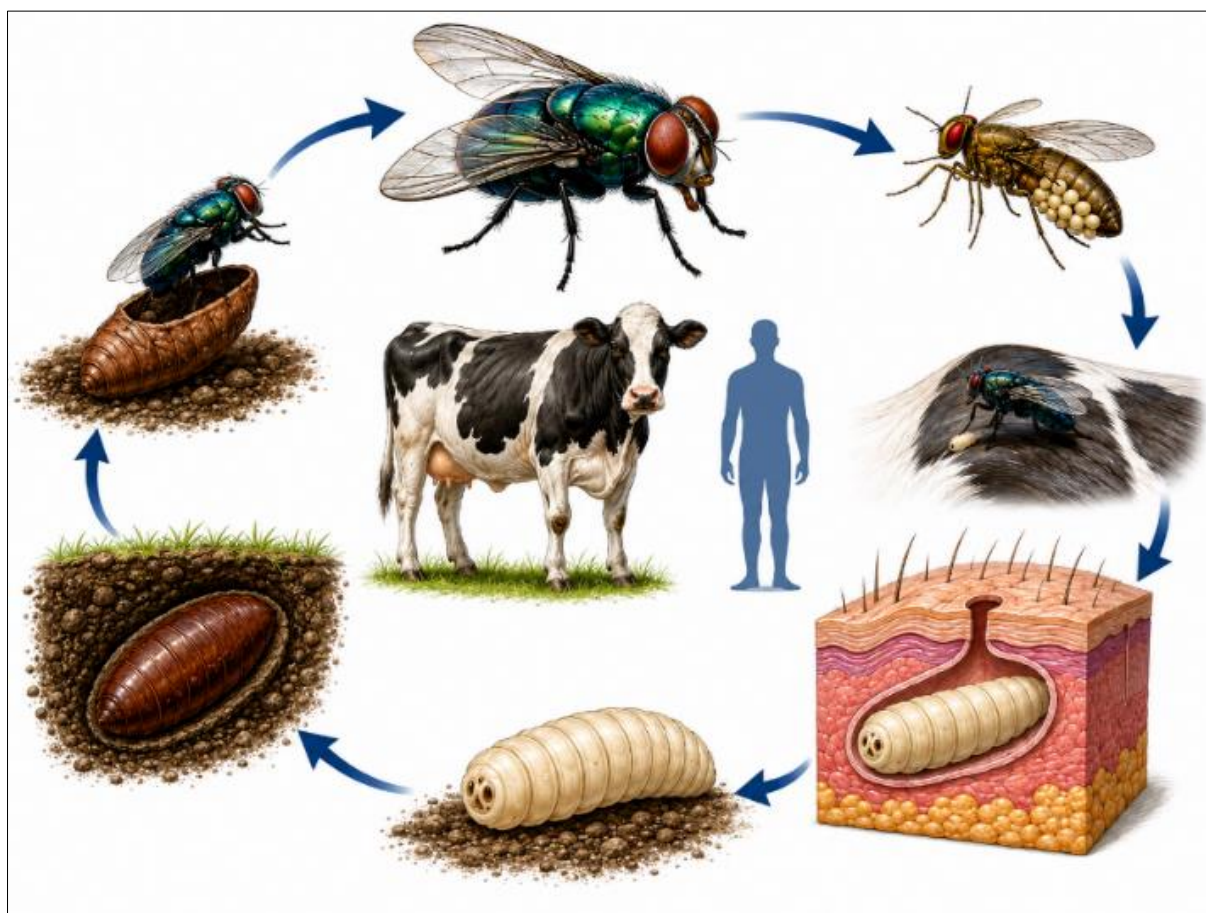


Figure 6: Life cycle of *Dermatobia hominis* (Linnaeus Jr., 1781) (Diptera: Oestridae). The cycle includes the adult fly, deposition of eggs on a carrier insect, transfer of larvae to a mammalian host, subcutaneous larval development, emergence of the mature larva, pupation in the soil, and emergence of a new adult. Cattle and other mammals, including humans, may serve as hosts. The figure highlights the biological stages involved in furuncular myiasis and the veterinary importance of this parasite

A total of 628 dipteran pupae were obtained from bovine feces, from which eight specimens of *S. calcitrans* emerged from eight pupae collected in pastures. This species is preferentially associated with feces found in stables, corrals, and similar environments. The sampled pastures were located approximately 500, 100, and 10 meters from stables and corrals. The feces were deposited in areas inaccessible to animals. The presence of sheep near the stables was observed only once during the study.

The percentage of individuals collected was 1.3% (8/628) low due to its high degree of synanthropy. *Stomoxys calcitrans* have most animals and humans as hosts, and their bite is painful. Food remains and vinasse, a byproduct of the sugarcane industry, attract and stimulate posture, and can be formed in straw and harvest residues, which remain for some time in the field,

especially if these materials are fermented or moistened with urine and animal feces (Marchiori, 2021b).

5. Diptera Collected in the Mountain of Caldas Novas in Goiás, Brazil

A total of 2.946 flies were collected in the Serra de Caldas Novas region, Goiás, Brazil. Of these, 1.255 individuals were recorded at 740 m and 1.691 at 1.000 m, indicating greater abundance at the higher altitude (Table 6). The original analysis of variance reported no significant preference between altitudes ($F = 0.16$; $p = 0.6949$), suggesting that altitude did not significantly influence the distribution of synanthropic Diptera under the analytical approach used in the original study. However, the altitude of 1.000 m showed higher species richness, with 13 species recorded, whereas eight species were recorded at 740 m (Table 6).

Table 6: Synanthropic Diptera collected at the altitudes of 740 m and 1.000 m from different substrates in the Serra de Caldas Novas Park, Goiás, Brazil

Altitude / Family / Species	Feces	Liver	Chicken	Fruit	Fish	Total
Altitude of 740 meters:						
Calliphoridae						
<i>Chrysomya albiceps</i>	10	30	0	0	4	44
<i>Chrysomya megacephala</i>	1	0	0	0	0	1
Drosophilidae						
<i>Drosophila</i> sp.	0	0	0	141	0	141
Fanniidae						
<i>Fannia pusio</i>	236	0	0	0	0	236
Muscidae						
<i>Brontaea</i> sp.	1	0	0	0	0	1
<i>Musca domestica</i>	2	0	0	0	0	2
Sarcophagidae						
<i>Oxysarcodexia thornax</i>	169	166	0	0	197	532
<i>Peckia chrysostoma</i>	138	0	160	0	0	298
Total	557	196	160	141	201	1.255
Altitude of 1000 meters:						
Calliphoridae						
<i>Chrysomya albiceps</i>	92	0	82	0	51	225
<i>Chrysomya megacephala</i>	0	0	58	0	0	58
Drosophilidae						
<i>Drosophila</i> sp.	0	0	0	66	0	66
Fanniidae						
<i>Fannia pusio</i>	71	67	0	0	11	149
Muscidae						
<i>Brontaea</i> sp.	10	0	0	0	0	10
<i>Musca domestica</i>	130	0	61	0	0	191
<i>Ophyra</i> sp.	74	25	23	0	0	122
<i>Synthesiomyia nudiseta</i>	57	0	0	1	0	58
Phoridae						
<i>Megaselia scalaris</i>	0	114	0	0	0	114
Sarcophagidae						
<i>Oxysarcodexia thornax</i>	0	17	101	0	16	134
<i>Peckia chrysostoma</i>	117	99	62	0	189	467
<i>Sarcodexia lambens</i>	50	0	0	10	36	96
<i>Squamatoides trivittatus</i>	0	0	0	0	1	1
Total	601	322	387	77	304	1.691
Total general	1.158	518	547	218	505	2.946

At 740 m, *O. thornax* was the most frequent species, representing 42.4% of the individuals collected at this altitude. This species was mainly associated with feces, bovine liver, and fish, reinforcing its synanthropic behavior and adaptability to different organic substrates. At 1.000 m, *P. chrysostoma* was the most frequent species, accounting for 27.6% of the individuals collected at this altitude. This species was recorded in feces, liver, chicken, and fish, indicating broad substrate use.

The original study reported no significant differences in substrate preference at either altitude (740 m: $F = 0.40$; $p = 0.9314$; 1.000 m: $F = 0.77$; $p = 0.7179$), indicating that substrate availability did not strongly influence species distribution according to the original ANOVA. As a complementary count-based analysis,

chi-square tests were applied to the data presented in Table 6. These tests indicated significant differences in total abundance between altitudes ($\chi^2 = 64.53$; $df = 1$; $p < 0.0001$), in substrate distribution between altitudes ($\chi^2 = 104.07$; $df = 4$; $p < 0.0001$), and in species distribution between altitudes ($\chi^2 = 1040.58$; $df = 12$; $p < 0.0001$). These results suggest that, although the original ANOVA did not detect preference, the distribution of individuals among altitudes, substrates, and species was not homogeneous when evaluated using frequency data.

Overall, the findings indicate that synanthropic Diptera were able to exploit multiple substrates at both altitudes, demonstrating ecological plasticity in the Serra de Caldas Novas region. The greater abundance and richness recorded at 1.000 m may reflect local environmental conditions, substrate availability,

microclimate, and species-specific responses to altitude. These results reinforce the importance of evaluating both environmental factors and substrate use when studying

the distribution of Diptera in natural and semi-natural areas (Figure 7) (Marchiori, 2019).

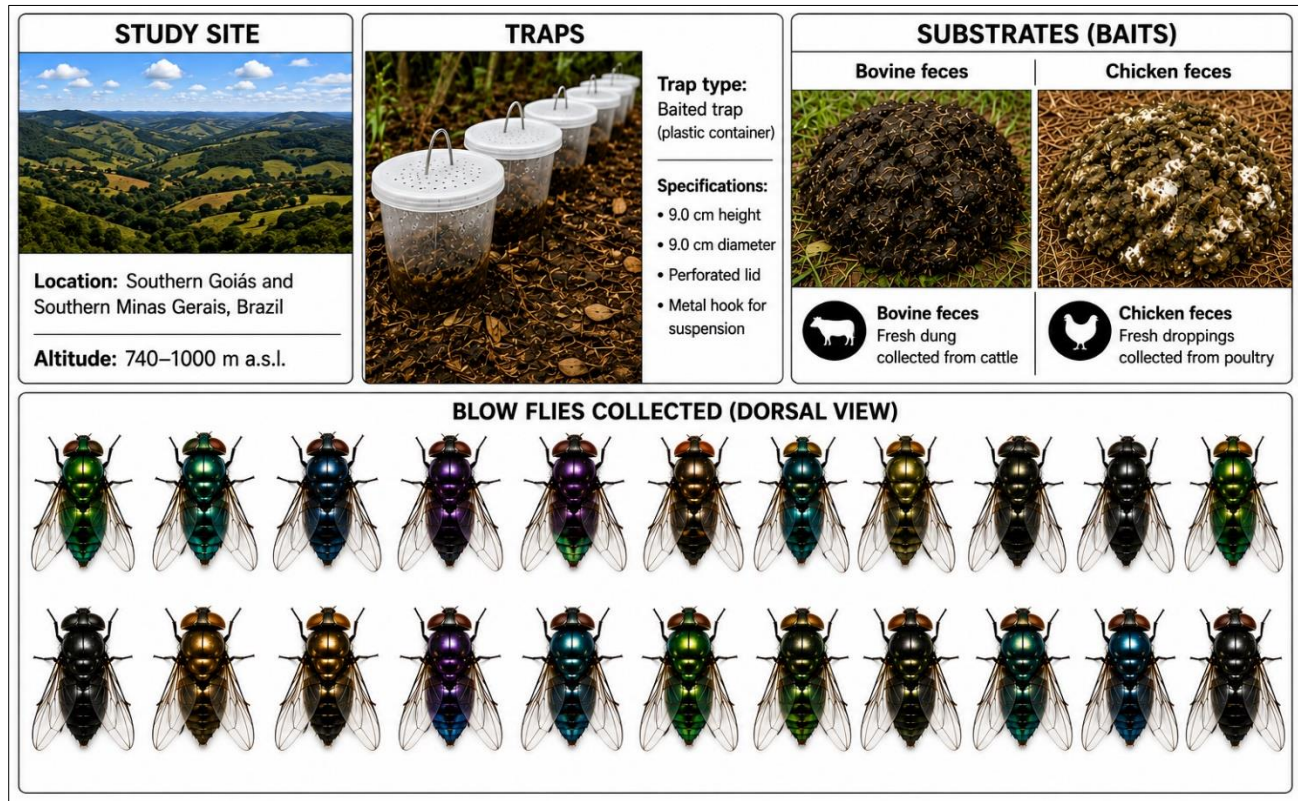


Figure 7: Synanthropic Diptera collected at 740 m and 1,000 m from different organic substrates in Serra de Caldas Novas State Park, Goiás, Brazil. The figure represents the distribution of Diptera among feces, bovine liver, chicken, fruit, and fish baits, highlighting differences in abundance and richness between altitudes and substrates

6. Parasitoids of the Subfamily Eucoilinae (Hymenoptera: Figitidae) Collected in a Remnant of Cerrado Forest in Itumbiara, Goiás, Brazil

A total of 20 specimens of Eucoilinae were collected in a remnant of Cerrado Forest using Malaise traps. The most abundant taxon was *Steleucoela* sp., with 10 individuals, representing 50.0% of the total collected specimens. The remaining taxa occurred at lower

frequencies: *Aganaspis pelleranoi* (Brèthes, 1924) (Hymenoptera: Figitidae) (1 individual; 5.0%), *Dettmeria* sp. (2; 10.0%), *Dicerataspis* sp. (1; 5.0%), *Odonteucoila* sp. (1; 5.0%), *P. egeria* (2; 10.0%), *Trybliographa* sp. (1; 5.0%), and *Zaeucoila unicarinata* (2; 10.0%). These results indicate a diverse but unevenly distributed Eucoilinae assemblage in the Cerrado fragment (Table 7).

Table 7: Eucoilinae parasitoids collected in a remnant of Cerrado Forest in Itumbiara, Goiás, Brazil. Taxonomic composition and relative frequency of specimens obtained using Malaise traps in a native vegetation fragment

Taxonomic group	Substrates	Hosts	Frequency	Percentage
<i>Aganaspis pelleranoi</i>	Decomposing fruits	Tephritidae larvae	1	5.0
<i>Dettmeria</i> sp.	Organic matter	Diptera pupae	2	10.0
<i>Dicerataspis</i> sp.	Forest litter	Diptera larvae	1	5.0
<i>Odonteucoila</i> sp.	Decomposing feces	Muscoid Diptera	1	5.0
<i>Paraganaspis egeria</i>	Bovine feces	Sarcophagidae pupae	2	10.0
<i>Steleucoela</i> sp.	Forest organic matter	Diptera pupae	10	50.0
<i>Trybliographa</i> sp.	Soil/litter	Diptera larvae	1	5.0
<i>Zaeucoila unicarinata</i>	Decomposing substrates	Cyclorrhaphous Diptera	2	10.0
Total			20	100

The high frequency of *Steleucoela* sp. may be related to ecological adaptability, local host availability, or a closer association with dipteran hosts present in the sampled area. In contrast, the low frequency of the other

taxa suggests that several Eucoilinae occurred as rare components of the community. The chi-square analysis showed a significant difference in abundance among the recorded taxa ($\chi^2 = 26.40$; $df = 7$; $p = 0.0004$), indicating

that the distribution of Eucoilinae was not uniform in the Cerrado fragment. Eucoilinae parasitoids are commonly associated with larvae of Cyclorrhapha Diptera, and their

presence in environments with decomposing organic matter reflects the availability of suitable hosts (Figure 8).



Figure 8: Representative adult specimen of *Steleucoela* sp., a parasitoid of Diptera associated with decomposing organic substrates. The image illustrates the morphological characteristics of Eucoilinae parasitoids and their relevance as natural enemies in Cerrado environments

Therefore, the diversity observed in this study reinforces the ecological importance of Cerrado remnants as reservoirs of parasitoid biodiversity. The record of *A. pelleranoi*, a species frequently associated with fruit-infesting Diptera, highlights its potential role as a natural biological control agent. Similarly, *P. egeria* and *Zaeucoila unicarinata* Ashmead, 1903 (Hymenoptera: Figitidae), are associated with Diptera larvae and may contribute to the natural regulation of fly populations. Overall, the results demonstrate that even small forest remnants can sustain representative assemblages of Eucoilinae species, contributing to ecological balance and providing useful information for biological control studies (Marchiori *et al.*, 2001a).

7. Agricultural Pests of Some Parasitoids Collected in Brazil

A total of 389 parasitoids were obtained from different agricultural pests collected in southern Goiás and southern Minas Gerais, Brazil. The hosts included *Lonomia* sp. (Lepidoptera: Saturniidae), *Leptoglossus zonatus* (Dallas, 1852) (Heteroptera: Coreidae), *Tuta absoluta* (Meyrick, 1917) (Lepidoptera: Gelechiidae), and *Z. indianus*. The highest abundance was recorded from pupae of *Z. indianus*, with 288 parasitoids, followed by eggs of *L. zonatus* with 54 parasitoids, pupae of *T. absoluta* with 35 parasitoids, and *Lonomia* sp. with 12 parasitoids.

From *Lonomia* sp., 12 parasitoids were obtained, including nine specimens of *Anastatus* sp. (Hymenoptera: Eupelmidae) and three specimens of *Aprostocetus* sp. (Hymenoptera: Eulophidae). *Anastatus*

sp. was the most frequent parasitoid associated with this host, representing 75.0% of the parasitoids recorded for *Lonomia* sp. This result indicates a higher occurrence of this genus in association with lepidopteran hosts under the sampled conditions.

In eggs of *L. zonatus*, 54 parasitoids emerged from 113 eggs, corresponding to a parasitism rate of 47.8%. The most abundant species was *G. gallardoi*, with 41 specimens, followed by *Trissolcus* sp. with six specimens, *Anastatus* sp. with five specimens, and *Brasema* sp. with two specimens. This high parasitism rate demonstrates the importance of egg parasitoids in the natural regulation of hemipteran pests.

Regarding *T. absoluta*, 35 parasitoids emerged from pupae, including *Bracon* sp. with 21 specimens, *Conura* sp. with 13 specimens, and *Earinus* sp. with one specimen. *Bracon* sp. (Hymenoptera: Braconidae) was the most frequent parasitoid in this host, suggesting greater efficiency in host location or better adaptation to greenhouse or cultivated conditions.

From pupae of *Z. indianus*, 288 parasitoids were obtained, including 285 specimens of *P. vindemmiae* and three specimens of *Leptopilina boulandi* (Barbotin, Carton & Kelner-Pillaut, 1979) (Hymenoptera: Figitidae). *P. vindemmiae* was the dominant species, representing 98.9% of the parasitoids associated with this host and 73.3% of all parasitoids recorded in the dataset. This predominance may be related to its generalist behavior and high capacity to exploit dipteran hosts (Table 8).

Table 8: Parasitoids obtained from insect pests in southern Goiás and southern Minas Gerais, Brazil. Distribution of parasitoid species associated with different agricultural pests and host developmental stages

Insect pest	Parasitoid species	Family	Number of specimens	Percentage
<i>Lonomia</i> sp.	<i>Anastatus</i> sp.	Eupelmidae	9	2.3
	<i>Aprostocetus</i> sp.	Eulophidae	3	0.8
<i>Leptoglossus zonatus</i>	<i>Anastatus</i> sp.	Eupelmidae	5	1.3
	<i>Brasema</i> sp.		2	0.5
	<i>Gryon gallardoi</i>	Scelionidae	41	10.5
	<i>Trissolcus</i> sp.		6	1.5
<i>Tuta absoluta</i>	<i>Bracon</i> sp.	Braconidae	21	5.4
	<i>Conura</i> sp.	Chalcididae	13	3.3
	<i>Earinus</i> sp.	Braconidae	1	0.3
<i>Zaprionus indianus</i>	<i>Leptopilina bouhardi</i>	Figitidae	3	0.8
	<i>Pachycrepoideus vindemmiae</i>	Pteromalidae	285	73.3
Total			389	100

The percentages presented in Table 8 refer to the relative frequency of each parasitoid species in relation to the total number of parasitoids collected (389 specimens). Therefore, these values should not be confused with parasitism rates, which can only be calculated when the total number of hosts or eggs is available.

Complementary chi-square analyses were applied to the quantitative data. The distribution of parasitoids among the four pest hosts differed significantly ($\chi^2 = 507.96$; $df = 3$; $p < 0.0001$), with *Z. indianus* showing the highest number of associated parasitoids. The abundance of parasitoid species also differed significantly across the dataset ($\chi^2 = 1977.30$; $df = 10$; $p < 0.0001$), mainly due to the predominance of *P. vindemmiae*. Within specific hosts, significant

differences were observed for *L. zonatus* ($\chi^2 = 75.33$; $df = 3$; $p < 0.0001$), *T. absoluta* ($\chi^2 = 17.37$; $df = 2$; $p = 0.0002$), and *Z. indianus* ($\chi^2 = 276.13$; $df = 1$; $p < 0.0001$). No significant difference was observed between the two parasitoids associated with *Lonomia* sp. ($\chi^2 = 3.00$; $df = 1$; $p = 0.0833$).

Overall, the results demonstrate that parasitoid abundance varied markedly among agricultural pests and parasitoid taxa. The dominance of *P. vindemmiae* in *Z. indianus* and the high parasitism of *L. zonatus* eggs by *G. gallardoi* highlight the ecological relevance of these parasitoids as natural enemies. These records reinforce the importance of parasitoids in the regulation of agricultural pests and indicate their potential contribution to biological control programs (Figure 9) (Marchiori, 2013).

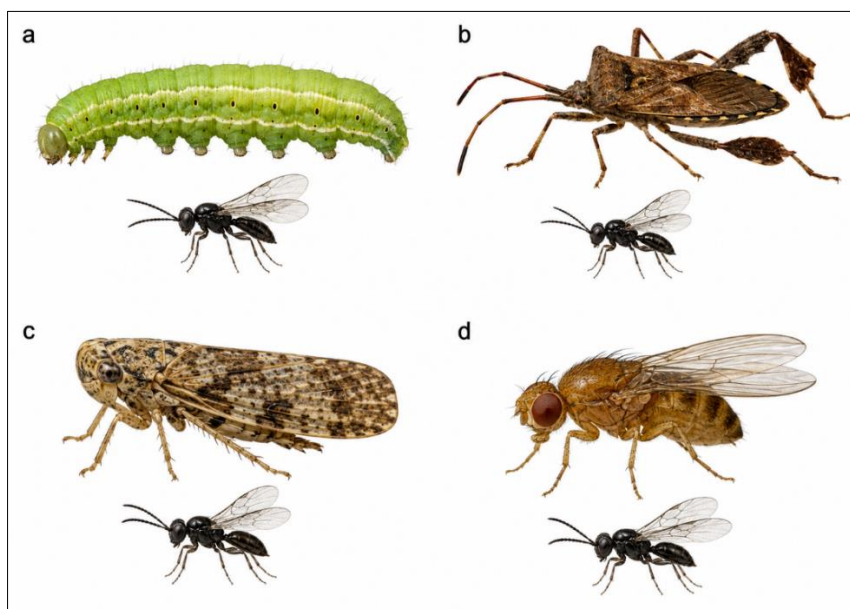


Figure 9: Main agricultural pest insects and associated parasitoids (Black) collected in southern Goiás and southern Minas Gerais, Brazil. (a) Lepidopteran larva and associated hymenopterous parasitoid; (b) hemipteran pest insect and associated hymenopterous parasitoid; (c) leafhopper and associated hymenopterous parasitoid; (d) fruit fly and associated hymenopterous parasitoid. The pest insects are shown as the principal hosts, whereas the parasitoids are represented in black, highlighting their ecological importance in the natural regulation of agricultural pest populations

Overall, the results demonstrate that different groups of parasitoids are associated with specific host insects and developmental stages (eggs, larvae, and pupae). The high parasitism rates observed, especially for *L. zonatus* and *Z. indianus*, reinforce the ecological importance of these parasitoids as natural biological control agents in agricultural systems (Marchiori, 2013).

A total of 151 engorged females of *Rhipicephalus microplus* (Canestrini 1888) (Acari: Ixodidae) were collected from cattle, from which one

specimen of *Chrysoperla externa* (Hagen) (Neuroptera: Chrysopidae) emerged. The observed occurrence rate was 0.6%, indicating a low frequency of association under the evaluated conditions. The emergence of *C. externa* from ticks suggests a possible interaction between this predator and *R. microplus*, likely associated with oviposition on engorged females. One of the collected ticks probably carried a pedicellate egg of *C. externa*, which later developed into a larva and completed its life cycle (Figure 10).



Figure 10: Association between *C. externa* and engorged females of *R. microplus* collected from cattle in Goiás, Brazil. The figure shows an engorged tick, the larval stage, and the emerged adult of *C. externa*. This record suggests a possible ecological interaction with potential importance in the biological control of the cattle tick

The low observed occurrence percentage may be related to the low degree of synanthropy of *C. externa*, as well as to ecological and behavioral factors that limit its interactions with tick populations. Despite this, the record is significant, as it demonstrates a little-documented association between Chrysopidae and ticks. Larvae of *C. externa* are known to be voracious predators, capable of consuming large numbers of insect eggs, such as *Anagasta* sp., *Diatraea* sp., and *Sitotroga* sp. This predatory capacity reinforces the potential of this species as a biological control agent in different agroecosystems.

Additionally, *C. externa* has a wide geographic distribution across the Neotropical region and is commonly found in agricultural environments, especially on crops such as tomato. Its ecological versatility, high reproductive potential, and tolerance to certain insecticides make it an important natural enemy in integrated pest management programs. The results of this study highlight a novel ecological interaction and reinforce the importance of further investigations on the role of generalist predators in the biological control of ectoparasites such as ticks (Marchiori, 2021c).

8. Eucilinae collected in Lavras, Minas Gerais, Brazil

A total of 34 individuals of Eucilinae (Hymenoptera: Figitidae) were collected using a Malaise trap in Lavras, Minas Gerais, Brazil. Among the recorded taxa, *Steleucoela* sp. was the most abundant genus, with 14 individuals, representing 41.2% of the total collected specimens. *Aganaspis pelleranoi* was the second most frequent taxon, with 8 individuals (23.5%). *Dettmeria* sp. represented 11.8%, whereas *Dicerataspis* sp., *K. nigra*, *P. egeria*, *Odonteucoila* sp., *Trybliographa* sp., and *Zaeucoila* sp. occurred at lower frequencies. The lowest frequency was observed for *Odonteucoila* sp., with one individual (2.9%).

The occurrence of *A. pelleranoi* is noteworthy because this species is widely distributed in Brazil and is commonly associated with larvae of frugivorous Diptera. Its presence reinforces its ecological importance as a parasitoid of economically relevant hosts. Species of *Zaeucoila* sp., although less abundant in this sample, are also known to parasitize immature stages of Cyclorrhapha Diptera and are restricted to the Neotropical region. The low number of specimens collected may be related to the limited sampling area, host availability, and vegetation structure in the forested environment (Table 9).

Table 9: Eucoilinae parasitoids collected in Lavras, Minas Gerais, Brazil. Number of individuals and percentage distribution of each genus obtained using Malaise traps. Relative frequency of parasitoid genera associated with Diptera hosts in a Cerrado-related environment

Taxonomic group	Substrates	Hosts	Frequency	Percentage
<i>Aganaspis pelleranoi</i>	Decomposing fruits	Tephritidae larvae	8	23.5
<i>Dettmeria</i> sp.	Organic matter	Diptera pupae	4	11.8
<i>Kleidotoma nigra</i>	Bovine feces	Diptera Muscidae pupae	2	5.9
<i>Odonteucoila</i> sp.	Decomposing feces	Muscoid Diptera	1	2.9
<i>Paraganaspis egeria</i>	Bovine feces	Sarcophagidae pupae	3	8.8
<i>Steleucoela</i> sp.	Forest organic matter	Diptera pupae	14	41.2
<i>Zaeucoila</i> sp.	Decomposing substrates	Cyclorrhaphous Diptera	2	5.9
Total			34	100

The chi-square analysis showed a significant difference in abundance among Eucoilinae taxa (chi-square = 26.53; df = 6; $p = 0.0002$), indicating that the distribution of individuals was not uniform. The predominance of *Steleucoela* sp. suggests that this species was better represented under the sampled conditions. The *Dettmeria* sp. has been reported to parasitize dipteran hosts such as *Euxesta* sp., with relatively high parasitism rates in agricultural systems, suggesting its potential importance in biological control. In addition, *K. nigra* and *P. egeria* have been associated with Diptera developing in decomposing organic substrates, reinforcing the role of Eucoilinae in

regulating populations of synanthropic flies (Figure 11) (Marchiori *et al.*, 2001a).

Although only one species of *Steleucoela* sp. has been reported to date, and its biology remains poorly known, its high abundance in this study suggests a strong adaptation to the local environmental conditions or to the availability of suitable hosts. Overall, the results highlight the diversity of Eucoilinae in the studied area and reinforce the importance of these parasitoids as natural regulators of dipteran populations, especially in environments associated with organic matter and agricultural systems (Siva *et al.*, 2003).



Figure 11: Specimen of Eucoilinae Endoparasitoids of cyclorrhaphous dipteran larvae, acting primarily as biological control agents of fruit flies, such as those belonging to the Drosophilidae, Lonchaeidae, and Tephritidae families. Its occurrence highlights the ecological importance of figitid parasitoids in the natural regulation of Diptera populations

9. Cynipoidea (Hymenoptera) Associated with Bovine Feces and Collected in Native Forest and Pasture Areas in Goiás, Brazil

A total of 5.786 dipterans were collected from bovine feces. The highest abundances were recorded for *C. paraescita*, with 2.362 individuals, and *S. occidua*, with 2.006 individuals. Other species, such as *B. debilis*, *B. quadristigma*, and Sepsidae species, were also recorded, demonstrating the diversity of Diptera associated with cattle dung.

Regarding parasitoids, 105 individuals were obtained from the dataset summarized in Table 10, with 104 belonging to Eucoilinae and one to another Figitinae taxon. Considering the total number of dipteran pupae presented in Table 10, the overall parasitism rate was 3.6%. The most abundant parasitoid was *Triplasta atrocoxalis* (Ashmead, 1865) (Hymenoptera: Figitidae: Eucoilinae), representing 68.6% of the parasitoids, followed by *P. egeria* with 30.5%. These results indicate that Eucoilinae were the dominant parasitoids in bovine feces under the studied conditions.

Table 10: Occurrence of parasitoids and their respective hosts in pasture areas in Itumbiara, Goiás. Number of pupae and parasitism percentages for each parasitoid species associated with Diptera. Distribution of Figitidae according to host species collected from bovine feces

Host species	No. pupae	<i>Triplasta</i> (N)	%	<i>Paraganaspis egeria</i> (N)	%	<i>Neralsia splendens</i> (N)
<i>Brontaea quadristigma</i>	720	0	0	1	0.1	0
<i>Palaeosepsis</i> spp.	147	67	45.6	0	0	0
<i>Sarcophagula occidua</i>	2.006	4	0.2	31	1.5	1
<i>Sphaeroceridae</i> sp.	27	1	3.7	0	00	0
Total	2.900	72	2.5	32	1.1	1

Triplasta sp. showed a strong association with pupae of *Palaeosepsis* spp., whereas *P. egeria* was mainly associated with *S. occidua*. The occurrence of *N. splendens* was rare and was recorded only once in the pupae of *S. occidua*. Complementary chi-square analyses showed significant variation in parasitism among host

species ($\chi^2 = 785.77$; $df = 3$; $p < 0.0001$), in abundance among parasitoid taxa ($\chi^2 = 72.40$; $df = 2$; $p < 0.0001$), and in host-parasitoid association ($\chi^2 = 88.60$; $df = 6$; $p < 0.0001$). These results indicate that parasitoid occurrence was not randomly distributed among host species (Figure 12) (van Nort, 2026).



Figure 12: The image emphasizes structural characteristics commonly used for taxonomic identification in parasitoid Cynipoidea Hymenoptera. These features are essential for distinguishing species and understanding their ecological roles. Source: Web author Simon van Noort (Iziko South African Museum)

The results also demonstrate that bovine feces with approximately eight days of exposure provide suitable conditions for the development of parasitoids, particularly for species of the genus *Triplasta* sp. The high parasitism observed for this *Palaeosepsis* spp. reinforces its importance as a natural enemy of dung-breeding flies. In relation to habitat, 414 parasitoid

individuals were collected using yellow pan traps, with 222 individuals in pasture areas and 192 in native forest. Although abundance was numerically higher in pastures, the difference between environments was not significant when only total abundance was compared ($\chi^2 = 2.17$; $df = 1$; $p = 0.1404$) (Table 11).

Table 11: Parasitoids collected in pasture and forest areas. Number of individuals and percentage distribution per environment. Figitidae according to habitat

Taxon	Pasture N	Pasture %	Forest N	Forest %	Total N	Total %
<i>Acanthaegilips</i> sp.	4	1.8	0	0.0	4	1.0
<i>Aganaspis pelleranoi</i>	2	0.9	0	0.0	2	0.5
<i>Agrostocynips</i> sp.	1	0.5	0	0.0	1	0.2
<i>Dieucoila</i> sp.	1	0.5	1	0.5	2	0.5
<i>Eucoilinae</i> sp.	183	82.4	157	81.8	340	82.1
<i>Neralsia splendens</i>	1	0.5	0	0.0	1	0.2
<i>Odonteucoila</i> sp.	1	0.5	9	4.7	10	2.4
<i>Paraganaspis egeria</i>	5	2.3	4	2.1	9	2.2
<i>Triplasta</i> sp.	6	2.7	1	0.5	7	1.7
<i>Zaeucoila</i> sp.	18	8.1	20	10.4	38	9.2
Total	222	100	192	100	414	100

The number of taxa was higher in pasture areas (10 taxa) than in native forest (6 taxa), suggesting that open environments may support greater parasitoid diversity. However, native vegetation also plays an important role as a reservoir of natural enemies, contributing to the maintenance of parasitoid populations in agroecosystems. Among the collected taxa, Eucilinae was the most abundant group (82.1%), followed by *Zaeucoila* sp. (9.2%). The distribution of taxa differed significantly between pasture and forest areas ($\chi^2 = 18.10$; $df = 9$; $p = 0.0341$), indicating that habitat type influenced taxonomic composition even though total

abundance did not differ significantly between environments.

Overall, the results demonstrate that both bovine feces and the surrounding environments, including pasture and forest, are important habitats for Cynipoidea parasitoids. These parasitoids contribute to the natural regulation of dipteran populations associated with decomposing substrates. The results also show that complementary statistical analyses can strengthen ecological interpretation by distinguishing between total abundance patterns and differences in taxonomic composition (Figure 13) (Marchiori *et al.*, 2000).



Figure 13: Lateral view of Eucoilinae sp., illustrating general morphological characteristics of the group, including a slender body, filiform antennae, and reduced wing venation. The illustration highlights key diagnostic features such as thoracic structure, scutellar plate, and abdominal configuration typical of Eucoilinae parasitoids

10. Ichneumonidae (Hymenoptera) as parasitoid of Diptera Muscomorpha in Brazil

A total of 50 pupae of *Hemilucilia segmentaria* (Fabricius, 1805) (Diptera: Calliphoridae) were collected, from which 10 specimens of *Pimpla* sp. (Hymenoptera: Ichneumonidae) emerged, resulting in a parasitism rate of 20.0%. The results demonstrate that *Pimpla* sp. acts as a parasitoid of *H. segmentaria*, representing the first record of this parasitoid-host association. This finding highlights the ecological

importance of Ichneumonidae species as natural regulators of Diptera populations, especially those of medical and veterinary relevance. *Hemilucilia segmentaria* showed a strong association with forest environments, where the highest abundance of individuals was recorded, particularly in substrates such as bovine liver. This pattern suggests that environmental conditions and substrate type directly influence host availability and, consequently, parasitoid occurrence (Figure 14).

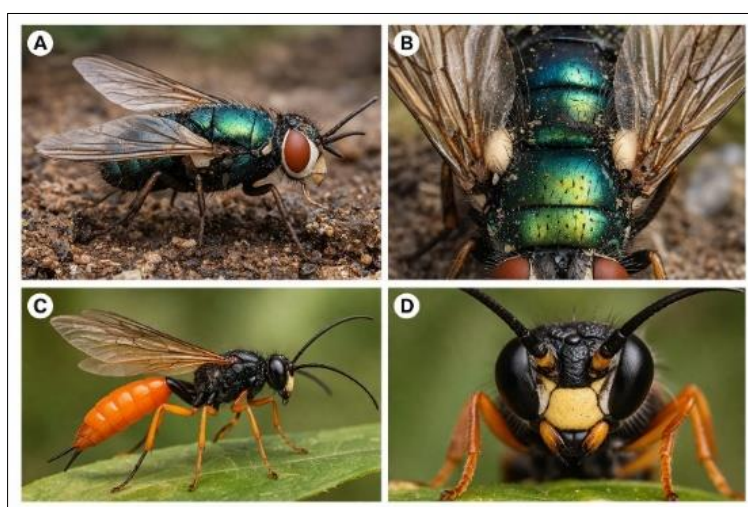


Figure 14: (A) Adult of *H. segmentaria* collected in decomposing organic substrate; (B) thoracic detail showing diagnostic morphological characteristics. (C) Adult of *Pimpla* sp. (Hymenoptera: Ichneumonidae) with orange abdomen, parasite of Diptera pupae; (D) frontal view of the head highlighting sensory and morphological structures

The presence of *Pimpla* sp. as a parasitoid reinforces the role of Ichneumonidae as important biological control agents. Members of the subfamily Pimplinae include parasitoids of pupae and larvae, exhibiting diverse strategies such as ectoparasitism and endoparasitism, which increase their efficiency in regulating host populations. In addition, the limited knowledge regarding parasitoids of Diptera in Brazil emphasizes the need for further studies aimed at identifying species with potential use in biological control programs. The ability of these parasitoids to exploit specific hosts, such as *H. segmentaria*, indicates their relevance in integrated pest management strategies (Marchiori, 2020).

11. Microhymenopterans of *M. Domestica* Collected on Different Substrates in Brazil

A total of 11,403 pupae of *M. domestica* were collected from different substrates, of which 59 were

parasitized, yielding 98 parasitoid individuals and an overall parasitism rate of 0.5%. Chicken manure was the substrate with the highest number of pupae, with 7,779 pupae, corresponding to 68.2% of the total collected hosts. However, despite the high abundance of hosts, parasitism in chicken manure remained low, indicating that host density alone did not determine parasitoid efficiency.

Pachycrepoideus vindemmiae was the most abundant species, confirming its importance as a generalist parasitoid of Diptera. This species is widely distributed and associated with several dipteran families, reinforcing its ecological role as a natural regulator of fly populations. The genus *Spalangia* was also well represented, with species such as *S. cameroni*, *S. endius*, *S. nigra*, and *S. nigroaenea*, all known as pupal parasitoids associated with synanthropic flies (Table 12).

Table 12: Parasitoids of *M. domestica* collected in different substrates in Itumbiara, Goiás, Brazil. Number of pupae, parasitized pupae, and occurrence of parasitoid species. Distribution of parasitoids according to substrate type

Substrate	Total pupae	Parasitoid species	N. Individuals	Parasitized pupae	Parasitism rate (%)
Bovine liver	48	<i>Nasonia vitripennis</i>	34	3	6.3
Cattle dung	3.370	<i>Spalangia cameroni</i>	1	1	0
Chicken manure	7.779	<i>Pachycrepoideus vindemmiae</i>	17	17	0.2
		<i>Spalangia cameroni</i>	12	12	0.2
		<i>Spalangia endius</i>	8	8	0.1
		<i>Spalangia nigra</i>	1	1	0
		<i>Spalangia nigroaenea</i>	6	6	0.1
Chicken viscera	45	<i>Nasonia vitripennis</i>	5	1	2.2
Food leftovers	50	<i>Pachycrepoideus vindemmiae</i>	9	9	18.0
Human feces	111	<i>Hemencyrtus herbertii</i>	5	1	0.9
Total	11.403		98	59	0.5

Higher parasitism rates were observed in bovine liver (6.3%) and chicken viscera (2.2%), suggesting that substrates rich in organic matter and nutrients may favor parasitoid development. In contrast, lower parasitism frequencies in chicken manure and cattle dung indicate that substrate type significantly influences parasitoid-host interactions. *Nasonia vitripennis* was recorded as a gregarious parasitoid, commonly associated with pupae of Diptera from several families. Additionally, *H. herbertii* was observed as a larval parasitoid, developing internally within the host and emerging from the pupal stage. These differences in parasitic strategies highlight the diversity of ecological adaptations among parasitoid species (Figure 15).

The results reinforce the importance of parasitoids as biological control agents of *M. domestica*, although their effectiveness depends on environmental conditions, substrate type, and host availability. Further studies are necessary to evaluate their potential application in integrated pest management programs (Marchiori, 2006).

13. Efficiency of Baited Traps for Capturing Synanthropic Diptera and Their Parasitoids

The use of baited traps demonstrated high efficiency in the collection of synanthropic Diptera and their associated parasitoids under field conditions. The trap model employed allowed not only the attraction of insects but also their retention and preservation, which is essential for subsequent identification and ecological analysis. The presence of decomposing organic material played a key role in attracting a high diversity of dipteran species (Figure 16).

The results indicated that Diptera abundance varied according to the type and stage of decomposition of the bait. Substrates in advanced stages of decomposition showed greater attractiveness, likely due to the release of volatile compounds that stimulate oviposition. This increase in host availability directly influenced the occurrence and diversity of parasitoids recorded in the samples. Parasitoids were consistently associated with dipteran hosts, confirming their ecological dependence on host density and availability. Some species exhibited generalist behavior, parasitizing multiple host species, which suggests a high level of

adaptability to different environmental conditions. This ecological plasticity enhances their importance in natural and applied biological control programs (Figure 17).

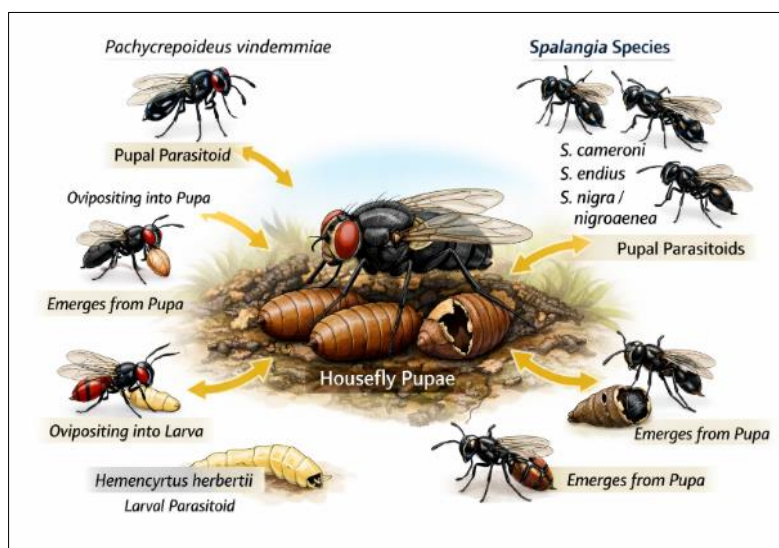


Figure 15: Parasitoid species associated with *M. domestica* pupae in different substrates, highlighting pupal and larval parasitism strategies. Main taxa include *P. vindemmiae* and species of *Spalangia*, which act as pupal parasitoids, and *H. herbertii*, a larval parasitoid. The figure illustrates interactions between parasitoids and host pupae, emphasizing their role in the biological control of synanthropic flies



Figure 16: Developmental stages of a parasitoid associated with Diptera, showing two pupae and one adult specimen. Variation in pupal coloration reflects different stages of development before adult emergence. The adult parasitoid exhibits morphological characteristics typical of hymenopteran parasitoids involved in the regulation of dipteran populations



Figure 17: Black cylindrical trap used for capturing synanthropic Diptera under field conditions, suspended from a tree branch. The trap design facilitates the attraction and retention of flies through decomposing organic bait placed inside the container. This method is widely used in ecological studies to evaluate dipteran diversity and associated parasitoid fauna

Furthermore, the trap design contributed significantly to the effectiveness of sampling, minimizing specimen damage and allowing accurate taxonomic identification. This is particularly relevant for studies involving parasitoid-host interactions, where precise identification is crucial for understanding ecological dynamics. The continuous presence of parasitoids throughout the sampling period indicates their establishment in the studied environment and their potential role in regulating dipteran populations. These findings reinforce the importance of parasitoids as natural enemies and highlight their potential application in integrated pest management programs, especially in environments with high organic matter accumulation, such as livestock systems (Marchiori, 2021d).

5.0. DISCUSSION

The results of this study demonstrate that parasitoid diversity and parasitism patterns are strongly influenced by environmental conditions, substrate type, and host availability. As summarized in the analyzed datasets, parasitoid-host associations varied considerably across different ecological contexts, indicating that both biotic and abiotic factors play a key role in structuring these interactions. Species such as *Aphaereta* sp., *H. herberti*, and *N. vitripennis* showed clear associations with specific hosts and substrates, reflecting differences in parasitic strategies and ecological adaptations. These patterns agree with studies showing that parasitoid occurrence is directly influenced by host availability, habitat structure, and ecological conditions (Marchiori, 2019d; Peralta-Aragón *et al.*, 2025; Rocha *et al.*, 2025; Gudín *et al.*, 2026; Guimarães *et al.*, 2026; Huang *et al.*, 2026; Krčmar *et al.*, 2026).

An important contribution of the present study was the use of quantitative information available in the reviewed articles to perform complementary chi-square analyses. In several original studies, the data were presented mainly in descriptive form, with values of abundance, frequency, number of pupae, number of parasitized pupae, and parasitism rates, but without formal statistical comparison. The present review, therefore, did not modify the original records but used the available numerical data to test whether differences in parasitoid abundance, substrate use, host association, habitat, altitude, and parasitism rates were statistically supported. This approach strengthened the interpretation of ecological patterns that had previously been described only descriptively and allowed a clearer evaluation of non-random patterns in parasitoid-Diptera interactions (Huang *et al.*, 2026; Krčmar *et al.*, 2026).

The predominance of parasitoids associated with decomposing organic matter highlights the importance of these substrates as key ecological niches.

Organic materials such as feces, carcasses, animal waste, and other decomposing substrates provide suitable conditions for the development of Diptera, which in turn support parasitoid populations. This relationship reinforces the idea that parasitoids are indirectly regulated by resource availability through their hosts. Environments with higher accumulation of organic matter, such as urban areas, livestock systems, poultry farms, and carcass-associated habitats, showed greater interaction frequency between parasitoids and hosts. These findings support the importance of decomposing substrates as ecological reservoirs for both synanthropic Diptera and their natural enemies (Marchiori, 2020; Marchiori, 2021d; Marchiori, 2022; Marchiori, 2023; Schuster and Sivakumar, 2024; Hopper and Leung, 2026; Krčmar *et al.*, 2026; Österman *et al.*, 2026; Teng *et al.*, 2026; Zhang *et al.*, 2026).

Parasitism rates varied widely among the studied systems, ranging from low values in some substrates to relatively high levels in others. This variability may be explained by differences in host density, environmental stability, substrate quality, and parasitoid behavior. High parasitism rates observed in certain hosts and substrates suggest favorable conditions for parasitoid development, while low rates may reflect environmental limitations, reduced host accessibility, or low suitability of the host species. The complementary chi-square analyses helped confirm that many of these differences were not random, especially when comparisons involved host species, substrate type, or parasitoid abundance. These findings are consistent with previous studies that highlight the dynamic nature of parasitoid-host interactions in heterogeneous environments (Marchiori, 2021a; Marchiori, 2023; Peralta-Aragón *et al.*, 2025; Rocha *et al.*, 2025; Hopper and Leung, 2026; Krčmar *et al.*, 2026).

The presence of both generalist and more specialized parasitoid species further emphasizes the complexity of these ecological relationships. Generalist species, such as *P. vindemmiae*, were able to exploit a wide range of hosts, demonstrating high ecological plasticity and adaptability. In contrast, parasitoids such as *Aphaereta* sp. showed stronger associations with specific hosts, suggesting a higher degree of host specificity. These differences in ecological strategies are important for understanding parasitoid efficiency and their potential application in biological control programs. Generalist parasitoids may be useful in environments with diverse dipteran hosts, whereas more specialized parasitoids may be more efficient in regulating particular host populations under specific ecological conditions (Figure 18) (Marchiori and Linhares, 1999; Marchiori, 2007; Jaume-Schinkel and Mengual, 2022; Schuster and Sivakumar, 2024; Peralta-Aragón *et al.*, 2025; Rocha *et al.*, 2025).

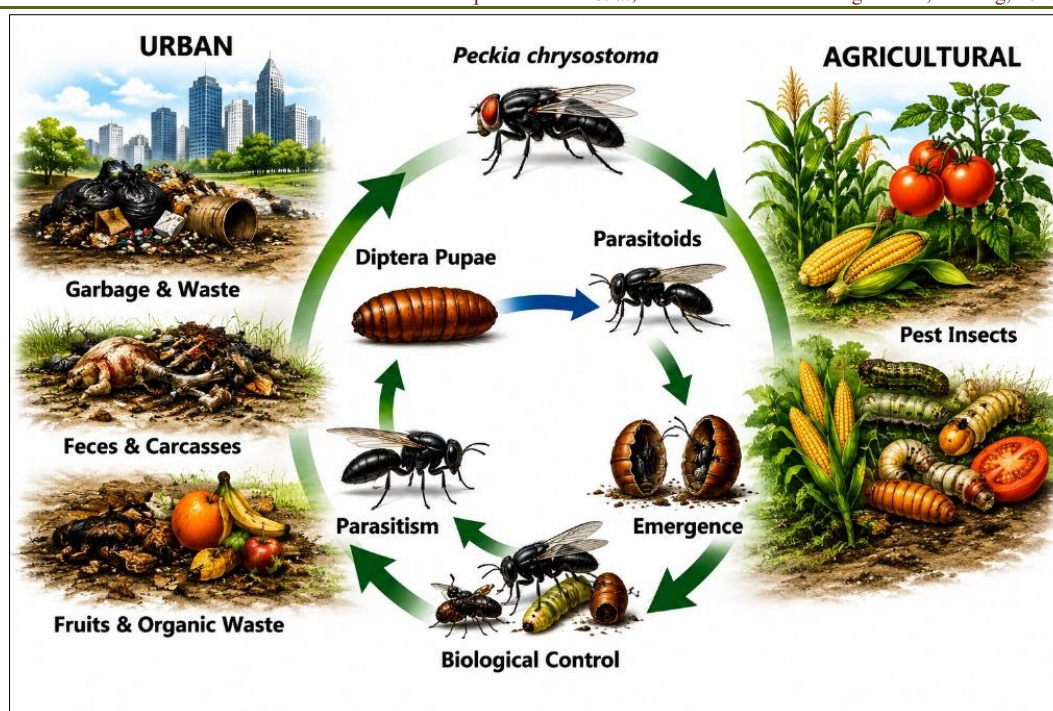


Figure 18: Parasitoid–Diptera interactions across urban and agricultural environments, highlighting the life cycle and parasitism process. Organic substrates support dipteran development, which in turn sustains parasitoid populations and ecological interactions. These relationships demonstrate the role of parasitoids as key agents in the natural regulation and biological control of pest insects

Another important aspect observed in this study is the influence of environmental context on parasitoid diversity. Forest environments, although sometimes presenting lower abundance, may act as important reservoirs of biodiversity, maintaining a wide variety of parasitoid species. In contrast, urban, livestock, and agricultural environments tend to support higher host densities, which may lead to increased parasitoid abundance. This complementary role of different habitats highlights the importance of landscape heterogeneity in maintaining ecological balance. The statistical analyses performed in the present review also showed that habitat, altitude, substrate type, and host distribution may significantly affect parasitoid occurrence, although some comparisons were not significant, indicating that parasitoid responses depend on the interaction of multiple ecological factors (Marchiori, 2021a; Huang *et al.*, 2026; Krčmar *et al.*, 2026; Österman *et al.*, 2026).

From an applied perspective, the results reinforce the importance of parasitoids as natural biological control agents of synanthropic and agricultural pests. Species associated with medically and economically important Diptera, such as *M. domestica* and *C. albiceps*, play a relevant role in reducing host populations and may contribute to integrated pest management strategies. The occurrence of parasitoids across different environments suggests that they can be useful in systems with high organic matter availability, especially where chemical control is limited or

undesirable (Marchiori, 2021b; Jaume-Schinkel and Mengual, 2022; Schuster and Sivakumar, 2024).

However, parasitoid efficiency may be influenced by environmental variability, host availability, substrate condition, and methodological differences among studies. As this work is based on compiled data from multiple sources, variation in sampling effort and experimental design is expected. Even so, the use of complementary chi-square analyses provided additional statistical support and strengthened the ecological interpretation of parasitoid diversity, host associations, and parasitism patterns in different Brazilian environments (Figure 18) (Gudin *et al.*, 2026; Guimarães *et al.*, 2026).

6.0. CONCLUSION

The present study demonstrates that parasitoids play a fundamental role in regulating populations of Diptera associated with decomposing organic substrates across different environments. The diversity and distribution of parasitoid species were strongly influenced by substrate type, host availability, and environmental conditions, highlighting the complexity of parasitoid–host interactions. The results revealed consistent associations between specific parasitoids and their hosts, with both generalist and more specialized strategies contributing to ecological balance. Environments with greater availability of organic matter, such as urban and livestock systems, supported higher interaction frequencies, while natural environments acted as important reservoirs of parasitoid diversity. The

reviewed articles provided quantitative data that enabled complementary chi-square analyses, although these statistical tests were not applied or reported in several of the original studies. These analyses added statistical support to ecological patterns previously described mainly in descriptive terms, reinforcing the non-random nature of parasitoid occurrence, host association, substrate use, and parasitism. Therefore, parasitoids represent an important component in the natural regulation of synanthropic and agricultural pest species, particularly those of medical and veterinary relevance. A better understanding of parasitoid diversity, behavior, host associations, and environmental responses is essential for developing sustainable biological control strategies and maintaining ecosystem stability.

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