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Research of Diptera (Insecta: Muscomorpha) and Their Parasitoids (Insecta: Hymenoptera Parasitica) Collected in a Forest Area in Brazil

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ABSTRACT

The objective of this study was to carry out a survey of the orders Diptera (Insecta: Muscomorpha) and Hymenoptera Parasitica (Insecta: Hymenoptera) in the forest area of Brazil. The study was carried out on a farm, located in the municipality of Goias, Midwest region, Brazil, from March to April 2015. Adult flies were collected using traps constructed with a matte black can. Fish, bovine kidneys, human feces and chicken viscera (300 g) deposited inside the cans, on a layer of earth, served as bait to attract the flies. After this sand was sieved (15 days after it was placed in the field), the pupae were extracted, and later placed individually in glass flasks, to obtain flies and/or parasitoids. A total of 734 pupae of flies were collected 89 specimens of parasitoids. The percentage of muscoid Diptera collected were Fannia pusio (Wiedemann, 1830) (Diptera: Fanniidae) (44/735) with 6.0%, Chrysomya megacephala (Fabricius, 1794) (Diptera: Calliphoridae) (14/735) with 1.9%, Oxysarcodexia thornax (Walker, 1849) (67/735) with 9.0%, Musca domestica L., 1758 (Diptera: Muscidae) (274/735) with 37.3%, Ophyra aenescens (Wiedemann, 1830) (Diptera: Muscidae) (46/735) with 6.3%, Tricharaea occidua (Fabricius, 1794) (Diptera: Sarcophagidae) (60/735) with 0.8% and Peckia chrysostoma (Wiedemann, 1830) (Diptera: Sarcophagidae) (230/735) with 31.3%. Two species of parasitoids were obtained: Pachycrepoideus vindemmiae (Rondani, 1875) (Hymenoptera: Pteromalidae) (25/89) with 28.1% and Brachymeria podagrica (Fabricius, 1787) (Hymenoptera: Chalcididae) (64/89) with 71.9%. The total percentage of parasitism was (89/735) 12.1%. In relation to the substrates: Bovine kidneys presented a percentage of parasitism (2/89) of 2.2%, chicken viscera (46/49) 51.7%, human feces (34/89) 38.2% and fish (7/89) 7.9%. This information contributes to studies aimed at the prevention of what can be transmitted by these insects' diseases as well as to the formulation of more effective methods of control.

Keywords: Biocontrol; Flies; Forensic Entomology; Natural Enemies; Public Health

INTRODUCTION

Some dipterans are of fundamental medical and veterinary importance since they can produce myiasis and act in the transmission of pathogens to humans and other animals. Diptera is valuable for Forensic Entomology, larval therapy, pollination, decomposition of organic, necrophagous, matter, predators, parasitoids, ectoparasitic mammals, and vectors of pathogens (Amendt, 2021; Brighton, 2020; Mendes et al., 2021; Perveen, 2021).

The occurrence, distribution and predominance of these flies in metropolitan areas are factors of great importance in public health. In rural areas, they can lead to a decrease in egg production and animal diseases, in addition to causing discomfort to the population neighboring the creations (Figure 1) (Brighton, 2020; Brighton, 2021; Marchiori, 2021g; Marchiori, 2022; Perveen, 2021).

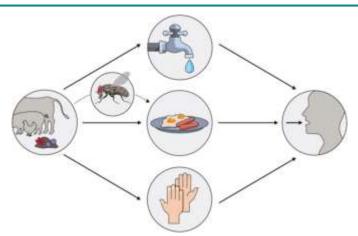


Figure 1. Pathways of fecal-oral transmission. Pathogens from feces can be transmitted to humans or animals through drinking water, food, or hands (smear infection). Flies can enhance the contamination of food, as no direct contact with food and feces is necessary

Source: https://www.sciencedirect.com/science/article/pii/S1477893918300371

Among the means of controlling flies, the most used are chemical insecticides that can lose their efficiency as populations become resistant to them. In addition, treatment with these substances has had an impact on the natural enemies of these insects, since some insecticides do not only affect but harm the fauna of parasitoids and fly predators, which are responsible for the natural reduction of populations of harmful insects. The emergence of resistance to insecticides justifies the growing need to implement alternative control programs aimed at controlling flies (Marchiori, 2019a; 2019b; 2021a; 2021g).

Cultural control is more a matter of awareness that seeks to show producers the need for periodic removal of feces accumulated in manure pits. Studies indicate that the most important controllers in cattle breeding are predators, competitors, and parasitoids. Most parasitoids belong to the insect group, mainly to the orders Diptera (flies) and Hymenoptera, and the females usually deposit their eggs on the hosts. Thus, when hatching from the eggs, the larvae consume the individual host that serves as a resource in the initial phase of development, killing it many times before it reproduces for the first time (Marchiori, 2019a, 2019b, 2021b; Wang et al., 2021; Zhang et al., 2021).

The feasibility of using controllers and the advantages of using them in agriculture and animal husbandry are indisputable, due to their low cost, ease of handling, selectivity and non-contamination of the environment. Parasitoids are significant regulators of biodiversity, pollination, manipulation of the host, and mutualistic relationships contributing to diversity and sustainability in tropical forests (Barbosa et al, 2021; Wang et al., 2021; Zhang et al., 2021).

The objective of this study was to carry out a survey of the orders Diptera (Insecta: Muscomorpha) and Hymenoptera Parasitica (Insecta: Hymenoptera) in the forest area of Brazil.

MATERIALS AND METHODS

The study was carried out on a farm, located in the municipality of Goias, Midwest region, Brazil, from March to April 2015. Adult flies were collected using traps constructed with a matte black can, measuring approximately 19 cm in diameter, height by 9 cm in diameter, with two shutter-type openings, located in the lower third, to allow insects to enter. Nylon funnels were attached to the upper part of the cans, open at the ends, with bases facing downwards and wrapped in plastic bags whose removal allowed the collection of flies (Figure 2).

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Figure 2: Map of Brazil with divisions of states and regions Source: https://www.mappr.co/political-maps/brazil-map/

Fish, bovine kidneys, human feces and chicken viscera (300 g) deposited inside the cans, on a layer of earth, served as bait to attract the flies. Five traps were used, hung from eucalyptus trees one meter from the ground, two meters from each other and 50 meters from household waste. The collected individuals were taken to the laboratory, sacrificed with ethyl ether and preserved in 70% alcohol for later identification. To obtain the parasitoids, the contents of the traps were placed in plastic containers containing a layer of sand to serve as the substrate for the pupation of the larvae. After this sand was sieved (15 days after it was placed in the field), the pupae were extracted, and later placed individually in glass flasks, to obtain flies and/or parasitoids (Figure 3).



Figure 3. General aspects of the trap Source: Author

Methods of Data Analysis

The preference of the parasitoids for the host and substrate was reached through the Chisquare with a probability of 5.0%.

Percentage of Parasitism

The total percentage of parasitism was calculated by counting the number of parasitized pupae/total number of collected pupae x 100. The percentage of parasitism of each parasitoid species was calculated by counting the number of parasitized pupae by each parasitoid species/total number of pupae from that host x 100.

RESULTS AND DISCUSSION

A total of 734 pupae of flies were collected 89 specimens of parasitoids (Table 1). The percentage of muscoid Diptera collected were *Fannia pusio* (Wiedemann, 1830) (Diptera: Fanniidae) (44/735) with 6.0%, *Chrysomya megacephala* (Fabricius, 1794) (Diptera: Calliphoridae) (14/735) with 1.9%, *Oxysarcodexia thornax* (Walker, 1849) (67/735) with 9.0%, *Musca domestica* L., 1758 (Diptera: Muscidae) (274/735) with 37.3%, *Ophyra aenescens* Wiedemann 1830 (Diptera: Muscidae) (46/735) with 6.3%, *Tricharaea occidua* (Fabricius, 1794) (Diptera: Sarcophagidae) (60/735) with 0.8% and *Peckia chrysostoma* (Wiedemann, 1830) (Diptera: Sarcophagidae) (230/735) with 31.3% (Table 1). *Musca domestica* had a higher percentage than the other species, probably due to its synanthropic rate or interspecific competition between flies.

Regarding substrate: *M. domestica* presented 274 individuals in human feces (Table 1), this is the highest number of individuals possibly due to the high presence of bacteria in the feces. *Musca domestica* had a higher percentage, probably due to its synanthropic rate synanthropic animals are those that have adapted to live with man, despite his will. They differ from domestic animals, which man raises and takes care of for the purposes of companionship dogs, cats, birds, among others, food production, or transportation). *Musca domestica* is the species of greatest sanitary interest, due to, its endophily, abundance in urban areas, ability to develop in various types of substrates, high reproduction power and being identified as the carrier of pathogens to man and animals (Hassan et al., 2020; Marchiori, 2021; Pereira et.al.; Santos et al., 2021; Zhang et al., 2021).

Substrates/ Taxonomic groups	Number/ individuals	Species	Number/ pupae	% Parasitism
Bovine kidneys				
Fannia pusio	44	Pachycrepoideus vindemmiae	2	4.5
Chicken viscera				
Chrysomya megacephala	14	Brachymeria podagrica	2	14.3
Oxysarcodexia thornax	67	Brachymeria podagrica	10	15.0
Peckia chrysostoma	174	Brachymeria podagrica	34	19.5
Human feces				
Musca domestica	274	Pachycrepoideus vindemmiae	18	6.6
Ophyra aenescens	46	Brachymeria podagrica	11	24.0
Tricharaea occidua	60	Pachycrepoideus vindemmiae	5	8.3
Fish				
Peckia chrysostoma	56	Brachymeria podagrica	7	10.5

 Table 1. Diptera and their parasitoids were collected in different substrates in a forest area, from March to September 2015 in Brazil

Regarding the other dipterans obtained: *Fannia pusio* belongs to the *canicularis* group and the *pusio* subgroup, whose males have a *trimaculate* abdomen dorsally with black legs and thorax. Adults are often found associated with the human-modified environment, with their larvae rearing in decaying organic matter (Grzywacz et al., 2022; Marchiori, 2021e).

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Chrysomya megacephala is often found associated with a human-modified environment, with its larvae rearing in decomposing animal organic matter. This dipteran is of great medicalsanitary interest and its occurrence, distribution and predominance in metropolitan areas are factors of great importance. They have already been observed in human and domestic animal cadavers. Adults can be attracted to substances in the process of fermentation, decomposition, blood and wounds (Hassan et al., 2020; Marchiori, 2021c; Pereira et al., 2021).

Ophyra aenescens included in Muscidae, Azeliinae and Hydrotaeini, in phase larvae is a facultative predator of fly larvae synanthropic species, especially *Musca domestica*, on pig and poultry farms are often found in poultry farms, but does not represent significant public health problem (Hassan et al., 2020; Pereira et al., 2021).

Oxysarcodexia thornax is mostly coprophagous or predator. However, some species are widely found associated with decaying vertebrate carcasses, having been considered the species with the greatest diversity found associated with carcasses (Barbosa et al., 2021; Derlinge, 2022; Dufe et al., 2020; Santos et al., 2021; Silva et al., 2019).

Peckia chrysostoma is a species that can be found in built-up areas close to forests and in urban centers. Larvae can usually be found feeding on various substrates, such as: Living tissue, animal carcasses, urban garbage and feces. These flies, because they frequent certain habitats for feeding and/or laying, become potential vectors of several pathogens of great medical-sanitary interest (Barbosa et al., 2021; Derlinge, 2022; Dufe et al., 2020; Santos et al., 2021; Silva et al., 2021).

Tricharaea occidua has a great variety of feeding and development habits, which take place in animal carcasses, excrement and decomposing organic matter; making them possible vectors of pathogens (Santos et al., 2022; Silva et al. 2021).

Larval Therapy

Biological debridement, also known as larval therapy, larva therapy, or Maggot therapy, consists of using sterilized fly larvae on complex lesions, with the presence of necrotic tissue, in order to optimize their healing. This technique consists of a type of biological debridement, in which the wound bed is cleaned by fly larvae, which are selective for necrotic tissue. Despite the scientifically proven benefits and the fact that it has been used for thousands of years, in Brazil, only Rio Grande do Norte performs this procedure (Figure 4) (Gama et al., 2021; Wilson et al., 2019).



Figure 4. Wounds presented for evaluation by the subjects Source: https://www.mdpi.com/1660-4601/19/5/2895

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Forensic Entomology

Uses the information that insects provide – biological, ecological and distribution data – to elucidate legal issues in general, especially criminal matters. It can be said that Forensic Entomology is the science that applies the study of insects and other arthropods to legal procedures. It is divided into three main areas focused on issues that are frequently the subject of litigation (Figure 5) (Marchiori, 2021f; Silva, 2023).

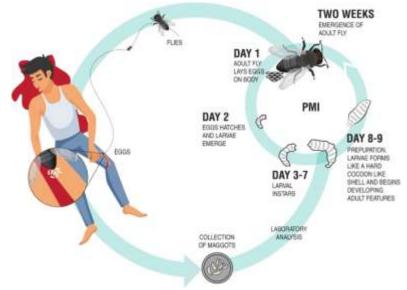


Figure 5. Figure Tools and Techniques in Forensic Entomology Source: https://link.springer.com/article/10.1007/s42690-022-00823-5

Two species of parasitoids were obtained: *Pachycrepoideus vindemmiae* (Rondani, 1875) (Hymenoptera: Pteromalidae) (25/89) with 28.1% and *Brachymeria podagrica* (Fabricius, 1787) (Hymenoptera: Chalcididae) (64/89) with 71.9%. This result may be due to the greater competitiveness of the parasitoid *B. podagrica* in the larval stage or intraspecific ecological relationships between harmonious and inharmonious parasitoids.

Pachycrepoideus vindemmiae is cited in works on population surveys, in addition to basic studies for control strategies. It is a solitary pupal ectoparasitoid of Diptera that has a cosmopolitan distribution. It is classified as a generalist, with records of attacks on more than 60 species, whose hosts belong to the Drosophilidae, Anthomyiidae, Calliphoridae, Muscidae, Sarcophagidae, Tachinidae and Tephritidae families (Figure 6) (Hood et al., 2021; Marchiori, 2021b; 2021d; Yang et al., 2020).



Figure 6. Pachycrepoideus vindemmiae (Rondani, 1875) (Hymenoptera: Pteromalidae) Source: https://mundoagro.cl/liberan-agente-de-control-biologico-para-drosophila-suzukii-encultivos-organicos/

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Regarding the wide range of dipterans associated with *P. vindemmiae*, the South American fruit fly *Anastrepha fraterculus* (Wiedemann, 1830) (Diptera: Tephritidae), the Mediterranean fruit fly *Ceratitis capitata* (Wiedemann, 1824) (Diptera: Tephritidae) and the spotted-wing drosophila *Drosophila suzukii* (Matsumura, 1931) (Diptera: Drosophilidae) constitute one of the most important groups of pests in Brazilian fruit production (Boughdad et al., 2021; Brighton, 2021; Marchiori, 2021d; Silva et al., 2019).

Brachymeria podagrica is a solitary endoparasitoid with a koinobiont strategy. This allows the host to continue its development while feeding upon it, the female ovipositing within the larval stages of the host while the adult emerges from the pupa. the wasp *B. podagrica* is recorded for the first time for Colombia, a solitary parasitoid of larvae of the *Peckia collusor* (Curran & Walley, 1934) fly (Figure 4) (Andrade-Herrera & Marchiori, 2018; Ramirez-Mora & Durango-Manrique, 2021) (Figure 7).



Figure 7. *Brachymeria podagrica* (Fabricius, 1787) (Hymenoptera: Chalcididae) Source: https://bugguide.net/node/view/552824/bgimage

Third instar larvae of *P. collusor* were exposed to outdoor ambient conditions for 6 hours in the garden of the Tecnológico of Antioquia (Medellín, Colombia). *Brachymeria podagrica* adults were obtained from the host pupae. The percentage of parasitism was 14.5 % (Schulter et al., 2023).

The total percentage of parasitism was (89/735) 12.1%, presumably due to the lower number of species reached, by the ability of parasitoids to search for hosts, the amount of substrate available and hypothetically the variety of substrate offered and interspecific competition and promotes coexistence among insect parasitoids.

In relation to the substrates: Bovine kidneys presented a percentage of parasitism (2/89) of 2.2%, chicken viscera (46/49) 51.7%, human feces (34/89) 38.2% and fish (7/89) 7.9%. Three hundred (300) g of each substrate were placed in the traps, perhaps the kidneys allowed for more moisture and odor release time than other baits. *B. podagrica* by and *M. domestica*, *O. aenescens*, *O. thornax* and *P. chrysostoma*; *P. vindemmiae* showed a preference for *F. pusio*, *M. domestica* and *T. occidua* (X^2 =361.8).

CONCLUSION

Dipterans are of fundamental medical and veterinary importance since they can produce myiasis and act in the transmission of pathogens to humans and other animals. Studies indicate that the most important controllers in cattle breeding are predators, competitors, and parasitoids. *Musca domestica* was the most collected species of dipterans with 37.3%, in relation to parasitoids the most obtained species was *B. podagrica* with 71.9%. The total percentage of parasitism was 12.1%. Among the substrates: Chicken viscera at 51.7% and

human feces at 38.2% were the most superior. It is important more intensive study of hymenopteran parasitoids is required before we can understand the general conditions that favor the evolution of insect parasitoids and the truly magnifying themes of their behavior and ecology.

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