Interaction and Distribution of Beetles (Insecta: Coleoptera) Associated with Heliconia bihai (Heliconiaceae) Inflorescences

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Interaction and distribution of beetles (Insecta: Coleoptera) associated with Heliconia bihai (Heliconiaceae) inflorescences

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Abstract

The insect fauna associated with the genus Heliconia (Heliconiaceae) is quite diversified and includes terrestrial and aquatic species. In plants with an upright inflorescence, the bracts may form phytotelmas. Insects are one of the main taxa that have adapted to phytotelma environments, including species of Diptera, Coleoptera, Hemiptera, Dermaptera, Plecoptera, Trichoptera, and Odonata, among other orders. The Coleoptera fauna associated with phytotelma is poorly studied, and the objective of this study was therefore to determine the distribution of beetles in Heliconia bihai (L.) (Zingiberales: Heliconiaceae) bracts in cultivated and uncultivated areas in northeastern Brazil. In addition, we describe their functional relationships. With respect to the coleopteran assemblage, 6 families were found in both areas and 3 were found only in the uncultivated Heliconia. The most abundant and frequently occurring family in both areas was Hydrophilidae, represented by a single species, Pelosoma lafertei (Mulsant) (Coleoptera: Hydrophilidae). Staphylinidae were the second most abundant and the second most frequently occurring family. The detritivores were most abundant (6 species), followed by herbivores (3 species), and predators (2 species). Abundance and richness were higher in the uncultivated area. Temperature and pH were the main factors affecting the coleopteran assemblages. A high acidity indicates an environment rich in organic matter, the main food source in phytotelma environments.

Key Words: phytotelma; aquatic insects; tropical flowers; bracts

Resumo

A fauna de insetos associada ao gênero Heliconia (Heliconiaceae) é bastante diversificada e inclui espécies terrestres e aquáticas. Em plantas com inflorescência vertical, as brácteas podem formar fitotelmas. Os insetos são um dos principais táxons adaptados aos ambientes fitotelmatas, incluindo espécies de Diptera, Coleoptera, Hemiptera, Dermaptera, Plecoptera, Trichoptera e Odonata, entre outras ordens. A fauna de Coleoptera associada ao ambiente fitotelma é pouco estudada, e o objetivo deste estudo foi determinar a distribuição de besouros nas brácteas de H. bihai (L.) (Zingiberales: Heliconiaceae) em áreas cultivadas e não cultivadas no Nordeste do Brasil. Além disso, descrevemos suas relações funcionais. Em relação à ocorrência de coleópteros, 6 famílias foram encontradas em ambas as áreas e 3 foram encontradas apenas em brácteas de Heliconia não cultivada. A família mais abundante e frequentemente encontrada em ambas as áreas foi Hydrophilidae, representada por uma única espécie, Pelosoma lafertei (Mulsant) (Coleoptera: Hydrophilidae). Os Staphylinidae foram a segunda família mais abundante e mais frequente. Os detritívoros foram mais abundantes (6 espécies), seguidos de herbívoros (3 espécies), e predadores (2 espécies). A Abundância e a riqueza foram maiores na área não cultivada. A temperatura e o pH foram os principais fatores que afetaram a assembléia de coleópteros. Uma alta acidez indica um ambiente rico em matéria orgânica, o principal recurso alimentar do ambiente fitotelma.

Palavras Chave: fitotelma; insetos aquáticos; flores tropicais; brácteas

In plants with upright inflorescences, microhabitats called phytotelmas may form in the bracts. Each phytotelma consists of a small water reservoir formed by plant structures that have the ability to retain water (Maguire 1971; Machado-Alisson et al. 1983; Richardson et al. 2000). There is a great diversity of species associated with these small ecosystems. They occur primarily in plants found in the Neotropical region, where more than 1,500 plant species possess phytotelma, for example in the carnivorous plants, bromeliads, and axillary buds of many species that accumulate water. Phytotelma may contain a rich fauna, mainly arthropods, forming simple to complex food webs (Fish 1983; Kitching 2001). In phytotelma environments, the combination of physical and chemical conditions of the water, such as volume, pH, temperature, electrical conductivity, and the amount of available resources (e.g., organic matter and debris), regulates the abundance of the species present (Kitching 2001).

The insect fauna associated with the genus Heliconia (Heliconiaceae) is quite diversified and includes both terrestrial and aquatic species. Insects are one of the principal groups that have adapted to

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phytotelma environments, including, among other orders, species of Diptera, Dermaptera, Coleoptera, Hemiptera, Hymenoptera, and Lepidoptera (Seifert & Seifert 1979; Seifert 1980; Frank & Barrera 2010; Oliveira et al. 2010; Jalinsky et al. 2014; Staines & Garcia-Robledo 2014). The order Diptera occurs widely in all types of phytotelmata; however, studies on other groups of insects in these environments are more limited (Campos 2010). In particular, information on coleopteran fauna associated with Heliconia phytotelmata is still limited. Most of the species inhabiting phytotelmata are terrestrial; however, a few families are semiaquatic (e.g., Chrysomelidae) or aquatic (e.g., Hydrophilidae) (Frank & Barrera 2010; Aristizábal et al. 2013).

In Brazil, publications addressing the phytotelmata fauna focus on bromeliad water reservoirs (called ‘tanks’) in the northern and southeastern regions of the country (Mestre et al. 2001; Torreias & Ferreira-Keppeler 2011). Studies associating the Coleoptera order with Heliconia are scarce. However, the presence of beetles of the families Hydrophilidae, Staphylinidae, Nitidulidae, and Curculionidae in Heliconia bihai (L.) (Zingiberaceae: Heliconiaceae) was reported in northeastern Brazil by Oliveira et al. (2010).

One approach to understand the role of biodiversity is functional diversity, in which the species are classified based on their effects on ecosystem processes. There are different ways of quantifying functional diversity, one of which is to form groups of species that have similar functions in the ecosystem (Díaz & Cabido 2001). This allows us to discern the trophic relationships between these organisms and to better understand the ecosystem structure.

According to Benítez-Malvido et al. (2016), tropical forest fragmentation affects biotic interactions; however, information on how this process affects animal trophic guilds and their pattern of interactions with host plants is scarce. Those authors sought to assess changes in biotic interactions in continuous forests and fragments, assigning arthropods associated with Heliconia aurantiaca Verschaff. (Zingiberaceae: Heliconiaceae) into broad trophic guilds such as omnivores, herbivores, and predators. In this paper, we investigated coleopterans associated with Heliconia, with the objective to determine the distribution of beetles in H. bihai bracts, to categorize their functional relationships, and to determine if there is a specific relationship between the species and the phytotelma environment (wild and cultivated plants). We hypothesized that species richness, abundance, and diversity of Coleoptera in inflorescences of cultivated species would be lower than in inflorescences of naturally occurring species.

Materials and Methods

Inflorescences of H. bihai were harvested on a farm containing areas with cultivated and wild (uncultivated) Heliconia, located in Camaragibe-PE (8.0219°S, 34.9925°W), in northeastern Brazil. The vegetation in this area is characterized by trees, grass, and sugar cane. The distance between the cultivated area and the wild area is 700 m.

The cultivated area represented the collection of Heliconia germplasm of the Federal Rural University of Pernambuco, planted in Jan 2007, with partial shade provided by trees. Plants were cultivated with spacing of 3 m, and 4 m between rows, prepared in blocks with 4 replicates per genotype. Each plot was formed by a clump and covered a total area of 12 m², representing 6 m² of useful area. Irrigation was provided through a high sprinkler, and insecticides were not applied in the area.

The wild (uncultivated) area was represented by an Atlantic Forest fragment with a total area of 145.3 m², classified as Lowland Ombrrophylous Forest (Veloso et al. 1992). It is an evergreen forest with a canopy height of 30 m, with emergent trees up to 40 m tall. The area features clumps of H. bihai that are located in a steep portion between the edge of the forest and the watercourse, forming a shaded and moist area.

Inflorescences were collected monthly from Aug 2011 to Aug 2012; field collections were initiated at 7:00 AM and continued until 9:00 AM. In each area, all available inflorescences were collected and the bracts were counted. Abiotic parameters were measured directly on all bracts. The pH of the water in the bract was measured using a Hanna Instruments® portable potentiometer (Model number HI98100, Woonsocket, Rhode Island, USA).

To determine water temperature, an Incoterm® portable digital thermometer (Model number 6132, Porto Alegre, Rio Grande do Sul, Brazil), was used. Water volume in the tank was measured with a 5-mL graduated pipette. The liquid contained within the bracts was then pipetted into individual plastic pots. To collect the insects, the inflorescences were dissected and a brush and water jets were used to dislodge adults and larvae from inside the floral bracts. Subsequently, this liquid was examined with a stereoscopic microscope to collect the remaining insects. The insects were stored in bottles in 70% alcohol for further sorting and counting.

For each species, abundance, relative abundance, and frequency of occurrence were calculated. The abundance of each species was calculated by totaling the number of individuals observed in that species. The relative abundance was the number of a species in relation to the total number of individuals of all species observed in each inflorescence. The frequency of occurrence corresponds to the proportion of inflorescences in which each species occurred relative to the total number of inflorescences sampled. Diversity and evenness of the beetle fauna were analyzed using the Shannon-Wiener (H’) and Pielou’s (J’) indices (Magurran 2004). The species were classified following Bouchard et al. (2011) and vouchers were deposited in the Entomological Collection of the Zoology Department of the Federal University of Paraná, Curitiba, Brazil. The functional categorization of the beetle fauna followed the classification of Kitching (2000) and Merrit & Cummins (1996).

The Shapiro Wilk Test was used to test the normality of the data. Levene’s test was used to test the equality of the variances (homooscedasticity) of the abiotic and biotic parameters. The Mann-Whitney U Test was used to compare the diversity, richness, and abundance between the 2 collection areas. Kruskal-Wallis analysis was used to compare abundance by species, richness, and abundance among inflorescences with 2, 3, 4, and 5 bracts. All analyses were carried out using the software package Statistica 8.0 (Statsoft 2010).

To test the effect of environmental variables on the distribution patterns of the coleopteran phytotelmata fauna, we used canonical correspondence analysis. The canonical correspondence analysis was performed to verify the degree of relationship among abiotic variables (pH, temperature, and volume) and the most abundant species in each area (uncultivated and cultivated). The Monte Carlo test was performed with a standard of 999 permutations and a significance level of 5%. The tests were conducted in the software Canoco 4.5 (Ter Braak & Šmilauer 2002).

Results

A total of 2,331 beetles was collected from 293 H. bihai inflorescences, representing 9 families of Coleoptera (Table 1). Six families were found in both areas and 3 were found only in the uncultivated Heliconia (Table 1). Six families occurred in both the larva and adult stages, but most individuals were adults. The families with aquatic habitats were Hydrophilidae and Elmidae (both Coleoptera), whereas the family with semiaquatic habits was Nitidulidae and those with terres-
trial habits were Chrysomelidae, Staphylinidae, Histeridae, Curculionidae, and Scarabaeidae (all Coleoptera). The number of families with terrestrial habits was higher than the number of those with aquatic or semiaquatic habits.

The most abundant and frequently occurring family in the 2 areas was Hydrophilidae, represented by a single species, *Pelosoma lafertei* (Mulsant) (Coleoptera: Hydrophilidae). Staphylinidae was the second most abundant and frequently occurring family, represented by the only 2 predator species observed in the bracts (Table 1). The majority of the species among the sampled Coleoptera were detritivores (6), followed by herbivores (3), and predators (2) (Table 1).

The total number of beetles collected from the uncultivated area was 1,574, whereas we collected 757 from the cultivated area. Twelve morphospecies were recorded in the uncultivated area and 8 in the cultivated area (Table 1). Nitidulidae, Staphylinidae, and Curculionidae were the most species-rich taxa, with 2 morphospecies each. The uncultivated area displayed greater species richness (uncultivated: 1.57 ± 0.9; cultivated: 0.79 ± 1.4) and abundance (uncultivated: 6.9 ± 9.0; cultivated: 2.4 ± 5.0) per inflorescence (richness: *U* = 7,059; *Z* = −4.7; *P* < 0.001; abundance: *U* = 6,701; *Z* = −4.5; *P* < 0.001). The diversity (*H*’) also differed significantly between areas (*U* = 2,251; *Z* = −13.8; *P* < 0.001), with higher diversity in the uncultivated area (1.16 ± 0.04) than in the cultivated area (1.02 ± 0.11). There was no difference in evenness between areas (uncultivated: 0.71; cultivated: 0.74).

We sampled approximately equal numbers of inflorescences (20–50), with 2 to 5 bracts per inflorescence, in each area. In the uncultivated area, when comparing the inflorescences composed of a smaller number of bracts (2) with those with larger numbers (4 and 5), the abundance of *P. lefertei* and *Paederomimus* sp. (Coleoptera), and the abundance and total richness of beetles were higher in inflorescences with more bracts (Table 2). Comparing inflorescences with 3 and 4 bracts, only the total abundance of beetles was significantly different (Table 2).

A similar pattern can be observed in the cultivated area, where the abundance of *P. lefertei* and *Paederomimus* sp., and the abundance and total richness of beetles also were higher in inflorescences with more bracts (4 and 5) when compared to those with fewer bracts (2 and 3) (Table 3). The abundance of *Tachyporus* sp. (Coleoptera) in inflorescences with 3 and 4 bracts also was higher in the inflorescences with more bracts (Table 3).

The abiotic factors showed a small range in all size inflorescences (2–5 bracts) (Table 4) in both cultivated and uncultivated areas. Inflorescences in cultivated area showed bracts with a more acidic environment (trace = 0.248; *F*-ratio = 2.24; *P* = 0.046) and uncultivated (trace = 0.158; *F*-ratio = 1.77; *P* = 0.041) areas. In the uncultivated area, *Pelosoma lafertei* was positively correlated with pH and volume, whereas *Tachyporus* sp. and *Paederomimus* sp. were positively correlated with temperature. In the cultivated area, *P. lefertei* and *Elmidae* sp. were positively correlated with volume in the bracts, and *Tachyporus* sp. and *Paederomimus* sp. were affected by the temperature (Fig. 1).

**Discussion**

The uncultivated area presented a greater diversity relative to the cultivated area. Inflorescences with higher numbers of bracts showed...
greater abundance and total richness of beetles. The abundance of *Paederomimus* sp. and *P. lafertei* also presented this pattern in both areas. As the number of bracts increased in the inflorescences, abundance and richness also tended to increase. This change in the colonizing fauna has been reported previously (Seifert 1982; Richardson & Hull 2000; García-Robledo et al. 2005; Yee & Willig 2007) and recently, Benítez-Malvido et al. (2016) have shown the influence of the fragmentation of the tropical forest on the phytotelma arthropod biodiversity in *Heliconia* bracts. The habitat affects species abundance, and the diversity and composition were higher in continuous forest (Benítez-Malvido et al. 2016).

The Coleoptera fauna was predominantly detritivorous. The species *P. lafertei*, similar to a large portion of the Hydrophilidae, feed on organic matter present in the environment; adult beetles are mainly saprobic, whereas the larvae are predators of several invertebrates (Hansen 1999; Short & Hebauer 2006; Fikácek et al. 2010). *Pelosoma lafertei* has been reported previously in Pernambuco associated with the bromeliad *Vriesea infleta* Warrw. (Bromeliaceae) in fragments of the Atlantic Forest (Mestre et al. 2001).

The fact that the number of beetles with terrestrial habits was higher than those with aquatic habits may be associated with the fact that these species are visitors. They usually are found foraging on different parts of the inflorescence. There is a high diversity of terrestrial species in phytotelma environments in the neotropical region (Campos & Fernández 2011), and many studies have reported terrestrial beetles in bromeliads (Campos 2010; Torreias & Ferreira-Keppler 2011). Some studies have reported terrestrial families in *Heliconia* bracts (Frank & Barrera 2010; Oliveira et al. 2010; Aristizábal et al. 2013). The family Scarabaeidae has a diverse biology, including adults that feed on excreta, carcasses, fungi, foliage, pollen, fruits, or roots; some species are diurnal and can be found on leaves (Silva et al. 2011). Many species are pollinators, recycling plant material and excreta, and are considered beneficial (Schiestl & Dotterl 2012). There are few records associating this family with phytotelma, but in *Heliconia* spp., they are more common in parts of the plant where water does not accumulate (Aristizábal et al. 2013). Species associated with the rhizome and the pseudostem of *Heliconia* inflorescences were reported in Colombia (Aristizábal et al. 2013).

Two predators of the family Staphylinidae, *Tachyporus* sp. and *Paederomimus* sp., were identified. In *H. bihai*, species of this family of beetles were recorded as top predators in the food chain formed inside the bracts, feeding on Diptera larvae (Frank & Barrera 2010). This family is particularly interesting from the functional perspective because they can be detritivorous, herbivorous, mycophagous, or predacious (Clough et al. 2007). These insects occupy different habitats, such as fallen parts of plants, tree holes, debris, vegetation, flowers, etc. (Honek & Kocian 2003; Greeney 2001; Sanabria et al. 2008; Brunke & Marshall 2011).

Histeridae also occupy different types of habitats such as manure, carrion, and decomposing plant material (Hawkeswood 2006). The adults also have been found in other Eryngium (Apiaceae) phytotelmata in the subtropical and temperate regions of Argentina (Campos & Fernández 2011). However, these species represent a small part of the species complex.

Among the herbivores, the family Curculionidae has the largest number of records of insects found in phytotelma environments (Campos & Fernández 2011); it was found in inflorescences of *Heliconia* spp. cultivated in Colombia (Aristizábal et al. 2013), as well as in bromeliad species in Argentina (Montero et al. 2010) and in Brazil (Mestre et

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**Table 3.** Mean occurrence of key species and richness and abundance of insects in the cultivated area when inflorescence size (number of bracts) is compared using the Mann-Whitney test.

| No. of bracts/inflorescence | Cultivated | Mann-Whitney |
|-----------------------------|------------|--------------|
|                             | Average ± SD |               |
|                             | 2          | 3            | 4            | 5            |
| Pelosoma lafertei           | 0.3 ± 0.5  | 3.5 ± 6.4   | 439          | −2.8         | 0.004        |
| Paederomimus sp.            | 0.03 ± 0.1 | 0.9 ± 1.9   | 465.5        | −3.2         | 0.001        |
| Richness                    | 0.6 ± 0.7  | 1.2 ± 1.0   | 472          | −2.3         | 0.017        |
| Abundance                   | 0.8 ± 1.2  | 5.0 ± 8.3   | 313          | −4.0         | 0.000        |
| Tachyporus sp.              | 0.03 ± 0.1 | 0.3 ± 0.6   | 1098         | −2.6         | 0.007        |
| Pelosoma lafertei           | 0.7 ± 2.4  | 3.5 ± 6.4   | 843          | −3.1         | 0.001        |
| Paederomimus sp.            | 0.2 ± 0.1  | 0.9 ± 1.9   | 652          | −3.8         | <0.0001      |
| Richness                    | 0.5 ± 0.7  | 1.2 ± 1.0   | 598.5        | −4.1         | <0.0001      |
| Abundance                   | 1.4 ± 2.8  | 5.2 ± 8.3   | 598.5        | −4.1         | 0.000        |
| Pelosoma lafertei           | 1.1 ± 3.2  | 3.5 ± 6.4   | 707.5        | −2.9         | 0.003        |
| Abundance                   | 2.2 ± 3.8  | 5.2 ± 8.3   | 666          | −2.9         | 0.002        |

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**Table 4.** Values (average ± SD) of pH, volume (mL), and temperature (°C) in the inflorescence groups with 2, 3, 4, and 5 *Heliconia bihai* bracts from cultivated and uncultivated areas in Camaragibe-PE.

| No. of bracts/inflorescence | Cultivated | Uncultivated |
|-----------------------------|------------|--------------|
|                             | pH         | Temperature (°C) | Volume (mL) |
|                             | 2          | 3            | 4            | 5            | 2          | 3            | 4            | 5            |
| 2                           | 6.5 ± 1.3  | 27.3 ± 2.9   | 2.0 ± 1.6    |               | 6.5 ± 1.1  | 25.9 ± 1.5   | 5.8 ± 1.1    |               |
| 3                           | 6.8 ± 1.4  | 28.4 ± 2.3   | 1.5 ± 0.9    |               | 6.6 ± 1.0  | 26.3 ± 1.8   | 4.7 ± 0.6    |               |
| 4                           | 7.1 ± 1.3  | 28.9 ± 2.4   | 1.5 ± 1.1    |               | 5.8 ± 1.4  | 25.2 ± 2.8   | 4.7 ± 0.5    |               |
| 5                           | 7.2 ± 1.7  | 28.7 ± 2.1   | 1.6 ± 1.2    |               | 6.6 ± 0.8  | 27.0 ± 1.6   | 4.5 ±0.7     |               |
The environmental condition seems to guarantee the occurrence of *P. lafertei*. The acidity indicates that the environment is rich in organic matter, which is the main food source of phytotelma fauna (Nishadh & Anoopdas 2014) supporting *P. lafertei*, which is a detritivorous species. The higher water volume in the uncultivated area could allow the occurrence of aquatic *Elmidae* species (Manzo 2005). Bracts with lower temperatures guarantee the occurrence of *Paederomimus* sp. in the uncultivated area. *Paederomimus* sp. and *Tachyporus* sp. are predators and consume Diptera larvae, which are extremely abundant in the bracts (Frank & Barrera 2010).

Our studies demonstrated a greater richness of species associated with the uncultivated area. Also, bract features such as greater volume, higher acidity, and lower temperature seem to influence the Coleoptera assemblage in *H. bihai* bracts. This is important because not only does the uncultivated area contribute to the maintenance of diversity, but these plants may also represent a source of species potentially harmful to crops.

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