

## Natural history of *Gynaikothrips uzeli* (Thysanoptera, Phlaeothripidae) in galls of *Ficus benjamina* (Rosales, Moraceae)

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**ABSTRACT.** Galls induced by thrips are simple structures when compared to those of other groups of arthropods, and little is known regarding many of their aspects. This study aimed to investigate aspects of the natural history of *Gynaikothrips uzeli* Zimmermann, 1900 in galls of *Ficus benjamina* L., 1753 using seasonal sampling (summer and winter). Twenty trees were sampled and divided into quadrants. From each of them, five galls were collected, forming a total of 400 galls per collection. Thrips showed greater abundance at higher temperatures (25.7°C) and no precipitation. Sex ratio was biased towards females (0.022 males per female), pointing to an inbred mating structure. Arthropod fauna associated with galls was more abundant (N=798) in winter, and it included representatives of the orders Hemiptera, Hymenoptera, Araneae, Coleoptera, Neuroptera, Psocoptera, Thysanoptera, Diptera and Blattodea.

**KEYWORDS.** Leaf galls, natural enemies, sex ratio, thrips.

**RESUMO.** História natural de *Gynaikothrips uzeli* (Thysanoptera, Phlaeothripidae) em galhas de *Ficus benjamina* (Rosales, Moraceae). As galhas induzidas por tripses são estruturas simples quando comparadas com outros grupos de artrópodes e pouco conhecidas quanto a diversos aspectos. Este trabalho teve como objetivo verificar aspectos da história natural de *Gynaikothrips uzeli* Zimmermann, 1900 em galhas de *Ficus benjamina* L., 1753 em coletas sazonais (verão e inverno). Vinte árvores escolhidas, aleatoriamente, foram divididas em quatro quadrantes, e em cada um deles coletadas cinco galhas, perfazendo um total de 400 galhas por coleta. Os tripses apresentaram uma maior abundância em temperaturas mais elevadas (25,7°C) e sem precipitação. A razão sexual (0,022 machos) encontrada foi baixa (tendenciosa às fêmeas), apontando para uma estrutura endogâmica de acasalamento. A fauna associada de artrópodes presentes nas galhas foi mais abundante (N=798) no período de inverno e incluiu representantes das ordens Hemiptera, Hymenoptera, Araneae, Coleoptera, Neuroptera, Psocoptera, Thysanoptera, Diptera e Blattodea.

**PALAVRAS-CHAVE.** Galha foliar, inimigos naturais, razão sexual, tripses.

The Thysanoptera, or thrips, exhibit a remarkable dietary diversity, encompassing predatory, mycophagous and phytophagous species (TERRY, 1997; MOUND, 2005; ROTENBERG & WHITFIELD, 2010). Their phytophagous diet can cause damage to several parts of plants, and many species are gall inducers – among these, the genus *Gynaikothrips* Zimmermann, 1900. Association between gall-forming thrips and host plants is a specialized trophic strategy and, in some cases, it is also species-specific (ANANTHAKRISHNAN, 1993).

*Ficus benjamina* L., 1753 is widely used in urban tree planting (SANTOS & RAMALHO, 1997) and it has been used in studies due to its antibacterial potential (RESCHKE *et al.*, 2007). The galls induced by *Gynaikothrips uzeli* Zimmermann, 1900 in *F. benjamina* are characterized by the folding of the leaf due to the insects feeding on the leaf tissue (RETANA-SALAZAR & Sánchez-CHACÓN, 2009), bringing this species into a pest status. Besides providing a food source, the galls also serve as shelter and protection from predators, as well as shelter to inquiline species belonging to different

arthropod orders (MOUND *et al.*, 1995; GONÇALVES-ALVIM & FERNANDES, 2001).

Despite its cosmopolitan distribution and economic importance, a few studies on the biology of *Gynaikothrips uzeli* have been conducted. Among them, we highlight FARONG *et al.* (1995), who studied biological characteristics (rate of emergence, average amount of spawned egg, rate of hatchability, and span of life) of *G. uzeli* in Jinggu, China; HELD *et al.* (2005), who conducted a study on the distribution and a revision of the biology of *G. uzeli* in the southwestern United States; and DE MELO *et al.* (2013), who accessed the behavior and predatory potential of *Androthrips ramanchandrai* Karny, 1926 to *G. uzeli*.

Therefore, this research aims to characterize the galls induced by *Gynaikothrips uzeli* in *Ficus benjamina* through seasonal sampling (summer and winter) regarding the abundance of adult and immature inducer individuals, the sexual rate and the relation with the leaf area and with the gall stage. Furthermore, we looked for the effects of average

temperature and precipitation on *G. uzeli* abundance, and we determined the fauna associated with the galls.

## MATERIALS AND METHODS

Galls were collected from trees of *Ficus benjamina* (Moraceae) in Jequié, state of Bahia (13°52'4.9"S, 40°04'26.7"W), during the period of February 1st to 10th (summer), and from July 25th to August 3rd, 2012 (winter). In order to collect galls, 20 trees with different canopy sizes were selected, and each one was divided into four quadrants. Then, five galls were sampled from each quadrant, totalizing 400 galls per collection period (800 in total).

The galls were placed individually into plastic bags and transported to the Laboratory of Insect Biology (LABI) at Universidade Estadual do Sudoeste da Bahia (UESB), where they were analyzed. Each gall was washed internally with 70% ethanol, and the obtained material was put into a Petri dish. The material was then inspected under a stereomicroscope to search for adults, immature (larval, pre-pupal and pupal stages) and associated arthropods, which were all counted. Individuals were fixed in absolute ethanol, placed in plastic vials of 2 mL, and identified to the most specific taxonomic level with support of experts. For identifying the sex of *Gynaikothrips uzeli*, we took into account the sexual dimorphism of the species, in which adult males are smaller than females (GARITA-CAMBRONERO & LIZANO-FALLAS, 2006). In cases which sex identification was ambiguous, specimens were dissected in order to inspect the genitalia.

In order to establish the relation between gall stages and abundance of individuals, galls were collected and classified into three stages considering leaf texture and folding capacity: young gall leaves, which were flexible and partially folded; mature gall leaves, which were minimally flexible and folded; and old galls, which had inflexible, overdried and twined leaves. Leaf area was calculated using the formula proposed by AMAZONAS *et al.* (2008):  $A_E = \pi.ab = (3.14.CW)/4 = 0.785.CW$  (C, length and W, width).

The sex ratio of adult thrips was determined as the ratio between the total number of males and number of females (QUICKE, 1997). The analysis of variance (ANOVA) was performed to verify a significant difference between the averages of leaf area, abundance of *Gynaikothrips uzeli*, general associated fauna, *Montandoniola moraguesi* Puton, 1896, and *Androthrips ramachandrai* Karny, 1926 in different gall stages of summer and winter periods. Pearson correlations were also calculated in order to test

the correlation between leaf area with following variables: total of *G. uzeli*, *A. ramachandrai*, *M. moraguesi* and other associated arthropods and the program SAS/STAT version 8.0 (SAS INSTITUT, 2003) was used.

## RESULTS AND DISCUSSION

Results obtained from the analysis of 800 galls induced by *Gynaikothrips uzeli* in *Ficus benjamina* are summarized in Tab. I.

The data revealed a greater abundance of *Gynaikothrips uzeli* in summer compared to winter. This decrease is partially due to the influence of the temperature. The summer average temperature was about 25.7°C, while in winter the average temperature was about 22.0°C. There was no rainfall during the period of collections. Under those conditions of high temperature and decreased precipitation the biological cycles are usually accelerated, promoting the population growth. In the analysis performed by PAINE (1992), e.g. developmental time (from egg to adult) in *Gynaikothrips ficorum* Marchal, 1908 varied from 48.99 days at 15°C to 16 days at 30°C (thermal limits were 12°C and 35°C). Thus, the abundance of *G. uzeli* in summer may be a consequence of a shorter life cycle and a higher chance of overlapping generations. Moreover, heavy rains could carry individuals away from galls when the latter are not completely formed, or inundate them, when they are already formed.

Another important factor is that, during the winter, the associated fauna was more abundant and diverse. Furthermore, *Gynaikothrips uzeli* predators were present in greater quantity, mainly *Montandoniola moraguesi* (Tab. II), resulting in a sharp decrease in the number of immature individuals (Tab. I).

According to ANANTHAKRISHNAN (1993), besides the galling species, galls harbor other secondary species that may be inquilines, parasites, predators, or occasional visitors. Among predators, there are hemipterans, other thrips, ants, flies, wasps, neuropteran larvae, mites, spiders and pseudoscorpions (MONTEIRO & MOUND, 2012).

In the current work, we found specimens of nine orders of arthropods (Tab. II), which shows that the ecological interactions involving galls induced by *Gynaikothrips uzeli* are quite diverse.

Among specimens of the gall associated fauna, we highlight *Montandoniola moraguesi* and *Androthrips ramachandrai* due to their abundance and previously known predatory relationship with *G. uzeli* (DOBBS & BOYD JR., 2006; DE MELO *et al.*, 2013).

Tab. I. Data from the analysis of 800 galls of *Ficus benjamina* L., 1753 induced by *Gynaikothrips uzeli* Zimmermann, 1900 with 400 galls collected in summer and 400 collected in winter.

|        | Adults |      | Total | Immature | Total<br><i>Gynaikothrips uzeli</i><br>(Zimmermann, 1900) | Associated<br>fauna |
|--------|--------|------|-------|----------|---|---------------------|
|        | Female | Male |       |          |   |                     |
| Summer | 4447   | 77   | 4524  | 18201    | 22725   | 369                 |
| Winter | 4843   | 93   | 4936  | 1302     | 6238  | 798                 |
| Total  | 9290   | 170  | 9460  | 19503    | 28963   | 1167                |

Tab. II. Taxonomic classification of the fauna associated with galls of *Ficus benjamina* L., 1753 induced by *Gynaikothrips uzeli* Zimmermann, 1900 during collections in summer and winter times, indicating the numbers of immature (IM) and adult individuals (AD).

| Orders       | Taxon             | Summer   |     | Winter |     |
|--------------|-------------------|--|-----|--------|-----|
|              |                   | IM   | AD  | IM     | AD  |
| Araneae      | Thomisidae        | -  | -   | -      | 2   |
|              | Sicariidae        | -  | -   | -      | 1   |
|              | -                 | 5  | -   | 23     | -   |
| Blattodea    | -                 | -  | -   | 1      | -   |
| Coleoptera   | -                 | -  | 1   | -      | 7   |
| Diptera      | -                 | -  | -   | -      | 1   |
| Hemiptera    | Aleyrodidae       | 14   | -   | 11     | -   |
|              | Anthocoridae      | 124  | 47  | 405    | 91  |
| Hymenoptera  | Formicidae        | <i>Montandoniola moraguesi</i> (Puton, 1896)     | -   | 1      | -   |
|              |                   | <i>Nylanderia</i> sp.                            | -   | 5      | -   |
|              |                   | <i>Crematogaster</i> sp.                         | -   | 1      | -   |
|              |                   | <i>Cardiocondyla</i> sp.                         | -   | 5      | -   |
|              |                   | -  | -   | -      | 39  |
| Neuroptera   | Eulophidae        | -  | -   | 5      | 1   |
|              | Chrysopidae       | 1  | 1   | -      | -   |
|              | Hemerobiidae      | -  | -   | 1      | -   |
| Psocoptera   | Ectopsocidae      | -  | 7   | -      | -   |
|              |                   | <i>Ectopsocus</i> sp. 1                          | -   | -      | -   |
|              |                   | <i>Ectopsocus</i> sp. 2                          | -   | -      | -   |
| Thysanoptera | Pseudocaeciliidae | <i>Pseudocaecilius citricola</i> (Ashmead, 1879) | -   | -      | -   |
|              | Thripidae         | <i>Selenothrips rubrocinctus</i> (Giard, 1901)   | -   | -      | -   |
|              | Phlaeothripidae   | <i>Androthrips ramachandrai</i> (Karny, 1926)    | 21  | 144    | 19  |
| TOTAL        |                   | 164  | 205 | 472    | 326 |

*Montandoniola moraguesi* was the most abundant insect in the analyzed galls (Tab. II). It is recognized as an effective predator of various species of thrips and it has been used in Hawaii as a biological control agent of *Gynaikothrips uzeli* since 1966 (DOBBS & BOYD JR., 2006). Although it is considered native from Southeast Asia, this species has already reached cosmopolitan status, being reported in some countries in Africa, Asia, Europe, the Americas and in Australia (FUNASAKI, 1966; DOBBS & BOYD JR., 2006; PLUOT-SIGWALT *et al.*, 2009; SEPÚLVEDA CANO *et al.*, 2009). In a study with *Gynaikothrips ficorum* performed by FUNASAKI (1966), it was reported that the nymphs in the 1<sup>st</sup> and 2<sup>nd</sup> instars of *M. moraguesi* feed on eggs and larvae of *G. ficorum*, and nymphs in the 3<sup>rd</sup>, 4<sup>th</sup> and 5<sup>th</sup> instars feed on thrips at any stage of development, including adults. DOBBS & BOYD JR. (2006) also reported *M. moraguesi* as a predator of *Androthrips ramachandrai*.

Another predator of thysanopterans which was abundant in the galls was *Androthrips ramachandrai* (Tab. II). This species was firstly described in India, and it was already reported in Trinidad and Tobago, Belize, Puerto Rico, United States (BOYD JR. & HELD, 2006), Mexico (CAMBERO-CAMPOS *et al.*, 2010), Colombia (SEPÚLVEDA CANO *et al.*, 2009), Argentina (BORBÓN & AGOSTINI, 2011) and Brazil (CAVALLERI *et al.*, 2011) as a predator of various galling thrips, among them *Gynaikothrips uzeli* (BOYD JR. & HELD, 2006). Immature and adult stages of *A. ramachandrai* were observed in laboratory feeding on immature stages of *G. uzeli* and *G. ficorum* (CAVALLERI *et al.*, 2011). This observation was confirmed by SEPÚLVEDA CANO *et al.* (2009), who observed that whenever *A. ramachandrai* populations increased, *G. uzeli* populations decreased. This is also supported by data obtained in the current study (Tabs I, II), given that a

decrease in *G. uzeli* numbers was observed in winter, when *A. ramachandrai* numbers increased.

The other sampled orders may eventually establish a predatory relationship with *G. uzeli*, and an inquiline or visitation to the formed galls in *F. benjamina*. KAUFMANN *et al.* (1991) and NADEL *et al.* (1991), e.g. reported that Formicidae and Coleoptera are related to seed dispersion in several species of *Ficus*.

We would also like to highlight that this is the first report of specimens of the orders Araneae (Sicariidae), Neuroptera (Hemerobiidae) and Psocoptera associated with galls of *Ficus benjamina*.

Tab. III presents the average abundance of *G. uzeli*, general associated fauna, *Montandoniola moraguesi* and *Androthrips ramachandrai* in different stages of galls collected in summer and winter. Although mature galls had the highest average – which agrees with the hypothesis proposed by ANANTHAKRISHNAN & RAMAN (1989) that mature galls would have larger populations, while galls in formation would have smaller populations – the ANOVA was generally non-significant ( $p > 0.05$ ), thus indicating no difference between gall averages. However, the relationships between gall stage and leaf area ( $F = 11.86$ ;  $df = 2.399$ ;  $p < 0.001$ ), and total of *G. uzeli* ( $F = 17.51$ ;  $df = 2.399$ ;  $p < 0.001$ ) in the summer, and the relationships between gall stage and leaf area ( $F = 15.83$ ;  $df = 2.399$ ;  $p < 0.001$ ) and females ( $F = 4.37$ ;  $df = 2.399$ ;  $p = 0.013$ ) in the winter were all statistically significant and indicated that mature galls were different from the others. The correlations considering leaf area were low (Tab. IV). These data indicate both larger and smaller leaves can harbor populations of various sizes.

Sex ratio observed in *G. uzeli* (Tab. V) suggested a bias toward females. According to CRESPI (1993), the way

Tab. III. Number of galls, average and standard deviation of leaf area and average of the abundance of *Gynaikothrips uzeli* Zimmermann, 1900, general associated fauna, *Montandoniola moraguesi* Puton, 1896 and *Androthrips ramachandrai* Karny, 1926 in different stages of galls collected in summer and winter.

|   | Summer |        |        | Winter |        |        |
|---|--------|--------|--------|--------|--------|--------|
|   | Young  | Mature | Old    | Young  | Mature | Old    |
| Number of galls                             | 78     | 296    | 26     | 103    | 258    | 39     |
| Leaf Area                                   | 7.64 ± | 9.39 ± | 6.95 ± | 5.44 ± | 7.46 ± | 4.31 ± |
| <i>Gynaikothrips uzeli</i> Zimmermann, 1900 | 3.81   | 3.46   | 2.47   | 3.71   | 3.07   | 1.93   |
| Associated fauna (general)                  | 18.59  | 69.1   | 31.58  | 9.58   | 18.79  | 10.31  |
| <i>Montandoniola moraguesi</i> Puton, 1896  | 0.55   | 1.6    | 0.69   | 1.28   | 2.36   | 1.36   |
| <i>Androthrips ramachandrai</i> Karny, 1926 | 0.12   | 0.53   | 0.19   | 0.44   | 1.57   | 1.1    |
|   | 0.21   | 0.48   | 0.23   | 0.8    | 0.43   | 0.08   |

Tab. IV. Pearson correlations between Leaf Area (LA) and total of *Gynaikothrips uzeli* Zimmermann, 1900 (TOT), *Androthrips ramachandrai* Karny, 1926 (AR), *Montandoniola moraguesi* Puton, 1896 (MM) and other associated arthropods (OT), collected in summer, winter and both periods (summer + winter) (\*, significant at 5%; \*\* significant at 1%; ns, non significant).

|    | TOT            | AR                   | MM             | OT                  |         |
|----|----------------|----------------------|----------------|---------------------|---------|
| LA | <b>0,321**</b> | -0,057 <sup>ns</sup> | <b>0,151**</b> | <b>0,145**</b>      | Summer  |
| LA | <b>0,147**</b> | 0,053 <sup>ns</sup>  | <b>0,190**</b> | 0,069 <sup>ns</sup> | Winter  |
| LA | <b>0,306**</b> | 0,016 <sup>ns</sup>  | <b>0,115**</b> | 0,048 <sup>ns</sup> | General |

Tab. V. Sample size (N) and sex ratio (males per female) of *Gynaikothrips uzeli* Zimmermann, 1900 collected in summer, winter and total (both periods), by gall stage and general data.

|        | N    | Gall stage |        |       | General        |
|--------|------|------------|--------|-------|----------------|
|        |      | Young      | Mature | Old   |                |
| Summer | 4524 | 0.016      | 0.052  | 0.006 | 0.023 ± 0.0944 |
| Winter | 4936 | 0.02       | 0.022  | 0.029 | 0.022 ± 0.1016 |
| Total  | 9460 | 0.018      | 0.037  | 0.018 | 0.022 ± 0.098  |

in which galls are founded has fundamental importance in understanding sex allocation in thrips, which is reflected in the sex ratio.

Based on studies of TREE & WALTER (2009), who observed that galls of *Gynaikothrips ficorum* in *Ficus microcarpa* L.f 1782 are induced by one or occasionally two females, and field observations in the current study, we believe that the formation of galls of *G. uzeli* is also likely to be carried out by one or a few females. Those founding females possibly mate with a male sibling inside the gall from which they migrated. Such data is ratified by the low heterozygosity observed in a population genetics analysis of *G. uzeli* in galls of *Ficus benjamina* (MASCARENHAS *et al.*, 2015).

TREE & WALTER (2009) also observed that *Gynaikothrips australis* Bagnall, 1929 leaves and returns to its galls throughout the day. This temporary dispersion could also occur in *G. uzeli*. Such flow could be reflected in the sex ratio, as the adults are the ones involved, but their gender is not clearly defined. Another factor that may explain the low amount of males in galls is that male larvae could be present in the galls but were not detected, since immature individuals were not sexed. Furthermore, the possible presence of *Wolbachia* Hertig, 1936 bacteria may justify the low amount of males found, given that it has already been reported to affect the reproductive process of some species in Thysanoptera (KUMM & MORITZ, 2008).

Data from this study indicate a greater abundance of *G. uzeli* at higher temperatures, and the low number of males may be an indication that one or a few fertilized females are responsible for the foundation process of galls and that inbreeding is a common reproductive process in *G. uzeli*.

Moreover, sex ratio may be further influenced by adult male dispersion throughout the day and by females infected by *Wolbachia* bacteria. The arthropod fauna found within galls of *Ficus benjamina* was diverse, demonstrating that this was a favorable microhabitat for numerous ecological interactions. This study also provided the first records of specimens in the orders Psocoptera, Neuroptera (Hemerobiidae), and Araneae (Sicariidae) associated with galls induced by *G. uzeli*. Additional studies on the pattern of gall foundation, as well as studies on the relatedness among individuals from the same gall may help to elucidate aspects related to reproductive strategies and female dispersal in this species.

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