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**Primary parasitoids and hyperparasitoids associated with the striped mealybug *Ferrisia virgata* (Hemiptera: Pseudococcidae) infesting the ornamental host plant, *Leucaenia leucocephala* in Giza region, Egypt**

Angel, R. Attia<sup>1</sup>; Kamal, T. Awadallah<sup>2</sup> and Mohamed, M. Abo-Setta<sup>1</sup>

<sup>1</sup>Plant Protection Research Institute, Agricultural Research Center, Dokki, Giza, Egypt.

<sup>2</sup>Biological Control Lab., Faculty of Agriculture, Cairo University, Giza, Egypt.

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**Abstract**

A survey of the parasitoids and hyperparasitoids associated with the striped pseudococcid mealybug, *Ferrisia virgata* (Cockerell) (Hemiptera: Pseudococcidae) infesting the ornamental host plant, *Leucaenia leucocephala* was carried out during October of the three years 2017, 2020 and 2023. Each October sample was presented by 400 mummified individuals. They were sorted as mummified third nymphal instar, female, and gravid female. The survey revealed the presence of three primary endoparasitoids and two hyperparasitoids. The two primary parasitoids, *Blepyrus insularis* (Cameron) and *Acerophagus gutierreziae* Timberlake (Are solitary and the third *Acerophagus* sp. is gregarious. The two hyperparasitoids, *Chartocerus subaeneus* (Forster) (Hymenoptera: Signiphoridae) and *Prochiloneurus aegyptiacus* (Mercet) (Hymenoptera: Encyrtidae) were presented. The first one is gregarious, while the second is solitary. *B. insularis* and *C. subaeneus* were recorded as the most dominant species over the three years. *P. aegyptiacus* was recorded only in October 2023. Its sex ratio (Females/total) varied according to the mummified host stage. It was 21.05, 58.33 and 86.15% for the considered stages, respectively. This was attributed to in reverse the host size and subsequently the nutritional value available to the parasitoid as a survival technique. During 2023, *B. insularis* was the dominant followed by *P. aegyptiacus*, *C. subaeneus*, *Acerophagus* sp., and *A. gutierreziae* at 59.25, 30.00, 18.33, 6.67 and 5.75%, respectively. The mean number of *C. subaeneus* that emerged from one mummified host stage were  $2.46 \pm 0.50$ ,  $3.90 \pm 0.82$ , and  $6.58 \pm 0.77$  from a mummified host of a third nymph, female and gravid female, respectively. A number of *Acerophagus* sp. that emerged from one mummy of a mummified adult female ranged from 2-4 individuals (mean of  $3.25 \pm 0.75$ ), compared with 6-8 individuals (Mean of  $7.11 \pm 0.75$ ) from a mummified gravid female.

## Introduction

Family Pseudococcidae is the second largest family of Coccoidea, with about 2000 spp.; belonging to 280 genera worldwide (Ben-Dov *et al.*, 2010). In Egypt, 50 species belonging to 29 genera are specified (Mohammad and Ghabbour, 2008).

*Ferrisia virgata* (Cockerell) (Hemiptera: Pseudococcidae) is a highly polyphagous mealybug; being reported on host-plants belonging to over 203 genera in 77 families and can damage many crops. It is also known to transmit plant badnavirus diseases in cocoa and black pepper (Watson, 2016). In Egypt, it was recorded for the first time, attacking *Acalypha* shrubs and a few ornamental plants growing in gardens at Port Said and on a number of host plants at Ismailia and Suez governorates. In 1941, heavy infestations were noticed at Meadi, Hawamdieh and Nag-Hammadi attacking a great number of plants (Hosny, 1943).

In Egypt, the population density of *F. virgate* was investigated by previous authors on different host plants (Ammar *et al.*, 1979; Attia, 1997; Ata *et al.*, 2019; Youssef, *et al.*, 2021 and Amer *et al.*, 2023). As for the primary parasitoids and hyperparasitoids associated with mealybugs, belonging to families Aphelinidae, Encyrtidae, Platygasteridae, and Signiphoridae (Hymenoptera) are reported to attack different species of mealybugs (Attia, 2012). In Egypt, the primary parasitoid, *Blepyrus insularis* (Cameron) (Hymenoptera: Encyrtidae) was first reported by Attia in 1997 and thereafter by Amer *et al.* in 2023.

Concerning the hyperparasitoids, *Chartocerus subaeneus* (Foerster) (Hymenoptera: Encyrtidae) was previously recorded on *Tetracnemoidea peregrine* (Compere) (Rosen *et al.*, 1992), on *Aenasius arizonensis* (Girault) (Attia *et al.*, 2017) and on *B. insularis* (Amer *et al.*, 2023). Also, the hyperparasitoid, *Prochiloneurus aegyptiacus* (Mercet) (Hymenoptera: Encyrtidae) was

recorded on the two primary parasitoids *Anagyrus* sp., and *Gyranusoidea indica* Shafee, Alam and Agarwal (Awadallah *et al.*, 1999) and *A. arizonensis* (Attia *et al.*, 2017).

The present work aims to study the survey of the parasitoids and hyperparasitoids associated with the striped pseudococcid mealybug, *F. virgata* infesting the ornamental host plant, *Leucaenia leucocephala* in Giza, Egypt.

## Materials and methods

The study was conducted in the Giza region in October of the years 2017, 2020, and 2023 when a high infestation of *F. virgata* was noticed on the ornamental plant, *L. leucocephala*. Four hundred mummies each year were collected. They were obtained after one week, and the emerging parasitoids were examined weekly until the emergence of all parasitoids. Each mummy contained parasitoid pupa was isolated in a small glass tube covered with cotton until emergence. Hosts were sorted as mummified third nymphal instar, female, and gravid female.

Emerged parasitoids and hyperparasitoids were counted as their species in each of the mummified stages. These parasitoids and hyperparasitoids species were previously identified in Natural History Museum by Dr. John Noyes (Awadalla *et al.*, 1999 and Attia and Awadallah, 2016). They were *B. insularis*, *Acerophagus gutierreziae* Timberlake (Primary ones) and *C. subaeneus*. *P. aegyptiacus* (Hyperparasitoids).

The occurrence rate of each parasitoid or hyperparasitoid was calculated as follows: No of parasitic or hyperparasitic host mummies / Total No. host mummies (400) X 100. Mean individuals emerged from one mummy of *Aceropagus* sp. collected during 2017 per female and gravid female was determined. The same was conducted for *C. subaeneus* for the same stages during 2020. This time was the optimum when the highest parasitism occurred.

## Results and discussion

During the present study, three primary, endoparasitoids and two hyperparasitoids were recorded associated with mummies of the striped mealybug, *F. virgate*. Two of the primary parasitoids (*B. insularis* and *A. gutierreziae*) are solitary, while the third (*Acerophagus* sp.) is gregarious. The hyperparasitoid, *C. subaeneus* was the most common and was recorded in the three years (2017, 2020, and 2023), while the hyperparasitoid, *P. aegyptiacus* was recorded only in 2023.

### 1. Primary parasitoids associated with mummies of *Ferrisia virgate* during 15<sup>th</sup> October of years 2017, 2020 and 2023:

#### 1.1. *Blepyrus insularis*:

The primary parasitoid, *B. insularis* is a solitary, endoparasitoid, and uniparental species, as most of its progeny are females with very rare numbers of males. It was recorded for the first time in Egypt in 1993 and during this period the primary parasitoid, *Leptomastix* sp. was also recorded (Awadalla *et al.*, 1999).

In the present study and in October 2017, the parasitoid, *B. insularis* emerged from the mummified hosts of third nymphal instar, adult, and gravid females of *F. virgate*, with respective numbers of 48, 121, and 71 mummies. The smallest number (48 mummies) was recorded for those of third nymphal instars while the greatest (121 mummies) was from those of adult females. The total percent of existence for this parasitoid was 60% (Table 1).

During October 2020, numbers of 21, 118, and 124 parasitoids emerged from mummies of third nymphal instar, adult, and gravid females respectively and the total occurrence rate reached 65.75% (Table 2). In October 2023, the total percent of the existence of this parasitoid decreased to 52%, with numbers of 8, 60, and 140 individuals emerging from mummies of third nymphal

instar, adult, and gravid females, respectively (Table 3).

In this concern, it was noticed that the primary parasitoid, *Leptomastix* sp. that was previously recorded together with *B. insularis* (Attia, 1997) was not surveyed in the present investigation; the state that may be referred to climatic changes or host plant or any other factor.

#### 1.2. *Aceropagus gutierreziae*:

Attia and Awadallah (2016) previously recorded this parasitoid, developing solitary in nymphs of the cotton mealybug, *Phenacoccus solenopsis* Tinsley (Hemiptera: Pseudococcidae). In the present study, *A. gutierreziae* emerged from the mummified hosts of the third nymphal instar of *F. virgate*, with respective numbers of 31 and 38 individuals during the 2017 and 2020 years; their respective percentages of its occurrence were 7.75 % and 9.50 % during the 2016 and 2020 years (Tables 1 and 2). However, no parasitic individuals were recorded in the year 2023 (Table 3).

#### 1.3. *Acerophagus* sp.:

This gregarious species was identified at the generic level only. In 2017 (Table 1), this parasitoid emerged from mummified hosts of adult and gravid females of *F. virgate*; being represented by the numbers of 12 and 36 individuals. The percentage of its existence was 12%. However, in 2020 and 2023; this percentage decreased to 4.5% and 3.5% (Tables 2 and 3).

**Number of *Acerophagus* sp. / mummy:** Data represented in Table (4) indicated that the number of the parasitic individuals of the primary gregarious parasitoid, *Acerophagus* sp. that emerged from one mummy of mummified adult female ranged from 2 - 4 individuals with a mean of  $3.25 \pm 0.75$ , as opposed to a range of 6 - 8 individuals with a mean of  $7.11 \pm 0.75$  from a mummified gravid female.

The total percent of the existence of the three secured primary parasitoids that

emerged from the different stages of its host *F. virgate* was respective 79.75, 79.75 and 55.5% during the three years of study, representing the lowest rate in 2023 owing to the high existence of the associated hyperparasitoids (Tables 1,2 and 3).

## 2. Hyperparasitoids associated with the previous primary parasitoids:

Two hyperparasitoids were secured during the present investigation; *C. subaeneus* and *P. aegyptiacus* emerging from the primary parasitoids associated with *F. virgate* during October of years 2017, 2020, and 2023 for the first one and during October of 2023 for the second. The biological study of *C. subaeneus* was investigated by Rosen et al. (1992) who mentioned that this hyperparasitoid is deuterotokous (Virgin females give rise to female progeny, and males are rather rare) and from random samples of Lab cultures, the percent of males reached 5.25%.

During this investigation, (Tables 1,2,3), the hyperparasitoid, *C. subaeneus* was recorded during the three years 2017, 2020, and 2023, with an existence percentage of 20.25% in both 2017 and 2020, while in the third year, this percentage decreased to 14.50%. In this year (2023), the hyperparasitoid, *P. aegyptiacus* was also secured with a percent of 30 %. The total existence rate of the two hyperparasitoids together was 44.5%, leading to a reduction in the existence percent (52%) of the primary parasitoid, *B. insularis* in October 2023.

In 2023, the competition between the solitary hyperparasitoid, *P. aegyptiacus* and the gregarious dominant one *C. subaeneus* was obvious. In 2017 and 2020, *C. subaeneus* was only secured and its existence rate reached 20.25% in both seasons. However, in 2023, when *P. aegyptiacus* was also secured, this rate decreased to 14.5%. It is to be noted that *P. aegyptiacus* is larger in size (More than two times compared with *C. subaeneus*) and may be able to compete.

In this concern, Attia et al. (2017) recorded the three hyperparasitoids; *C. subaeneus*, *P. aegyptiacus*, and *Pachyneuron* sp. associated with the primary parasitoid, *A. arizonensis* associated with *P. solenopsis* on different host plants during June and July 2016. The common hyperparasitoid, *C. subaeneus* reduced the population of *A. arizonensis*, to 18 %. The highest count was located on the host plant, *Lantana camara*, while the lowest was on *Hibiscus rosa – sinensis*. The other two hyperparasitoids; *P. aegyptiacus* and *Pachyneuron* sp. were represented by rear numbers. The host plant may play in role in this concern.

**Number of *Chartocerus subaeneus* / mummy:** As indicated in Table (4), the number of the gregarious hyperparasitoid, *C. subaeneus* that emerged from one mummified host of third nymphal instar ranged from 2-3 individuals with a mean of  $2.46 \pm 0.50$ , and from 3-5 individuals with a mean of  $3.90 \pm 0.82$  from a mummified host of the adult female. In the case of a mummified host of gravid females, the number ranged from 6-8 individuals with a mean of  $6.58 \pm 0.77$ . In this concern, Attia et al. (2017) recorded a mean number of  $3.95 \pm 0.22$  individuals/ mummy emerged from one mummified cotton mealybug *P. solenopsis* adult female, as opposed to  $9.05 \pm 0.60$  individuals/mummy from those who emerged from mealybug gravid female.

A greater mean number of individuals ( $9.05 \pm 0.60$ ) emerged from a *P. solenopsis* mummified host of gravid females (Attia et al., 2017) than that ( $6.58 \pm 0.77$ ) emerged in the present investigation from the same stage of *F. virgate* may be due to the larger size of gravid female in cotton mealybug than in the striped mealybug.

## 3. Sex ratio among the progeny of the hyperparasitoid *Prochiloneurus aegyptiacus*:

As shown in Table (5) the sex ratios of the hyperparasitoid, *P. aegyptiacus*

emerged from the mummified host of the third nymphal instar, adult female, and gravid female for 2023 years varied completely. Females constituted respectively 21.05, 58.33, and 86.15, as opposed to the percent of 78.95, 41.67, and 13.85 for males. The highest percentage of females was recorded for the gravid females, while the lowest was

for the third nymphal instar. The vice versa occurred with the percent of males. This variation in sex ratios could be referred to as the size of the host; being the biggest in the case of gravid females compared to that in the third nymphal instars or adult females. The total percentage of males was 32.50% opposed to 67.50 for females (Table 5).

**Table (1): Primary and secondary parasitoids associated with the pseudococcid *Ferrisia virgata* infesting the ornamental host plant *Leucaenia leucocephala* in October 2017 in Giza region.**

Mummified host of <i>Ferrisia virgata</i>	Primary parasitoids				Hyperparasitoid
	Solitary		Gregarious	Total primary parasitoids	<i>Chartocerus subaeneus</i>
	<i>Blepyrus insularis</i>	<i>Acerophagus gutierreziae</i>	<i>Acerophagus</i> sp.		
Third nymphal instar	48	31	0	79	0
Adult female	121	0	12	133	45
Gravid female	71	0	36	107	36
Total	240	31	48	319	81
Percent of existence	60%	7.75%	12%	79.75%	20.25%

**Table (2): Primary and secondary parasitoids associated with the pseudococcid *Ferrisia virgata* infesting the ornamental host plant *Leucaenia leucocephala* in October 2020 in the Giza region.**

Mummified host of <i>Ferrisia virgata</i>	Primary parasitoids				Hyperparasitoid
	Solitary		Gregarious	Total primary parasitoids	<i>Chartocerus subaeneus</i>
	<i>Blepyrus insularis</i>	<i>Acerophagus gutierreziae</i>	<i>Acerophagus</i> sp.		
Third nymphal instar	21	38	0	59	11
Adult female	118	0	10	128	39
Gravid female	124	0	8	132	31
Total	263	38	18	319	81
Percent of existence	65.75	9.5	4.5	79.75%	20.25%

**Table (3): Primary and secondary parasitoids associated with the pseudococcid *Ferrisia virgata* infesting the ornamental host plant, *Leucaenia leucocephala* in October 2023 at Giza region.**

Mummified host of <i>Ferrisia virgata</i>	Primary parasitoids				Hyperparasitoids		
	Solitary		Gregarious	Total primary parasitoids	<i>Chartocerus subaeneus</i>	<i>Prochiloneurus aegyptiacus</i>	Total hyperparasitoids
	<i>Blepyrus insularis</i>	<i>Acerophagus gutierreziae</i>	<i>Acerophagus</i> sp.				
Third nymphal instar	8	0	0	8	0	19	19
Adult female	60	0	6	66	38	36	74
Gravid female	140	0	8	148	20	65	85
Total	208	0	14	222	58	120	178
Percent of existence	52%	0	3.50%	55.50%	14.50%	30%	44.50%

**Table (4): Mean numbers of emerged parasitoids from one mummy of the gregarious primary parasitoid, *Acerophagus* sp. during October 2017 and the secondary parasitoid, *Chartocerus subaenus* during October 2020.**

Year	2017		2020		
The two gregarious parasitoids	Primary parasitoid		hyperparasitoid		
	<i>Acerophagus</i> sp.		<i>Chartocerus subaenus</i>		
Mummified host of <i>Ferrisia virgata</i>	Adult female	Gravid female	Third nymphal instar	Adult female	Gravid female
Number of mummies	12	36	11	39	31
Number of emerged parasitoids	39	256	27	152	204
Mean number / one mummy	3.250	7.111	2.455	3.897	6.581
SD	0.754	0.747	0.498	0.852	0.765
Range	(2-4)	(6-8)	(2-3)	(3-5)	(6-8)

**Table (5): Numbers and percent of males and females of the hyperparasitoid *Prochilonurus aegyptiacus* on different stage of mummified host and their percent during October 2023.**

Mummified host of <i>Ferrisia virgata</i>	Male	%	female	%	Total
Mummified host of third nymphal instar	15	78.95	4	21.05	19
Mummified host of adult female	15	41.67	21	58.33	36
Mummified host of gravid female	9	13.85	56	86.15	65
Total	39	32.50	81	67.50	120

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