

# Life History, Diagnosis, and Parasitoids of *Zale phaeograpta* (Lepidoptera: Erebidae)

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## Life History, Diagnosis, and Parasitoids of *Zale phaeograpta* (Lepidoptera: Erebidae)

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ABSTRACT Lepidopteran erebid Zale phaeograpta (Hampson) has recently been found feeding on blackberry leaves (Rubus sp., cultivar 'Tupi') in two sites of Michoacan, Mexico. The life cycle and several life parameters of this insect, including parasitism, were examined in the laboratory. The egg, larva (seven instars), prepupa, pupa, and adult stages lasted 4.1, 35.9, 3.1, 14.5, and 29.1 d, respectively. Eggs are turquoise blue. The prolegs of first-instar larvae have an enlarged dark area that runs laterally. From second instar onwards, a whitish longitudinal dorsal band is observed along the entire body. Third-instar larvae wear a pair of dorsal protuberances in abdominal segment VIII. The fourth- and fifth-instar larvae are cream-colored, whereas sixth instars are entirely creamy. Pupa is covered with a waxy bloom. The apical margin of the forewings in males has two silvery bands, whereas in females, a dark band is present. Female and male pupae had the same weight (0.5 g) and measured 2.21 and 2.25 cm, respectively. The adult sex ratio of females to males was 1.1:1. The cumulative number of eggs laid by female during her lifetime was 281, with preoviposition and oviposition periods of 9.9 and 7.3 d, respectively. Maximum oviposition occurred 3.9 d after the start of oviposition (87.7 eggs per female). Larvae of Z. phaeograpta were parasitized by Microplitis sp. (Braconidae) (33%) and a currently unidentified Ichneumonidae species (9%), and pupae were parasitized by Brachymeria comitator (Walker) and another unidentified *Brachymeria* sp. (Chalcididae).

KEY WORDS Zale phaeograpta, life parameter, developmental stage, head capsule, parasitoid

The genus *Zale* (Hübner) belongs to the Ophiusini tribe, one of the seven tribes of the subfamily Erebinae (Lepidoptera: Erebidae) (Lafontaine and Schmidt 2010). This genus is widely distributed in the American continent (Lafontaine and Schmidt 2010). Its larvae are commonly known as false loopers because of the reduced or absent prolegs on abdominal segments III and IV, which cause their movements to be similar to larvae of the Geometridae family (Toimil 1987). Species of the genus *Zale* overwinter in the pupal stage between the leaf litter of their hosts or in the few centimeters of soil or humus. In the United States, species of *Zale* may produce more than two

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generations per year and enter diapause in late June or early July (Wagner et al. 2011).

There have been 37 species described in the genus *Zale* (Lafontaine and Schmidt 2010), and all of these species feed on several types of pine trees (*Pinus* spp.) (Lafontaine and Schmidt 2010, Wagner et al. 2011). Interestingly, caterpillars of the genus *Zale* have now been observed on commercial blackberry groves (*Rubus* sp., cultivar 'Tupi') in Zirimicuaro, Municipality of Ziracuaretiro, Michoacan, Mexico, as of 2007, and they were identified in 2009 by the Spanish expert Dr. A. Vives (Sociedad Hispano-Luso-Americana de Lepidopterología [SHILAP], Madrid, Spain) as *Zale phaeograpta* (Hampson) (López 2009).

The blackberry crop is the second most important crop after avocados in the state of Michoacan, based on its production value and increases in employment. Currently, this state is the world's main producer and exporter of blackberry fruit because it is responsible for 97% of the total national production, with an area of  $\approx$ 9,000 ha (SIAP 2012). Since 2002, Z. phaeograpta larvae have been observed feeding on the newly emerged leaves of this crop, causing extensive defoliation if they are not controlled in a timely manner. Although the occurrence of this insect is sporadic, to control it, growers use synthetic organic insecticides and

Ann. Entomol. Soc. Am. 107(1): 170-177 (2014); DOI: http://dx.doi.org/10.1603/AN13093

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some biorational agents such as spinosad, azadirachtin, and *Bacillus thuringiensis* Berliner.

Many aspects of the life histories of the following species of the Erebinea subfamily have been studied: *Mocis latipes* (Guenée) (Euclidiini) (King and Saunders 1984), *Achaea janata* (L.) (Ophiusini) (Leong 2010), *Melipotis indomita* Walker (Melipotini) (Oda and Mau 1974), *Catocala nymphaea* (Esper) (Toimil 1987), *Catocala nymphagoga* (Esper) (Cánovas 2004), and *Cometaster pyrula* (Hopffer) (the last three species of Catocalini tribe). However, beyond the early morphological description of adults of Z. *phaeograpta* conducted by Hampson (1913), other characteristics of this species remain unknown.

With the goal of contributing to a better understanding of the Z. *phaeograpta* that are feeding on blackberries, a survey was taken in two different localities in the state of Michoacán, and different aspects of its morphology (including a diagnosis of life stages) and biology (duration of life cycle) and several life parameters (fecundity, preoviposition and oviposition periods, larval size, larval growth ratio, pupal weights and lengths, and sex ratio) were explained. In addition, the parasitoids from both larvae and pupae were recorded. This knowledge is essential to develop a better monitoring and management program for the species.

### **Materials and Methods**

Insects and Rearing. In 2009 (22 September and 13 October), 14 larvae from the third and fourth instars and four pupae of Z. phaeograpta were collected from a field crop of blackberries (Rubus sp. cultivar Tupi) in Zirimicuaro (19° 23' 80" N, 101° 57' 78" W and 1,254 m altitude) and Rancho El Barreno (19° 34' 565" N, 102° 27′ 786″ W and 1,351 m altitude) in the state of Michoacan, Mexico. In 2010 (21 August), another 33 larvae from the same instars were collected in Zirimicuaro. Larvae were collected from crop leaves and pupae from the leaf litter near the blackberry plants. Following collection, larvae were placed in ventilated plastic boxes (25 by 16 by 4 cm) with blackberry leaves as food, whereas pupae were individually placed in ventilated plastic containers (7.5 cm in diameter by 5 cm in height). Collected insects were transported to the Instituto de Investigaciones Agropecuarias y Forestales, Universidad Michoacana de San Nicolas de Hidalgo, Morelia, Michoacan. Once in the laboratory, larvae were individually placed into 3.5-cm<sup>2</sup> cylindrical wells of six-well Castor tissue culture plates and reared on a semisynthetic diet (Poitout and Bues 1974).

Z. phaeograpta adults that resulted from either the larvae or the pupae collected in the field between 22 September and 13 October 2009 were considered generation zero ( $G_0$ ) and placed in cylindrical plastic containers (14 cm in diameter by 28 cm in height), with paper sheets covering both ends, and were provided with a 15% (wt:vol) honey solution made with distilled water. The interior walls of the plastic containers were covered with tissue paper for oviposition. Eggs were collected daily and placed in ventilated

plastic boxes (25 by 16 by 4 cm) until larvae emerged, at which point they were reared on the semisynthetic diet mentioned earlier. The whole rearing process was completed in an environmental chamber at  $25 \pm 2$  °C with  $75 \pm 5\%$  relative humidity (RH) and a photoperiod of 16:8 (L:D) h.

Life History and Diagnosis. Z. phaeograpta development in the laboratory was determined by recording the duration of the different stages (eggs, larva, prepupa, pupa, and adult). To do this, a cohort of 276 eggs (<24-h-old) was randomly selected from adults of Go. Paper sheets containing eggs were cut into small circles (≈15 mm in diameter) and individually placed into the 3.5-cm<sup>2</sup> wells already described. After emergence, each larva was kept in the same well and fed a semisynthetic diet until it completed the pupal molt. Each well was examined at 12-h intervals to see if the larva had molted, and the shed head capsules, if present, were collected and saved in vials for growth ratio assessment, as described further in the text. From molting times, the duration and number of larval instars were determined. The different life stages of Z. phaeograpta were described considering the most distinctive morphological aspects of every one. The images of the different life stages were obtained with a photomicroscope Carl Zeiss III Tessovar, equipped with a PAXcam 3 camera, and they were edited using GIMP 2.8.4 photo editing software.

Pupal Weights and Lengths and the Sex Ratio. The weights and lengths of the 37 female and 26 male 3-d-old pupae were individually assessed 3 d after pupation. Pupae were sexed based on an examination of the seventh, eighth, and ninth sterno-abdominal segments (Sannino et al. 1897) using a stereoscopic microscope ( $40 \times$ ) (Zeiss Stemi DV4). The sex ratio was also calculated.

Fecundity. To determine fecundity, 18 pairs of adults (<24-h-old) from the  $G_1$  were used. Each pair was placed in a separate oviposition container (13 cm in diameter by 11 cm in height) lined with tissue paper and supplied with a 15% honey solution. The tissue paper was replaced every 2 d, and fecundity was determined by counting the total number of eggs laid by each female until death. Duration of preoviposition and oviposition periods and maximum oviposition were also evaluated.

Growth Ratio of Larvae. Dyar's rule states that after sclerotization, the size of the head capsule remains more or less constant during the same larval instar with a regular geometric progression from one molt to the next (Dyar and Rhinebeck 1890). To know if larval growth of Z. phaeograpta proceeds according to Dyar's rule, the size (in micrometers) of the head capsule was estimated by measuring the distance between both genae (Dyar and Rhinebeck 1890) and the distance between the head frontal setae (Podoler and Klein 1978) in all successive larval instars. The head capsules shed after molting were fixed in 70% ethanol for 5 d and then mounted on glass slides with transparent gel to ensure that their facial areas stayed parallel to the surface of the slide so that the planes of the surface of the width being measured were perpendicular to the microscope (Calvo and Molina 2008). The images of the head capsules were obtained with a photomicroscope and analyzed with UTHSCSA Image Tool version 3.0 (Wilcox et al. 2002). A minimum of 11 and a maximum of 35 head capsules of each of the seven instars of *Z. phaeograpta* were measured. The larval length (in cm) was also measured 6–8 h after molting on between 10 and 20 larvae from each instar.

The growth ratio for each molt was calculated as the quotient between the dimensions of the parameters measured in two consecutive instars (distance between genae and distance between frontal setae) according to Calvo and Molina (2008).

Parasitoids' Identification. The identification of the emerged parasitoids from both the larvae and the pupae of *Z. phaeograpta* that were collected in the field was made using the keys developed by Whitfield (1997), Gibson (1997), and Goulet and Huber (1993). All adult parasitoids were deposited in the parasitic Hymenoptera collection of the Instituto de Investigaciones Agropecuarias y Forestales–Universidad Michoacana de San Nicolas de Hidalgo.

Data Analysis. Weights and sizes of pupae by sex, adult longevity, and larval growth ratio for the widths between the genae and the distances between the head frontal setae of each instar of *Z. phaeograpta* were subjected to Student *t*-tests or Mann–Whitney U non-parametric tests using SPSS version 10.0 program (SPSS Inc., Chicago, IL).

#### **Results and Discussion**

Life History and Diagnosis. It is well known that the life parameters of insects can be influenced by environmental conditions and food quality (Silva et al. 1991). For example, an increase in temperature, within a favorable range, may accelerate the metabolism of the insects and consequently increase their rates of development. To our knowledge, there is no information about the life cycle of *Z. phaeograpta*, which under  $25 \pm 2^{\circ}$ C and 75% RH lived 87 d. This value is similar to what is reported for *M. latipes* (80 d) at  $27 \pm 2^{\circ}$ C and 80% RH (Reinert 1975).

Eggs. Wagner et al. (2011) reported that depending on the environmental conditions, eggs of the species of the genus Zale hatch in  $\approx$ 5–15 d. In this study, Z. phaeograpta eggs hatched after a duration of 4 d (Table 1), similar to that reported for *Me. indomita* (3 d; Oda and Mau 1974). Z. phaeograpta eggs were laid singly; they are spherical-shaped, flattened on the lower surface, and measure 0.8 mm in diameter (Fig. 1a), similar to the eggs of *Me. indomita* (0.8 mm) (Oda and Mau 1974) and Co. purula (0.7 mm) (Palmer and Senaratne 2007). In contrast, the diameters of M. latipes (González 1995) and Catocala promissa Denis & Schiffermüller (Catocalini) (Fry 2007) eggs were 0.5 and 1.25 mm, respectively. Freshly laid eggs are turquoise blue (Fig. 1a), but 48 h after being laid, brown spots were visible through the reticulated chorion. The color of the egg continued to darken until it was completely brown 72 h later (Fig. 1b). At this time, the larvae could be seen moving inside. The micropile,

	Table 1.	Length	(d ±	SE)	of	diffe	erent	dev	elo	pmei	nt sta	ages	of
Z.	phaeograp	ta reare	ed at	<b>25</b>	±	2°C	with	<b>75</b>	±	5%	RH	and	a
ph	otoperiod o	of 16:8 (	L:D)	h									

Development stages	(n)	Length in d		
Egg	276	$4.1 \pm 0.0$		
First instar	254	$6.9\pm0.0$		
Second instar	217	$3.5\pm0.0$		
Third instar	144	$4.0 \pm 0.0$		
Fourth instar	114	$5.1 \pm 0.1$		
Fifth instar	93	$5.4 \pm 0.1$		
Sixth instar	44	$5.1 \pm 0.1$		
Seventh instar	30	$5.9 \pm 0.3$		
Prepupa	53	$3.1 \pm 0.1$		
Pupa	41	$14.5 \pm 0.1$		
Adult	34	$29.1 \pm 1.4$		

*n*, number of specimens.

which is the structure that permits the spermatozoids to enter the egg, is rosette-shaped (Fig. 1c). Female *M. latipes*, *C. nymphaea*, and *C. nymphagoga* also deposit their eggs individually, but unlike *Z. phaeograpta* eggs, they are completely spherical and brownish-green (González 1995, Cánovas 2004).

Larva. Larvae of Z. phaeograpta underwent seven instars. The first instar had the longest duration (7 d), followed by the seventh (6 d); fourth, fifth, and sixth (5 d); third (4 d); and second (3 d) instars (Table 1). M. latipes larvae also underwent seven instars (Reinert 1975), but C. nymphaea and C. nymphagoga had only five instars (Cánovas 2004). In our study, only three individuals had an additional larval instar. An increase in instar number is well documented in the literature for several insect species, such as *M. latipes* (Reinert 1975), Malacosoma disstria Hübner (Esperk et al. 2007), and Streblote panda Hübner (Calvo and Molina 2008) (Lepidoptera: Tortricidae), among others. This largely depends on the nutritive value of the food on which they are fed, which is affected by environmental factors (Esperk et al. 2007, Calvo and Molina 2008).

The newly hatched first instar of Z. phaeograpta is creamy white with a yellowish-brown head capsule (Fig. 1d). The abdominal segments V, VI, and X have a pair of well-developed prolegs, which have an enlarged dark area that runs laterally. The prolegs of the abdominal segments III and IV are very minimal until at least the third larval instar (Fig. 1d, e, and f). For this reason, from abdominal segment V up to the thoracic legs, there is a curve in the body when the insect walks similar to what is observed in "looper" larvae of the Geometridae family (Toimil 1987). The first-instar larvae have setigerous tubercles the same color as the head, distributed in a pattern that is typical of Erebinae larvae (Wagner et al. 2011). The diameter of the prothoracic spiracle is smaller than the base of the closest setigerous tubercle (Fig. 1k); this characteristic persists until the third larval instar.

Second-instar larvae also have the yellowish-brown color of the cephalic capsule, but with darker reticulation (Fig. 1e). In this instar, two light-brown, diffused longitudinal lateral bands are present, except for in abdominal segments I and II. From this instar on-



Fig. 1. Zale phaeograpta. (a) Lateral view of a freshly laid egg; (b) dorsal view of an egg, 72 h later; (c) micropile in rosette shape; (d) lateral and dorsal views of the first-instar larva; (e) lateral and dorsal view of second-instar larva, arrows indicate: 1. cephalic capsule with reticulations. 2. lateral bands, 3. mid-dorsal band; (f) lateral and dorsal view of third-instar larva, arrows indicate: 1. prolegs of abdominal segments III and IV, 2. dorsal protuberances of abdominal segment VIII; (g) lateral and dorsal views of the fourth-instar larva, arrow indicates: 1. dorsal bands; (h) lateral and dorsal views of fifth-instar larva; (i) lateral and dorsal views of sixth-instar larva, arrow indicates: 1. mid-dorsal band; (j) lateral, dorsal, and ventral views of seventh-instar larva, arrows indicate: 1. prolegs of the abdominal segments III and IV, 2. dark irregular spots; (k) first-instar larva arrow indicates: 1. mid-dorsal band; (j) lateral, dorsal, and ventral views of seventh-instar larva, arrows indicate: 1. prolegs of the abdominal segments III and IV, 2. dark irregular spots; (k) first-instar larva seventh-instar larva, arrows indicates: 1. prolegs of the abdominal segments III and IV, 2. dark irregular spots; (k) first-instar larva seventh-instar larva, arrows indicates: 1. prolegs of the abdominal segments III and IV, 2. dark irregular spots; (k) first-instar larva seventh-instar larva, arrows indicates: 1. prolegs of the abdominal segments III and IV, 2. dark irregular spots; (k) first-instar larva showing prothoracic spiracles and setigerous tubercles.

wards, a whitish longitudinal dorsal band is observed along the entire body (Fig. 1e).

In the third-instar larvae, the prolegs of abdominal segments III and IV are more evident (Fig. 1f) than in the previous stages. At this stage, the loop formed during walking becomes less pronounced than before. A pair of dorsal protuberances are also observed in abdominal segment VIII (Fig. 1f), and they are more evident in the later larval stages (Fig. 1g, h, and i). The color of the lateral bands also becomes darker.

The fourth-instar larvae are cream-colored. The whitish longitudinal dorsal band, differentiated in the second instar, is more evident and also outlined by two

dark bands (Fig. 1g). Ventrally, a dark irregular spot that runs along the entire body is visible until the seventh instar (Fig. 1j).

The fifth-instar larvae are similar to those of the fourth instars, except in size (Table 2). However, sixth-instar larvae have entirely creamy brown bodies, and only the whitish mid-dorsal band can be distinguished (Fig. 1i). In the seventh larval instar, the prolegs of the abdominal segments IV to X increase in size, whereas those from segment III continue to be small (Fig. 1j). In this larval instar, the whitish longitudinal dorsal band is almost imperceptible. In the union between abdominal segments I and II, there is

Table 2.	Larval size (cm	± SE) per instar	when measured 6-	-8
h after Z. ph	aeograpta molt			

Larval instar	n	Length	Growth ratio
Ι	20	$0.3 \pm 0.0$	
II	20	$0.8\pm0.0$	2.3
III	20	$1.3 \pm 0.0$	1.6
IV	15	$1.7 \pm 0.0$	1.3
V	11	$2.4 \pm 0.0$	1.4
VI	10	$2.9 \pm 0.0$	1.2
VII	10	$4.1 \pm 0.1$	1.4
Avg			1.5

n, number of larvae measured.

an interruption of the dark dorsal bands, giving the appearance of an hourglass. In every larval instar, the prothoracic spiracle, as well as that of the abdominal segment VIII, is larger than the rest.

The sizes of the first- and seventh-instar larvae of *Z. phaeograpta*, 6-8 h after molting, ranged between 0.35 and 4.1 cm (Table 2). However, when the seventh-instar larvae are fully grown, they measure 6 cm in length (Fig. 1h), similar to *M. latipes* (5.5 cm) (González 1995) and *C. promissa* (5 cm) larvae (Fry 2007). The average growth ratio from one instar to the next was 1.5.

In Lepidoptera species, the recognition of larval instars is important because feeding habits can vary according to growth stadium. Larvae can occupy different regions in the plant, which is information that must be known to ensure pesticide efficacy (Flores-Pérez et al. 2005).

**Prepupa.** The duration of prepupal stage in days was  $3.1 \pm 0.1$  d (Table 1) and the average size in centimeters  $3.2 \pm 0.15$  cm (n = 50). Although determination of the prepupal stage can be difficult in some insects, this was not the case in *Z. phaeograpta*. At the prepupal stage, larvae stop feeding and dig a cavity in the semisynthetic diet, where they remain immobile and close this space with silk threads mixed with feces, forming a cocoon. In this stage, the intersegmental unions of both the thorax and the abdomen are more pronounced (Fig. 2a), making their bodies thicker and considerably shorter. The prepupal is spindle-shaped and light brown in color.

**Pupa.** The duration of the Z. phaeograpta pupal stage was 14.5 d (Table 1), similar to what has been reported for C. nymphagoga (18 d) (Cánovas 2004). Under field conditions  $(30^{\circ}C)$ , the *M. latipes* pupa lasted 7 d (Álvarez and Sánchez 1981), but it may last up to 37 d at 15-16°C (Mahadeo 1977). The Z. pha*eograpta* pupa is dark brown and covered with a waxy bloom (Fig. 2b), characteristics that it shares with M. latipes (King and Saunders 1984), C. nymphaea and C. nymphagoga (Cánovas 2004), and C. promissa (Fry 2007). The cremaster, which is the terminal part of the abdomen, of the Z. phaeograpta pupa is ornamented with eight hooks. The two central hooks are the most robust, and their distal ends face outwards (Fig. 2c). On the outer edge of these two hooks, there are three more hooks that are thinner and smaller, and their tips are pointed toward the base of the most robust hooks.

This cremaster structure is very similar to that of *C. nymphaea*, except in the number of small hooks (To-imil 1987).

Morphological differences between Z. phaeograpta male and female pupae can be seen on abdominal segments VII, VIII, IX, and X (Fig. 2c). On males, there is a small ridge on a circular area on abdominal segment IX that is signaled by an incision. On females, there is no tegumentary relief but rather a differentiated longitudinal incision that reaches the edge of abdominal segment VII and continues through segments VIII, IX, and X. These characteristics are similar to those reported for several lepidopteran species of the Noctuidae family (Sannino et al. 1987). The weights of the female and male Z. phaeograpta pupae were 0.5  $\pm$ 0.02 g and 0.5  $\pm$  0.03 g, respectively, which is slightly higher than what is reported for female (0.3 mg) and male (0.4 mg) pupae of Co. pyrula (Palmer and Senaratne 2007). Female and male pupae measured 2.21  $\pm$ 0.02 and 2.25  $\pm$  0.02 cm, respectively, which is not significantly different (U = 424; P = 0.41). These results are very similar to those reported for other species of Erebinae, such as M. latipes (King and Saunders 1984), C. nymphaea (Toimil 1987), Me. indomita (Oda and Mau 1974), and C. promissa (Fry 2007), which measured between 2 and 2.5 cm.

Adults. No significant differences (t = 0.21, df = 32, P = 0.83) were observed in the longevity of adult male and female *Z. phaeograpta*. Male (n = 16) and female (n = 18) adults lived for 28.8 ± 2.0 and 29.4 ± 2.1 d, respectively. The proportion of males to females was 1:1.1.

Z. phaeograpta adults are sexually dimorphic (Fig. 2d and e). Females are light brown in color, except for the last abdominal segments, which are dark brown. The costal margin of the forewings has a dark area as well as a band of the same color that begins at the posterior edge and ends in the apical margin (Fig. 2d). The apical margin of the forewings in males has two silvery bands as well as a transverse line, which is also silvery, in the first third of these wings (Fig. 2e). In the hind wings, the silver spot occupies most of the preapical margin (Fig. 2e). The frenulum, a structure present on the hind wings of lepidopterans, consists of a long, thick seta in males, whereas in females, this structure is made up of three setae, the outermost seta being thinner and smaller than the other two. These differences in the frenulum are typical of the genus Spodoptera (Sannino et al. 1987). Adults have filiform antennae with >80 segments, 1.8–2-cm-long. The wingspan oscillates between 5 and 5.2 cm, similar to that of C. nymphaea (4.8-5.1 cm) (Toimil 1987) and slightly higher than that of *M. latipes* (3.5-4.0 cm)(González 1995).

**Fecundity.** The mean number of eggs laid by each *Z. phaeograpta* female, when fed for her entire lifetime with 15% honey under our environmental conditions  $(25 \pm 2^{\circ}\text{C} \text{ and } 75 \pm 5\% \text{ RH})$ , was  $281 \pm 20$ , with preoviposition and oviposition periods of  $9.9 \pm 0.4$  and  $7.3 \pm 0.6$  d, respectively. The maximum oviposition took place  $3.8 \pm 0.5$  d after the start of oviposition, with 87.7  $\pm$  16.8 eggs/female. Based on the literature, the



**Fig. 2.** *Z. phaeograpta:* (a) Prepupal stage showing intersegmental joints; (b) dorsal, lateral, and ventral views of a pupa; (c) ventral view of the last abdominal segments of a pupa, arrows indicate: a male with a small ridge on a circular area on an abdominal segment IX, cremaster of female, a female showing a differentiated longitudinal incision; (d) female habitus; (e) male habitus. Parasitoids: (f) Microplitis sp.; (g) Ichneumonidae; (h) *Brachymeria comitator*; (i) *Brachymeria* sp.

recorded fecundity over the life span of this species is similar to what has been previously reported for the genus Zale (up to 200 eggs per female; Wagner et al. 2011) and for some species of the genus *Catocala* (*C*. nymphaea, Toimil 1987; C. nymphagoga, Cánovas 2004), although another species of this genus can lay up to 340 eggs/female (Co. pyrula, Palmer and Senaratne 2007). The available amounts of carbohydrate and protein and their origins are important in determining the fecundity values of insects, and their intake can be modified in relation to reproductive needs during adulthood (Viñuela and Arroyo 1983). Another species of the same subfamily, M. latipes, laid more eggs/female when fed with 5% honey ( $\approx 291$ ) compared with 5% sucrose, fructose, or glucose (between 40 and 211) under identical environmental conditions to ours. Moreover, the number of eggs/female in this species decreased when the concentration of honey was increased to 15% (70–170) (Silva et al. 1991).

**Growth Larval Ratio.** Width between the genae and the distance between the frontal setae of *Z. phaeograpta* head capsule ranged between 399 and 3,270 and 49 and 439  $\mu$ m at the beginning and end of the larval instars, respectively (Table 3). The growth ratio of larvae of this species was approximately constant from one instar to the next. The average growth ratios were also very similar for both measured variables (1.42 for the width between genae and 1.44 for the distance between frontal setae) and no significant differences were observed (t = -1.7, P = 0.143). These values are consistent with the growth constant of Dyar's rule, which states that after each molt, the larval growth increases geometrically, with an average constant ratio for each species of  $\approx 1.40$  (Dyar and

Table 3. The size of the head capsule ( $\mu m \pm EE$ ) per instar of *Z. phaeograpta* when measuring both the distance between the genae and the distance between the frontal setae

Larval instar	n	Distance between genae	Growth ratio	Distance between frontal setae	Growth ratio
I	11	$398.7 \pm 1.9$		$49.2 \pm 0.7$	
Π	21	$576.2 \pm 5.4$	1.44	$73.4 \pm 1.0$	1.48
III	34	$858.0\pm9.3$	1.48	$113.7 \pm 1.0$	1.55
IV	22	$1285.9 \pm 19.5$	1.49	$168.7 \pm 2.7$	1.48
V	23	$2046.6 \pm 19.3$	1.59	$268.1 \pm 3.0$	1.59
VI	31	$2363.2 \pm 17.5$	1.15	$310.9 \pm 3.3$	1.15
VII	16	$3270.0 \pm 18.3$	1.38	$439.2 \pm 5.6$	1.41
Avg			1.42		1.44

n, number of head capsules measured.

The growth ratio is calculated as the ratio between sizes measured at one instar and the next.

Rhinebeck 1890). Similar values have been reported in the literature for many other insect species of different orders: Conotrachelus psidii Marshall (Coleoptera: Curculionidae) (1.37 times) (Bailez et al. 2003), Heliothis zea (Lepidoptera: Noctuidae) (1.41 times) (Boddie) (Zúñiga 2005), and Comadia redtenbacheri Hamm (Lepidoptera: Cossidae) (1.42 times) (Hernández-Livera et al. 2005). However, not every insect species follows Dyar's rule; known exceptions are as follows: Ochrogaster lunifer Herrich-Schäffer (Lepidoptera: Thaumetopoeidae) (Floater 1996), Copitarsia incommoda (Walker) (Lepidoptera: Noctuidae) (Flores-Pérez et al. 2005), and Zabrotes subfasciatus (Boh.) (Coleoptera: Bruchidae) (Rodríguez-Quiroz et al. 2000), for which growth ratios are 1.30, 1.35, and 1.88 times, respectively. These differences may be because of interspecific characteristics, which are conditioned by geographical and environmental factors and the type and quality of food supplied (namely, the plant species) (Calvo and Molina 2008).

Parasitoids. Little information is available about larval and pupal parasitoids of the genus Zale. The braconids Protomicroplitis foacetosa (Weed) and Apanteles paleacritae Riley can attack larvae of an unknown Zale species (Raffa 1977). Barbosa and Caldas (2004) reported 13 and 15 species of parasitoids developing on Zale lunata Drury and Zale galbanata (Morrison), respectively. Of the parasitoids found, Euplectrus sp. (Eulophidae); Cotesia sp. and Diolcogaster facetosa (Weed); Glyptapanteles sp. and Aeiodes nolophanae Ashmead (Braconidae); Casinaria sp. (Ichneumonidae); Campylochaeta platipenae (Sabrosky); and Ceromya americana (Townsend) (Tachinidae) could parasitize both Zale species. In our study, however, neither of the previously cited parasitoids of the genus Zale were found to attack Z. phaeograpta. Larvae collected on 22 September 2009 in Zirimicuaro were not parasitized. However, in the field collection realized 1 yr later (on 21 August 2010) in this same place, larvae were parasitized by Micro*plitis* sp. (Braconidae) (33%, n = 33) (Fig. 2f) and an unidentified Ichneumonidae species (9%, n = 33) (Fig. 2g). From four pupae collected in Rancho El Barreno on 13 October 2009, emerged two parasitoids: Brachymeria comitator (Walker) (Fig. 2h) and an unknown species of the same genus (Chalcididae) (Fig. 2i). Although all the species of the genus *Microplitis* are parasitoids of Lepidoptera larvae, and all of *Brachymeria* attack a wide variety of pupae of this same insect order, this is the first report of *Z. phaeograpta* as hosts of both parasitoids. The parasitism of *Microplitis* sp. was notable in one of the field collections of larvae. However, because of the small size of the sample, new *Z. phaeograpta* larval and pupal collections are underway to determine the real importance of parasitism in populations of this insect and to identify the most important species.

In conclusion, this is the first report of several different aspects of the life cycle of *Z. phaeograpta:* life parameters, morphology, and parasitoids. This information will help to implement management programs for this potential pest of blackberries, especially between bud break and development of fruiting laterals. This crop is very valuable for the Mexican economy, and there is a need to prevent damages by pests to make the crop more economically profitable.

### Acknowledgments

This work was financially supported by the Coordinacion de la Investigacion Cientifica, Universidad Michoacana de San Nicolas de Hidalgo and by the Fundacion Produce Michoacan, through grants to S. P. and A. R., respectively. This research forms part of the undergraduate thesis of Y.-M. V. Authors are grateful to American Journal Experts for reviewing the English style, which has contributed to clarify the manuscript.

#### **References Cited**

- Álvarez, R.J.A., and G. G. Sánchez. 1981. Ciclo de vida y descripción del gusano agrimensor, *Mocis latipes*. Revista ICA (Colombia) 16: 57–63.
- Bailez, O. E., A. M. Viana-Bailez, J.O.G. De Lima, and D.D.O. Moreira. 2003. Life-history of the guava weevil, *Conotrachelus psidii* Marshall (Coleoptera: Curculionidae), under laboratory conditions. Neotrop. Entomol. 32: 203– 207.
- Barbosa, P., and A. Caldas. 2004. Patterns of parasitoid-host associations in differentially parasitized macrolepidopteran assemblages on Black Willow Salix nigra (Marsh) and Box Elder Acer negundo L. Basic Appl. Ecol. 5: 75–85.
- Calvo, D., and J. M. Molina. 2008. Head capsule width and instar determination for larvae of *Streblote panda* Hübner (Lepidoptera: Lasiocampidae). J. Econ. Entomol. 1001: 881–886.
- Cánovas, G. P. 2004. La encina y la tierra agrícola. (http:// encina.etotana.com/4-6-1.htm#4.6.1.4).
- Dyar, H. G., and N. Y. Rhinebeck. 1890. The number of moles of lepidopterous larvae. Psyche 5: 420-422.
- Esperk, T., T. Tammaru, and S. Nylin. 2007. Intraspecific variability in number of larval instars in insects. J. Econ. Entomol. 100: 627–645.
- Floater, G. J. 1996. The brooks-Dyar rule and morphometrics of the processionary caterpillar Ochrogaster zunifer Herrich-Schaffer (Lepidoptera: Thaumetopoeidae). Aust. J. Entomol. 35: 271–278.
- Flores-Pérez, L. R., N. Bautista-Martínez, J. Valdéz-Carrasco, O. Galván-Morales, and S. Quiñones-Luna. 2005. Com-

paración de dos técnicas de medición de cápsulas cefálicas para separar estadios larvales de *Copitarsia incommoda* (Walker) (Lepidoptera: Noctuidae). Acta Zoológica Mexicana (n.s.) 21: 109–113.

- Fry, R. 2007. Light crimson underwing (*Catocala promissa*). (http://www.ukleps.org/Noctuids/2454s.jpg).
- Gibson, G. 1997. Morphology and terminology, pp. 16–44. In G.A.P. Gibson, J. T. Huber, and J. B. Woolley (eds.), Annotated Keys to the Genera of Nearctic Chalcidoidea (Hymenoptera). National Research Council of Canada, Ottawa, Canada.
- **González, R. M. 1995.** Susceptibilidad de *Mocis latipes* (Guenée) (Lepidoptera: Noctuidae), al nematodo entomopatogeno *Heterorhabditis bacteriophora* Poinar (Rhabditida: Heterorhabditidae). M. S. thesis, Facultad de Ciencias Biológicas y Agropecuarias. Universidad de Colima, Colima, Mexico.
- Goulet, H., and J. T. Huber. 1993. Hymenoptera of the world: an identification guide to families. Centre for Land and Biological Resources Research. Ottawa, Canada.
- Hampson, G. F. 1913. Catalogue of the Lepidoptera Phalaenae in the British Museum, Vol. XIII. Taylor and Francis. London, United Kingdom.
- Hernández-Livera, R. A., C. Llanderal-Cázares, L. E. Castillo-Márquez, J. Valdéz-Carrasco, and R. Nieto-Hernández. 2005. Identificación de instares larvales de *Comadia redtenbacheri* (Hamm) (Lepidoptera: Cossidae). Agrociencia 39: 539–544.
- King, A.B.S., and J. L. Saunders. 1984. Las plagas invertebradas de cultivos anuales alimenticios en América Central: Una guía para su reconocimiento y control. Bib. Orton IICA/CATIE. Turrialba, Costa Rica.
- Lafontaine, J. D., and B. Ch. Schmidt. 2010. Annotated check list of the Noctuoidea (Insecta, Lepidoptera) of North America north of Mexico. ZooKeys 40: 1–239.
- Leong, T. M. 2010. Final instar caterpillar and metamorphosis of Achaea janata (Linnaeus, 1758) in Singapore (Lepidoptera: Erebidae: Erebinae). Nat. Singapore 3: 297–303.
- López, C. I. 2009. Identificación, dinámica poblacional y parasitoides de dos lepidópteros plaga de zarzamora, *Rubus fruticosus L.*, en tres regiones productoras del estado de Michoacán. B. S. thesis, Facultad de Biología. Universidad Michoacana de San Nicolás de Hidalgo.
- Mahadeo, C. R. 1977. Mocis latipes and Spodoptera frugiperda on sugarcane. Plant Protect. Bull. 25: 211–212.
- Oda, C. S., and R.F.L. Mau. 1974. Description and life cycle of the monkeypod-kiawe caterpillar, *Melipotis indomita* Walker (Lepidoptera: Noctuidae). Proc. Hawaiian. Entomol. Soc. 21: 435–441.
- Palmer, W. A., and K.A.D.W. Senaratne. 2007. The host range and biology of *Cometaster pyrula*; a biocontrol agent for *Acacia nilotica subsp. indica* in Australia. Bio-Control 52: 129–143.
- Podoler, H., and M. Klein. 1978. Distance between frontal setae: a new tool for determining caterpillar instars. J. Nat. Hist. 12: 341–347.

- Poitout, S., and R. Bues. 1974. Elevage de chenilles de veingt-huit espeses de lépidoptères Noctuidae. Annals of Annales de Zoologie Ecologie Animale 6: 341–411.
- Raffa, K. F. 1977. Some parasites of lepidoptera larvae recently collected in Delaware. Entomol. News 88: 81–84.
- Reinert, J. A. 1975. Life history of the striped grassworm, Mocis latipes. Ann. Entomol. Soc. Am. 68: 201–204.
- Rodríguez-Quiroz, M., J. Valdéz-Carrasco, J. Vera-Graziano, and A. Castillo-Morales. 2000. Identificación de ínstares larvales de Zabrotes subfasciatus (BOH.) (Coloptera: Bruchidae) mediante las dimensiones de sus cápsulas cefálicas. Agrociencia 34: 83–90.
- Sannino, L., A. Balbiani, and B. Espinosa. 1987. Osservacioni morfobiologiche su alcune specie del genere *Spodoptera* (Lepidoptera: Noctuidae) e rapporti di parasitismo con la coltura del tabacco in Italia. Informatore Fitopatologico 11: 29–40.
- (SIAP) Servicio de Información Agroalimentaria y Pesquera. 2012. Anuario estadístico de la producción agrícola. (http://www.siap.gob.mx).
- Silva da, R. A., C. C. Freire, and M. J. Claret. 1991. Efeitos de diferentes dietas na biología da fase adulta de *Mocis atipes* (Guenée, 1852) (Lepidoptera: Noctuidae). Pesquisa Agropecuária Brasileira 26: 45–50.
- Toimil, F. J. 1987. Algunos lepidópteros defoliadores de la encina (Q. Ilex L.) y alcornoque (Q. suber L.), en la provincia de Huelva. Boletín de Sanidad Vegetal Plagas 13: 331–346.
- Viñuela, E., and A. Arroyo. 1983. Effect of nutrition on the susceptibility of *Ceratitis capitata* Wied. (Dip.: Tephritidae) adults to malathion. Influence of adult food, physiological stage and age. Acta Oecologica Oecologia Applicata 4: 123–130.
- Wagner, L. D., D. F. Schweitzer, J. Bolling, J. B. Sullivan, and R. C. Reardon. 2011. Owlet caterpillars of Eastern North America. Princeton University Press, Oxford, United Kingdom.
- Whitfield, J. B. 1997. Subfamily microgastrinae, pp. 333–364. In R. A. Wharthon, P. M. Marsh, and M. J. Sharkey (eds.), Manual of the New World Genera of the Family Braconidae (Hymenoptera). Special Publication of the International Society of Hymenopterists No. 1. Washington, DC.
- Wilcox, D., B. Dove, D. David, and D. Greer. 2002. Image tool version 3.0. University of Texas Health Science Center, San Antonio, TX.
- Zúñiga, A.A.H. 2005. Determinación etárea en Heliothis zea (Boddie) a través de la medición de las setas frontales: una herramienta para la gestión de recursos agrícolas. B.S. thesis, Facultad de Recursos Naturales, Universidad Católica de Temuco, Chile.

Received 9 June 2013; accepted 16 November 2013.