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## Studies on morphology and biology of immature stages of the predator *Rhyzobius lophanthae* Blaisdell (Col.: Coccinellidae)

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### Abstract

Morphological characteristics of immature developmental stages of *Rhyzobius lophanthae* Blaisdell (Col.: Coccinellidae), predator of scale insects of the family Diaspididae (Homoptera), are described. Data about the morphology of eggs, the four larval instars, and the pupa is presented. Larval instars are described in more detail: size of head capsule, length of tibia, distribution of dorsal and lateral setae of abdomen segments for each larval instar is reported. Distribution of spiracles is also given. Study on the biology concerns the influence of prey on the duration of instar periods. The diaspids *Aspidiotus nerii* and *Aonidiella aurantii* were used as food. The mean duration of the developmental period from egg to adult was 27.1 days, and no mortality was observed (0%) when *A. nerii* was used as prey. The respective numbers when the prey was *A. aurantii* were 48.8 days and 84%.

### 1 Introduction

The Australian native coccidophagous predator, *Rhyzobius lophanthae* Blaisdell (Col.: Coccinellidae), has been reported as an important natural enemy of most armored scale species of the family Diaspididae (Hom.: Coccoidea) (YUS, 1973; ROSEN, 1990). Its significance as a natural enemy of scale insects derives from its ecological and biological characteristics (STATHAS, 2000a,b). Furthermore, inundative releases of *R. lophanthae* in nature are recommended in applications of IPM programs (KATSOYANNOS, 1996).

Despite its importance, *R. lophanthae* has not been the subject of much study, and only little information is reported in literature about its morphology. SMIRNOFF (1950), provided some data about egg morphology and specified the way that eggs are laid under the scale of the host. More detailed reference of morphological charac-

teristics of immature stages of *R. lophanthae* is made by RICCI (1983), who studied the distribution of setae on head and thorax and the range of number of ventral setae on the 4th instar larvae and pupae.

Additional data on the morphology of immature stages of *R. lophanthae*, which is not reported in previous papers, is presented in this study. This data concerns not only the size of head capsule and length of tibia of all legs, but also distribution of setae and microtrichia of abdomen segments of all larval instars. Distribution of setae of larvae is regarded as an essential taxonomic characteristic for the species identification. For this reason, similar studies about distribution of setae on the body of coccinellid larvae have been conducted for many coccinellid species (HODEK, 1973; Klausnitzer in HODEK, 1973; Iperti et al., 1977). Moreover, number and distribution of spiracles of the larva and pupa of *R. lophanthae* are also examined.

The kind of prey influences significantly the duration of the developmental period of the instars of Coccinellidae, as well as many other biological characteristics, such as prey consumption, adult longevity and fecundity (HODEK, 1973; HODEK and HONĚK, 1996). During the present study, presentation of some additional data about biology of *R. lophanthae* concerning the influence of prey on the duration of instar periods, was regarded as necessary. The diaspids *Aspidiotus nerii* Bouché and *Aonidiella aurantii* (Maskell) were used as food.

### 2 Materials and methods

Individuals of *R. lophanthae* used for the study of morphology originated from a laboratory population reared on the diaspidid *Aspidiotus nerii*. Measurements of dimensions of eggs, head capsule, tibia and number of setae and microtrichia were made under stereoscope microscope on 25 individuals of *R. lo-*

Table 1. Length of fore, mid and hind tibia (mm) of larval instars of *Rhyzobius lophanthae*.

Leg	Length of Tibia (Mean ± SD, n=25)			
	1 <sup>st</sup> instar	2 <sup>nd</sup> instar	3 <sup>rd</sup> instar	4 <sup>th</sup> instar
I	0.068 ± 0.04	0.102 ± 0.09	0.150 ± 0.010	0.241 ± 0.011
II	0.072 ± 0.04	0.110 ± 0.07	0.164 ± 0.008	0.265 ± 0.013
III	0.078 ± 0.04	0.120 ± 0.07	0.179 ± 0.007	0.294 ± 0.012

Table 2. Number and distribution of setae and microtrichia on the dorsal and lateral side of abdomen segments of larval instars of *Rhyzobius lophanthae* (n = 25).

2A: 1 <sup>st</sup> instar						
Abdomen segment	Dorsal side				Lateral side	
	Internal line		External line		setae	microtrichia
	setae	microtrichia	setae	microtrichia		
I	0	2-3	0	1-3	0	1-3
II	0	2-3	0	1-3	0	1-3
III	0	2-3	0	1-3	0	1-3
IV	0	2-3	0	1-3	0	1-3
V	0	2-3	0	1-3	0	1-3
VI	0	2-3	0	1-3	0	1-3
VII	0	2-3	0	2-3	0	1-3
VIII	0	2-3	0	2-3	0	1-3
IX	0	1	2	1-3	0	0

2B: 2 <sup>nd</sup> instar						
Abdomen segment	Dorsal side				Lateral side	
	Internal line		External line		setae	microtrichia
	setae	microtrichia	setae	microtrichia		
I	2	0-2	2	0-2	1-2	1-3
II	2	0-2	2	0-2	1-2	1-3
III	2	0-2	2	0-2	1-3	0-3
IV	2	0-2	2	1-3	0-2	1-3
V	2	0-2	2	0-2	1-2	0-3
VI	2	0-2	2	1-2	1-2	0-3
VII	2	0-2	2	0-3	0-2	0-3
VIII	2	0-2	2	1-3	1-2	0-3
IX	0	1	2-3	1-3	0	0

2C: 3 <sup>rd</sup> instar						
Abdomen segment	Dorsal side				Lateral side	
	Internal line		External line		setae	microtrichia
	setae	microtrichia	setae	microtrichia		
I	2	2-5	2	2-4	2-3	2-4
II	2	3-5	2	2-4	2-4	2-5
III	2	2-4	2	2-5	2-5	2-4
IV	2	2-5	2	2-5	2-4	2-5
V	2	2-5	2	2-5	2-4	2-4
VI	2	2-5	2	2-5	2-4	1-5
VII	2	2-4	2	2-5	2-4	2-5
VIII	2	2-3	2	2-5	1-4	1-4
IX	0	1	2-4	2-6	0	0

2D: 4 <sup>th</sup> instar						
Abdomen segment	Dorsal side				Lateral side	
	Internal line		External line		setae	Microtrichia
	setae	microtrichia	setae	microtrichia		
I	2	4-7	2	2-7	2-5	3-9
II	2	2-9	2	2-8	3-7	2-10
III	2	3-8	2	3-7	3-7	3-7
IV	2	3-7	2	2-7	2-6	2-7
V	2	3-8	2	4-7	2-7	2-7
VI	2	3-7	2	2-8	4-7	3-7
VII	2	2-8	2	2-9	3-6	2-6
VIII	2	1-5	2	3-9	4-7	2-5
IX	0	1	3-6	2-6	0	0

*phanthae* for each larval instar. For biological studies, potato tubes infested by *A. nerii* and orange fruits infested by *Aonidiella aurantii* (Maskell) were used. Twenty-five individuals of the predator were reared from egg to adult, at 25 °C, 65 % relative humidity and 18h light per day. Observations were made daily.

### 3 Results

#### 3.1 Morphology

##### 3.1.1 Eggs

Eggs were laid under scales of predated female or male nymphs of *A. nerii*, singly or in groups of 2–5 eggs (fig.1) or even more rarely outside of the scale. Shape is elliptic with a narrowing front edge. Their length is  $0.50 \pm 0.02$  mm (Mean  $\pm$  SD) and width  $0.25 \pm 0.01$  mm. The colour depends on the host on which the ovipositing females feed. Adult females reared on *A. nerii* laid yellow eggs, whereas eggs found in nature laid by females who had consumed individuals of the diaspidid *Parlatoria pergandei* Comstock, were rose (STATHAS, unpubl. data).

##### 3.1.2 Larvae

Larvae of *R. lophanthae* consist of a strongly sclerotized head, 3 less sclerotized thoracic segments (legs), and 9 slightly sclerotized abdomen segments. Body colour is green-gray, and it is darker in the beginning of the development. Average width of head capsule of 1<sup>st</sup>, 2<sup>nd</sup>, 3<sup>d</sup> and 4<sup>th</sup> instar was  $0.22 \pm 0.01$ ,  $0.29 \pm 0.01$ ,  $0.42 \pm 0.01$  and  $0.55 \pm 0.01$  mm (Mean  $\pm$  SD), respectively. Length of tibia of all legs of each instar is presented in table 1. As shown in table 1, tibia I was the shortest on all instars, followed by tibia II and tibia III. On the dorsal side, there are two internal (fig. 2A) and two external (fig. 2B) parallel, longitudinal lines of setae and microtrichia. Two dorsolateral lines of setae are also present (fig. 2C). Numbers and distribution of dorsal setae and microtrichia of the abovementioned lines are presented in tables 2A-D. As shown in these tables, on 1<sup>st</sup> instar there are only 2 dorsal setae on the 9<sup>th</sup> abdomen segment. On 2<sup>nd</sup>, 3<sup>d</sup> and 4<sup>th</sup> instar, the internal and external dorsal line of setae is consisted of 2 setae on each abdomen segment, except the last one. Higher numbers of setae were observed on the lateral side and followed an increasing order from 2<sup>nd</sup> to 4<sup>th</sup> instar (tables 2B-D). Length of all setae exceeded 0.7 mm, whereas respective size for microtrichia did not reach 0.01 mm.

Larvae of all instars and pupa of *R. lophanthae* have the same number and distribution of spiracles. There are two parallel,

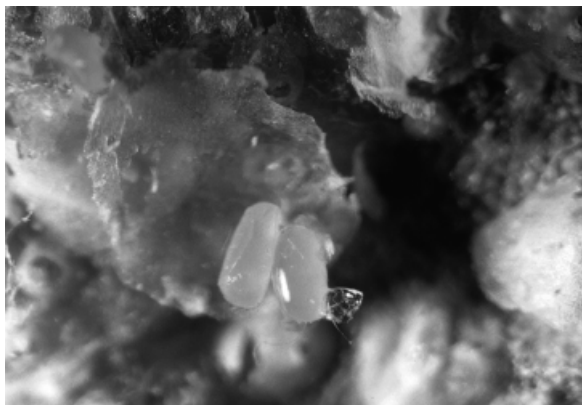


Fig. 1. Eggs of *Rhyzobius lophanthae* under scale cover of *Aspidiotus nerii*.

longitudinal lines of spiracles (9 spiracles each) along each lateral side of the body of larva and pupa. The 1<sup>st</sup> pair is situated on mesothorax and the remaining 8 pairs are located on the first 8 abdomen segments (one pair on each segment).

### 3.1.3 Pupae

During the last 2–3 days of 4<sup>th</sup> instar, the larva is tied up by sticking the last abdomen segment on a fixed point, and pupation follows. The shape of the pupa is broad, oval, with a narrow end (fig. 3). Its length and width vary between 2.9–3.6 mm and 1.7–1.9 mm, respectively. The colour of the pupa depends on the body colour of the scale which the individual had fed on as a

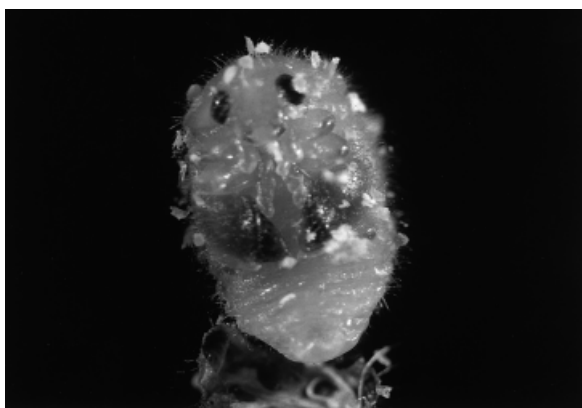


Fig. 3. Ventral view of pupa of *Rhyzobius lophanthae*.

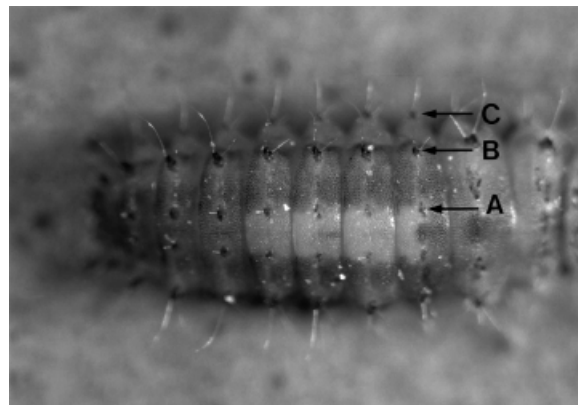


Fig. 2. Setae and microtrichia on abdomen of 4<sup>th</sup> instar larva of *Rhyzobius lophanthae* (Dorsal side: A: internal line, B: external line. Lateral side: C).

larva. Pupae originating from larvae previously fed on *Aspidiotus nerii* were yellow in colour, but when the prey was *Parlatoria pergandei*, pupae were rose (STATHAS, unpublished data). The puparium is entirely covered with setae. The thorax, a part of elytra and abdomen are distinguished dorsally, whereas head, legs and a larger part of elytra are visible from the ventral side.

### 3.2 Biology

As shown in table 1, development of *R. lophanthae* on *A. nerii* was shorter (Mean: 27.1 days) than on *A. aurantii* (Mean: 48.8 days). All eggs developed to adults on *A. nerii*, whereas significant mortality occurred on *A. aurantii* (table 3), and only four eggs reached the adult stage (total mortality 84 %).

## 4 Discussion

Size of eggs of *R. lophanthae* reared on *A. nerii*, as measured in the present study, showed little difference from respective numbers reported in other studies. These differences may be attributed to the different feeding conditions of adults. SMIRNOFF (1950), reported that eggs of *R. lophanthae* laid by adults which had fed on several diaspidid species were 0.4 mm long. Futhermore, RICCI (1983) indicates that individuals, which had also fed on various diaspidid species, laid eggs of 0.5 mm length and 0.3 mm width. The eggs are laid under scales (SMIRNOFF, 1950; STATHAS, 2000a), probably for protection from predators and parasites (HODEK and HONĚK, 1996). Average width of head capsule, as well as average length of tibia did not differ significantly between larvae of dif-

Table 3. Duration of development and mortality of immature stages of *Rhyzobius lophanthae* on two diaspidid scales.

	Egg	1 <sup>st</sup> instar	2 <sup>nd</sup> instar	3 <sup>rd</sup> instar	4 <sup>th</sup> instar	pupa
<i>Aspidiotus nerii</i>						
days (Mean ± SD)	7.4 ± 0.51	3 ± 0.45	2.2 ± 0.37	2.7 ± 0.61	6.2 ± 0.5	5.6 ± 0.51
N	25	25	25	25	25	25
mortality (%)	0	0	0	0	0	0
<i>Aonidiella aurantii</i>						
days (Mean ± SD)	12.4 ± 1.54	5.5 ± 1.22	4 ± 0.94	5 ± 0.79	11.3 ± 1	10 ± 1.29
N	17	14	10	8	7	4
mortality (%)	32	17	28.6	20	12.5	42.8

ferent instars (table 1). This is due to the sclerotization of head and legs, which make their size changeless, despite the expanding of the rest body during larval development (CHAPMAN, 1969). The increase in number of setae of tergum with larval development, which was observed in the present study for *R. lophanthae*, has also been recorded for the coccinellid *Exochomus quadripustulatus* Linnaeus (KATSOYANNOS, 1976). Number and distribution of spiracles of *R. lophanthae* indicate that the respiratory system of this coccinellid is of peripneustic type (CHAPMAN, 1969).

As presented in table 3, *A. aurantii* does not seem to be suitable prey for *R. lophanthae*. This explains the fact that *R. lophanthae* is not regarded as an important natural enemy of *A. aurantii* despite its frequent occurrence on this diaspidid (BATTAGLIA and VIGGIANI, 1982; SMITH, 1981). HONDA and LUCK (1995) reported that this phenomenon can be attributed to the hardened scale cover and sclerotized scale body of *A. aurantii*.

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