STUDY ON MORPHOLOGICAL AND BIOLOGICAL CHARACTERISTICS OF *OENOPIA KIRBYI* (COLEOPTERA: COCCINELLIDAE) IN DA LAT CITY, LAM DONG, VIETNAM

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Abstract

Ladybirds are natural enemies of many agricultural crop pests, such as aphids, scale insects, mealybugs, whiteflies, and thrips. This paper focuses on the morphological and biological characteristics of Oenopia kirbyi (Coleoptera: Coccinellidae) in Da Lat city (Lam Dong province). Field-collected adults of Oenopia kirbyi were reared in plastic boxes under laboratory conditions and fed on aphids. The egg phase lasted 4.45 ± 0.71 days; the four larval stages lasted 2.63 ± 0.52 days, 1.91 ± 0.73 days, 1.70 ± 0.46 days, and 5.78 ± 0.95 days, respectively; and pupae lasted 7.02 ± 0.91 days. The total duration of the immature stages was 23.49 days. The adult longevity of males and females was 67.13 ± 18.77 days and 82.83 ± 20.25 days, respectively. The fecundity was 414.00 ± 131.33 eggs on average. The rate of hatched eggs, emerged adults, and the female/male ratio were 80.85%, 96.53%, and 1.78, respectively. These results provide basic data for rearing Oenopia kirbyi for use as a biological control of crop pests.

Keywords: Biological characteristics; Morphological characteristics; *Oenopia kirbyi*; Predatory ladybird.

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1. INTRODUCTION

Da Lat city (Lam Dong province) is a famous vegetable and flower-growing region of Vietnam. With a cool temperate climate, Da Lat city is very suitable for the production of vegetables and flowers all year round. This fact has allowed pests such as aphids, mealybugs, scale insects, whiteflies, thrips, and others to thrive.

Currently, the local people mainly use chemical pesticides to control crop pests. This has affected the health of humans and domestic animals and caused environmental pollution. It has also caused resistance in pests and decreased the population of their natural enemies.

Biological control is increasingly used in the integrated pest management programs of many countries around the world and has obtained significant results compared with chemical pesticides.

Ladybird beetles (Coleoptera: Coccinellidae) comprise 6,000 described species worldwide, of which 90% are considered natural predators. This is one of the natural enemy groups that play an important role in agricultural production. Their food includes aphids, mealybugs, scale insects, whiteflies, thrips, insect eggs, small larvae, and phytophagous mites (Ajar & Akhtar, 2017).

The number of ladybird beetle species in the fauna of Vietnam has been discovered to be over 220 species, belonging to 65 genera, 15 tribes, and 6 subfamilies. The number of known beneficial ladybird species in the Vietnam coccinellid fauna has reached 165 species, belonging to 5 subfamilies and 60 genera (Hoàng, 1982). Some studies on the biological characteristics of predatory ladybird beetles have been published for *Chirocorus politus* (Nguyễn & Trần, 2010), *Coccinella tranversalis* (Mai et al., 2005), *Stethorus* sp., and *Harmonia sedecimnotata* (Nguyễn et al., 2005).

During surveying and collecting of predatory ladybird beetle species in Da Lat city, we recorded that *Oenopia kirbyi* commonly appeared on aphid-infected plants and weeds, but until now there have been almost no detailed studies on this species. With the hope of contributing to research on the use of predatory ladybird beetles in pest control, we studied the morphological and biological characteristics of *Oenopia kirbyi* in Da Lat city.

2. MATERIALS AND METHODS

Field-collected adults of *Oenopia kirbyi* were reared in plastic boxes and fed on aphids grown on black beans in the laboratory. In each plastic box, we put 3-4 pairs of adults together. The planting of beans and aphid culture was carried out regularly during the experiment to ensure that a sufficient supply of food was provided for the ladybirds. When the ladybirds reproduced, the eggs were collected daily to study morphological and biological characteristics.

Freshly laid eggs of *Oenopia kirbyi* were collected on the same day and put in a separate box. When the eggs hatched, individuals from the first instar were kept in 4 x 6 cm plastic cups covered with a perforated plastic cap and fed on aphids. New food was provided daily. Photographs were taken, and the developmental stages and size of the ladybirds were recorded.

Photographs of the development stages of *Oenopia kirbyi* were taken under a stereoscopic microscope or magnifying glass. At each stage, the length and breadth of 20 individuals were measured using a stereoscopic microscope fitted with a measuring tool (for egg and larval stages) or using a caliper (for pupal and adult stages). The colors and morphological characteristics of the stages were observed and described.

When the adults emerged from pupae, males and females were differentiated and paired. Thirty adult pairs of *O. kirbyi* were reared individually in 4 x 6 cm plastic cups covered with a perforated plastic cap and fed on aphids. Eggs were collected daily, and the number of eggs laid by each female was recorded.

Experiments were conducted at a temperature of 21 ± 2 °C and relative humidity of 80 ± 5%.

The data were statistically processed using Microsoft Excel software. Means (X) were calculated using the AVERAGE function, and standard deviations (SD) were calculated using the STDEV function.

3. RESULTS

3.1. Morphological characteristics

The ladybirds *Oenopia kirbyi* are holometabolous insects, undergoing a complete metamorphosis with four discrete life stages: egg, larva, pupa, and adult beetle.

3.1.1. Egg stage

Eggs are elongate ovals with an average size of 1.04 ± 0.02 mm x 0.52 ± 0.01 mm (n = 20) (n is the number of observed individuals), laid in clusters or scattered. Freshly laid eggs are pale yellow (Figure 1a). When they are about to hatch, they gradually turn dark gray (Figure 1b).

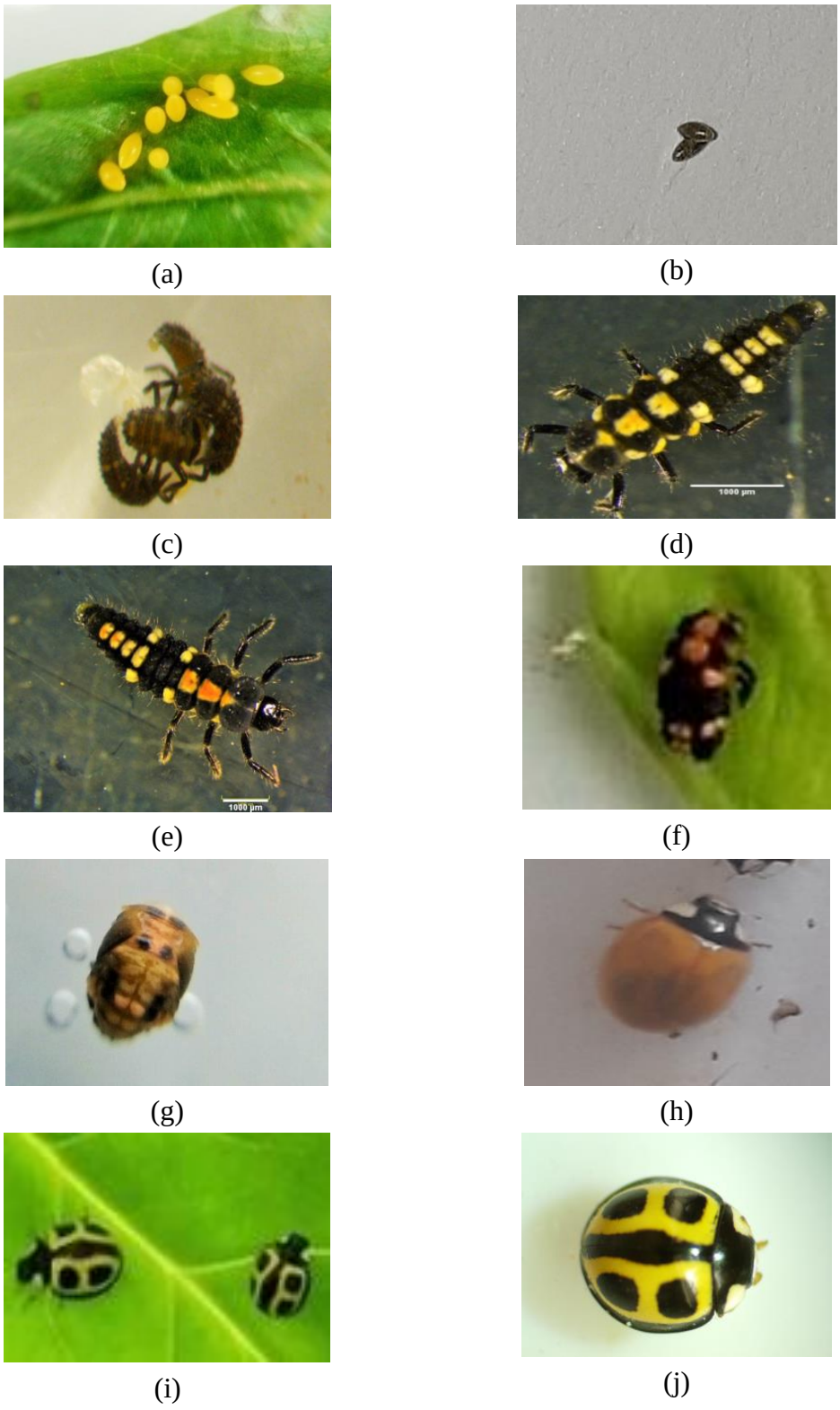

Figure 1. Development stages of *Oenopia kirbyi*

Note: (a) Egg cluster; (b) Egg before hatching; (c) First larval instar; (d) Third larval instar; (e) Fourth larval instar; (f) Pre-pupa; (g) Pupa; (h) Teneral adult; (i) Females; (j) Male.

3.1.2. Larval stage

Larvae of *O. kirbyi* are elongate, tapering posteriorly. The body is divided into three parts: head, thorax, and abdomen. There are four instars in larval life with three molts.

Larvae newly hatched from eggs are called first instar larvae. They are dark gray, covered with many small hairs, and have no distinctive color or pattern (Figure 1c). The first instar and second instar larvae are quite similar in shape. They only differ in size, with average sizes of 2.16 ± 0.04 mm x 0.74 ± 0.01 mm and 2.77 ± 0.03 mm x 0.78 ± 0.02 mm for first and second instar larvae, respectively (n = 20).

The third instar larvae have ivory-white and orange-yellow blotches on the thorax and abdomen. The blotches on the abdomen are arranged in a T-shape (Figure 1d). Third instars have an average size of 3.51 ± 0.04 mm x 1.11 ± 0.03 mm (n = 20).

The fourth instar larvae are not much changed in morphology compared to the third instar larvae, but they have grown significantly in size. Their average size is 5.16 ± 0.04 mm x 1.46 ± 0.03 mm (n = 20). The older larvae have orange-yellow blotches that turn darker and darker (Figure 1e).

The fully grown fourth instar larvae turn into the pre-pupal stage. At this stage, they cease to feed and become inactive. They attach themselves to the substrate in the box using their anal pad and their bodies shrink (Figure 1f).

3.1.3. Pupal stage

Pupae are initially orange-yellow in color with patterns similar to those of fourth instar larvae, then they darken (Figure 1g). The average pupal size is 3.50 ± 0.17 mm x 2.33 ± 0.18 mm (n = 20).

3.1.4. Adult stage

Adult beetles are oval, convex, and glabrous. Both male and female adults have black heads. The male beetles are recognised with a white forhead (Figure 1j). The elytra are pale yellow with a black margin and two black spots in each elytron. The lateral edge is uniformly black. The black strip along the meeting line of both elytra is irregular, bulging in the center (Figure 1i and 1j). The mean sizes of male and female adults are 3.37 ± 0.13 mm x 2.65 ± 0.16 mm and 3.95 ± 0.14 mm x 2.94 ± 0.17 mm (n = 20), respectively.

When newly emerged from pupae, the elytra of teneral adults are quite soft, yellow in color, and have no pattern (Figure 1h). It takes about half a day to develop the characteristic color pattern of the species.

3.2. Biological characteristics

3.2.1. Duration of immature stages

The immature development period consists of three stages: egg, larva, and pupa.

The egg incubation period was 3-6 days, with an average of 4.45 ± 0.71 days (Table 1). The group with the 4-day duration accounted for the highest percentage (58.19%), and the group with the duration of 3 days had the lowest percentage (3.14%).

The newly hatched larvae did not move immediately but usually remained stationary for several hours to half a day. Their first meal was their eggshells and the unhatched eggs. Then they began to move freely in search of food.

The first instar larvae lasted from 2-4 days, with a mean duration of 2.63 ± 0.52 days (Table 1). The group with a 3-day duration accounted for the most individuals (59.09%). The group with a duration of 4 days had the fewest individuals (1.71%).

The duration of the second and third instars was quite short, only 1-3 days, with an average of 1.91 ± 0.73 days for the second instars and 1.70 ± 0.46 days for the third instars (Table 1). The group with the 2-day duration accounted for the highest percentage of both instars, with 46.39% for the second instars and 69.54% for the third instars, respectively.

Table 1. Duration of immature stages of *Oenopia kirbyi*

Developmental stage	Number of observed individuals (*n*)	Duration (days) (X ± SD)
Egg	287	4.45 ± 0.71
First instar	176	2.63 ± 0.52
Second instar	166	1.91 ± 0.73
Third instar	151	1.70 ± 0.46
Fourth instar	144	5.78 ± 0.95
Pupa	139	7.02 ± 0.91
Immature development period		23.49

The fourth instar larvae stage lasted from 4-7 days, with an average duration of 5.78 ± 0.95 days (Table 1). The group with a 6-day duration accounted for the highest percentage (39.58%), and the group with 4 days of duration accounted for the lowest (11.11%).

Thus, the total larval period (first to fourth instar) was 8-16 days, with an average of 12.02 days (Figure 2).

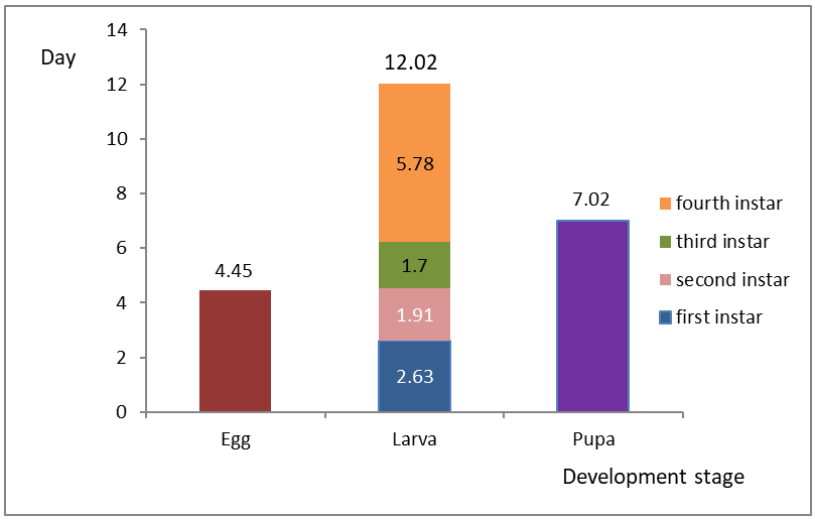

Figure 2. Duration of the immature stages of *Oenopia kirbyi*

The duration of the pupae lasted from 6-11 days, averaging 7.02 ± 0.91 days (Table 1). The group with the 7-day duration accounted for the highest percentage of individuals (55.40%).

In general, the duration of the egg, larval, and pupal stages of *Oenopia kirbyi* was slightly different from that of other predatory ladybirds. In *Coccinella tranversalis*, the average egg period was 3.86 days, the larval stage varied from 14 to 17 days, and the pupal period was 5.05 days (Mai et al., 2005). The average incubation period of *Chilocorus politus* was recorded by Nguyễn and Trần (2010) to be 3.7 days. The larval stages (from first to fourth) were 1.9, 1.8, 2.2, and 3.9 days, respectively. The pupal period was 7.1 days (Nguyễn & Trần, 2010).

3.2.2. Duration of adult stage

Adult longevity of *Oenopia kirbyi* was significantly different among observed individuals. In general, the longevity of males was shorter than that of females. The average longevity of males was 67.13 ± 18.77 days; the shortest was 28 days and the longest was 103 days. The longevity of females varied from 45 to 111 days, with an average of 82.83 ± 20.25 days (Table 2).

Table 2. Duration of adult stage of *Oenopia kirbyi*

Criteria	Mean (X ± SD)
Male longevity (days)	67.13 ± 18.77
Female longevity (days)	82.83 ± 20.25
Pre-oviposition period (days)	12.50 ± 2.26
Oviposition period (days)	65.53 ± 21.88

Note: n = 30 (n: number of observed individuals).

The average pre-oviposition period and oviposition period of females was 12.50 ± 2.26 days and 65.53 ± 21.88 days, respectively (Table 2).

Our results are quite similar to those of Nguyễn and Trần (2010). In *Chilocorus politus*, there is a big difference in longevity between males and females. The average longevities of males and females were 45 days and 80.6 days, respectively (Nguyễn & Trần, 2010).

3.2.3. Fertility

Usually, ladybird eggs were laid in clusters, sometimes scattered. In the laboratory, eggs were laid mainly on covers, on bean leaves, or in the boxes. There was a large variation in the number of eggs laid by observed individuals. The results in Table 3 show that under laboratory conditions, an *Oenopia kirbyi* female can lay 128 to 630 eggs, with an average of 414.00 ± 131.33 eggs. According to our observations, the number of eggs laid per female in a day was up to 22 eggs.

Compared to other species, the number of eggs laid by *Oenopia kirbyi* is quite high. The number of eggs laid per female varies among species: in *Chilocorus politus* the average is 183.5 eggs (Nguyễn & Trần, 2010), in *Coccinella tranversalis* it is 177 eggs (Mai et al., 2005), in *Stethorus sp*. it is 28.6 eggs, and in *Harmonia sedecimnotata* it is 414.7 eggs (Nguyễn et al., 2005).

Under laboratory conditions (temperature 21 ± 2 °C, relative humidity 80 ± 5%), the percentage of hatched eggs was 80.85%. The percentage of adults to emerge from pupae was 96.53%, of which males accounted for 35.97% and females 64.03%. The female/male ratio was 1.78. (Table 3). This result indicates the potential for rearing *Oenopia kirbyi* to control aphids and other crop pests in Da Lat city (Lam Dong province).

Table 3. Fertility of *Oenopia kirbyi*

Criteria	Mean (X ± SD)
Fecundity (eggs/female)	414.00 ± 131.33 (*n* = 30)
Rate of hatched eggs (%)	80.85 (*n* = 355)
Rate of emerged adults (%)	96.53 (*n* = 144)
Female/male ratio	1.78 (*n* = 139)

Note: *n*: number of observed individuals.

4. CONCLUSIONS

The average incubation period of *Oenopia kirbyi* was 4.45 ± 0.71 days. Mean duration of larval stages (from first to fourth) were 2.63 ± 0.52 days, 1.91 ± 0.73 days, 1.70 ± 0.46 days, and 5.78 ± 0.95 days, respectively. The pupal period was 7.02 ± 0.91 days. In general, the total period of the immature stages was 23.49 days.

The average longevity of adult males was 67.13 ± 18.77 days, and that of adult females was 82.83 ± 20.25 days.

The average number of eggs laid per female was 414.00 ± 131.33 eggs.

The rate of hatched eggs, rate of emerged adults, and female/male ratio were 80.85%, 96.53%, and 1.78%, respectively.

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