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# Effect of photoperiod on biological attributes of *Harmonia dimidiata* (Fab) (Coleoptera: Coccinellidae) fed on *Schizaphus graminum* (Rond.) (Homoptera: Aphididae) aphid

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## Abstract

Ladybird beetle *Harmonia dimidiata* Fab. (Coleoptera: Coccinellidae) is voracious predator of aphid pests. The present study was conducted to check the effect of long photoperiod (16: 8) hours light: dark and short photoperiod (8: 16) hours light: dark on the biological parameters of *H. dimidiata* at 24 ±1 $^{0}$C with 60±5 % relative humidity fed on *S. graminum* aphid in growth chamber. The results showed that egg incubation period was 4.22±0.15 and 4.21±0.14 days under both long and short phtoperiod. The duration of first and second instar larvae was 2.43±0.09, 2.42±0.09 and 2.24±0.10, 2.28±0.11 days which was non significantly different from each other under both long and short photoperiods. The duration of third and fourth instar larvae were 4.41± 0.16, 3.65± 0.17 and 8.0±0.10, 7.21± 0.16 days respectively, which was significantly different from each other. The larvae duration was 17.08± 0.12 and 15.56± 0.34 days respectively, which was significantly different from each other. The results showed that the effect of photoperiod was not extended to all stages. The preoviposition period of adult female was 9.0±0.45 and 11.65±0.26 days respectively which was significantly different from each other while the oviposition and post oviposition period was non significantly different. The female fecundity was 668.7±32.46 and 612.6±36.08 eggs per female which was non significantly different from each other. The results showed that *H. dimidiata* completed their life cycle successfully and no reproductive diapauses were observed under both photoperiods. On the basis of the present results, *H. dimidiata* should be reared successfully under long photoperiod (16: 8) hours (light: dark), owing to shorter pre oviposition period and high fecundity rate of female lady bird beetles

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## Introduction

Among the environmental factors, photoperiod is the most regular one, which usually provide the most consistent long lasting indication for future (Tauber *et al.*, 1986). It is known that day length could act as cue factor in inducing adult (reproductive) diapauses in aphidophagous coccinellids. However, the rate of coccinellid larval development is also dependent on photoperiod and diet (Reznik and Vaghina, 2011). Some insects show greater developmental rate under long-day conditions as compared to short-day (Danks, 2003). The short photoperiod not only slows down reproductive maturation but also speed up pre imaginal development (Reznik and Vaghina, 2011). *H. dimidiata* commonly known as fifteen spotted ladybird beetle is native to China and also found in South-East Asia, Vietnam, India, Nepal, Pakistan (Kuznetsov and pang, 2002). The biology and predatory potential of this species is least studied as compared to other coccinellid beetles worldwide. In Pakistan, it is found in subtropical hilly Northeren areas, Swat, Azad Kashmir and Islamabad. The beetles have the capability to live at low temperature. Both adults and larvae are voracious predator of aphids and fed on various aphid species like *M. persicae*, *S. graminum*, *A. craccivora*, *A. gossypii*, *A. glycines*, *M. rosae*, *A. fabae* and *A. pisum* in the field (Kuznetsov, 1997).

*Schizaphis graminum* (R.) generally recognized as green bug is a notorious insect pest of minute seed with a broad host range of about sixty plant species together with wheat, barley, sorghum and corn (Bowling *et al.*, 1998). Comprehensive research has been conducted in the past on the consequences of photoperiod on the life cycle of coccinellid beetles. i.e Rznike and Vaghina (2011), Tauber *et al.* (1986), Lopatina *et al.* (2007), Nijhout *et al.* (2010), Chen *et al.* (2014), Hodek (2012), Kucherove *et al.* (2011) and Semyanov (2009). However, apart from diapause, little research has been done to explore the influence of photoperiod on fecundity and oviposition of coccinellid beetles.

Therefore, the present study was conducted to check the effect of photoperiod, long day 16 hours light and short day 8 hours light on development, fecundity and induction of diapause at any of the development stages in order to find out the most suitable photoperiodic regimes for quality mass rearing of the predator under controlled conditions.

## Materials and methods

The present research work were conducted to observe the effect of long photoperiod (16: 8) hours light : dark and shorte photoperiod (8: 16) hours light: dark on biological attributes of *H. dimidiata* at 24 ±1 $^{0}$C, with 60± 5% relative humidity, fed on *S. graminum* aphid in growth chamber at Insect pest management program, National Agriculture Research Centre Islamabad Pakistan.

*Effect of long (16: 8) and short (8: 16) hours (light: dark) photoperiods on the developmental time of immature stages of H. dimidiata*

A total of 100 eggs in different bataches of the same age laid by *H. dimidiata* female were collecte from stock culture, maintained on *S. graminum* aphid in laboratory. Eggs were kept in petri dishes (4 × 2cm) for hatching in growth chamber under both photoperiodic regimes. Upon hatching a total of 20 first instar larvae were separated and kept individually in plastic vials of size (6 ×3 cm) under both photoperiodic regimes.The vials were covered with muslin cloth tightened with rubber band. Sufficient amount of aphids were provided on infested leaves of wheat plants in each vials daily. After each 24 hours the old infested leaves were replaced with fresh diet. The insects were observed for molting daily. The insect passes through four larval instars. The developmental period for each larval instars and total larval period were recorded. If the insect was found dead in vials than it was replaced with another insect of the same age from stock culture. The procedure was followed till all the larvae changed to pre pupal and subsequently to pupal stages. The data was recorded on incubation period (days), developmental period of different larval instars and total larval durations, pre-pupal and pupal durations, total developmental time taken from egg to adult emergence, and No. of Male/female beetles emerged.

*Effect of photoperiod on the biological attributes of adult female ladybird beetle*

Upon adult emergence from pupae at each photoperiod, 10 pair was transferred to rearing jars (4″×12″) under long and short photoperiodic regemes. The rearing jars were lined with filter paper to facilitate egg lying. *S. graminum* aphid mixed stages (i-iv) nymphal instars were provided inside the rearing jars in excess daily on wheat leaves. After 24 hours the old infested leaves were replaced with new diet in each jar. The insects were observed daily twice for egg laying. The first egg observed in each pair was the indication for the ending of pre oviposition period. The eggs laid on wheat leaves or on filter paper inside the jars in masses were collected immediately. The total eggs per female and mean number of eggs per female/day were counted under binocular microscope. The observations were carried out till all the male and female beetles were died in each jars under both photoperiod. The longevity of both sexes was recorded. During the female longevity experiment, the first and last egg laying was also recorded in each pairs to separate pre-oviposition, oviposition and post-oviposition periods. Data was recorded on pre-oviposition, oviposition and post oviposition period of female beetle, developmental period of male and female beetles, mean number of eggs per female beetles, and mean number of eggs per female per day.

The data was analyzed by analysis of variance "One way ANOVA using Statistix 8.1 computer package. The means data were compared using LSD test at ($P$< 0.05).

## Results and discussion

*Effect of long (16:8) hours and short photoperiod (8:16) hours on the developmental duration of immature stages of H. dimidiata (Fab.)*

The results showed that incubation period of *H. dimidiata* eggs was (4.22±0.15 and 4.21±0.14 days) under long and short photoperiodic regemes. This proved that both photoperiods have no significant effect on egg incubation period.

The duration of first instar larvae were (2.43±0.09 and 2.42±0.09) and second instar larvae were (2.24 ±0.10 and 2.28 ±0.11) days under long and short day photoperiods respectively which are non significantly different from each other. While, the duration of third instar larva were (4.41± 0.16, 3.65± 0.17) and fourth instar larvae (8.0±0.10, 7.21± 0.16) days under long and short photoperiodic regemes, which are significantly different from each other.

**Table 1.** Effect of photoperiod (16: 8) and (8: 16) hours (light: dark) on pre-imaginal development of *H. dimidiata* (Fab.), fed on *S. graminum* aphid**.**

| Developmental stages | 16:8 h (L: D) Photoperiod | 8:16 h (L: D) photoperiod | LSD value(P≤ 0.05) |
|---|---|---|---|
| | Developmental time ±SE | Developmental time ±SE | |
| Egg Incubation | 4.22±0.15a | 4.21±0.14a | 0.4095 |
| 1st instar | 2.43±0.09a | 2.42±0.09a | 0.2646 |
| 2nd Instar | 2.24±0.10a | 2.28±0.11a | 0.3089 |
| 3rd Instar | 4.41± 0.16a | 3.65± 0.17b | 0.4735 |
| 4th instar | 8.0±0.10a | 7.21± 0.16b | 0.3880 |
| Larva | 17.08± 0.12a | 15.56± 0.34b | 0.7251 |
| Pre pupa | 1.05±0.06a | 1.04±0.06a | 0.1792 |
| Pupa | 5.02±0.61a | 5.0 ±0.22a | 0.6098 |
| Egg to adult emergence | 27.55±0.51a | 25.81±0.30b | 1.1933 |

Means with different lower case letters rows wise are significantly different from each other at *P value* ≤ 0.05 (One-way ANOVA), using LSD test.

The results revealed that short day photoperiod accelerated the developmental time of third and fourth instar larvae as compared to long day photoperiod. These results are in accordance with those of Aksit *et al.* (2007) who reported that in insects, the response to photoperiod not necessarily extends to all life stages and the photoperiod responsive phases mostly vary among different species. While Omkar and Pathak (2006) reported that all the developmental stages of aphidaphagous ladybird *Coelophora saucia* were responsive to photoperiod. Chen *et al.* (2017) found that that short day photoperiod significantly extended the developmental time of early life stages of *C. nipponensis* and 1st and 2nd instar larvae completed their development in significantly longer time than those reared under long day photoperiod and thus the the total preadult developmental time was also longer. Danks (1987) reported that photoperiod sensitivity in different stages varies in different insects. Veerman (2001) reported that short day length may often induce diapause, while a long day photoperiodic condition may result in uninterrupted development as well as reproduction. The duration of pre pupae (1.05±0.06 and 1.04±0.06 days) and pupae was (5.02±0.61 and 5.0 ±0.22 days) under long and short photoperiod, respectively. Their differences were non-significant from each other. Which showed that the pupae completed their development independent of photoperiodic effect. The duration from egg to adult emergence was (27.55±0.51)days under longer potoperiod and (25.81±0.30) days under shorter photoperiod, which was significantly different under both photoperiodic regemes. It was found that short day photoperiod accelerated the growth rate from egg to adult emergence (Table 1).

Reznik *et al.* (2015) and Lopatina *et al.* (2007) also reported that short day length accelerated the developmental durations of immature stages in *H. axryidis* species. This ensures the timely development of the diapausing stage prior to the beginning of winter. However, the species specific growth rate is naturally limited and hence fast development often results in small and light adults (Salminen *et al.*, 2012). Some scientsists have also described the photoperiod temperature interactions as well (Tauber *et al.*, 1986; Zaslavski, 1988; Saunders *et al.*, 2002).

*Effect of long and short photoperiod on the biological attributes of adult female ladybird beetle*

*Pre oviposition, Oviposition and Post oviposition period*

The pre-oviposition period of female lady bird beetle was (9.0±0.45 and 11.65±0.26) days under long and short day photoperiodic regems, which was significantly different from each other (Table 2). This revealed that the females matured earlier under lond day photoperiod as compared to short day photoperiod. While on the other hand, the oviposition period was (37.35±1.09 and 35.45±0.75) days with non significantly different from each other. Similarly, the post oviposition period was (7.81± 0.25 and 8.24± 0.33) days under long and short day photoperiod, respectively.

**Table 2.** Effect of photoperiod (16: 8) and (8: 16) hours (light: dark) on biological attributes of adult female *H. dimidiata* fed on *S. graminum* aphid.

| Developmental stages | 16:8 h( L: D ) Photoperiod | 8:16 h( L: D ) Photoperiod | LSD value(P≤ 0.05) |
|---|---|---|---|
| | Developmental time± SE | Developmental time ± SE | |
| Pre-oviposition period | 9.0±0.45a | 11.65±0.26b | 1.0855 |
| Oviposition period | 37.35±1.09a | 35.45±0.75a | 2.7859 |
| Post oviposition period | 7.81± 0.25a | 8.24± 0.33a | 0.8569 |
| Female longevity | 54.16±0.75a | 55.34±0.90a | 2.4611 |
| No of eggs/female | 668.7±32.46a | 612.6±36.08a | 101.96 |
| No of eggs pe female/day | 17.9± 0.98a | 17.5± 0.37a | 2.2055 |

Means with different lower case letters, rows wise are significantly different from each other at *P value* ≤ 0.05 (One-way ANOVA), using LSD test.

The post oviposition period was also non-significantly different under both photoperiods. This shows that short day photoperiod not only accelerated the development of pre imaginal stages but also delayed the reproductive maturation in adult females and oviposition period was reduced as compared to long day photoperiod but still all the females reached maturity under both photoperiods and no diapause was observed at any developmental stage. These results are in conformity with Hodek (2012), Nedved and Honek (2012), who reported that short-day often, accelerates pre imaginal development and delay reproductive maturation in several species of predacious coccinellid beetles.

The results are in contradiction with Semyanov (2009), who determined the effect of photoperiod on *Lies dimidiata*. He found that the maturation of the female occurred simultaneously under different photoperiodic regimes at a temperature of 25 and 30 $^0$C (from clock-round day to clock-round darkness and also under short day (6 and 8) hours light but this also showed that in *L. dimidiata*, photoperiodic adult diapause is absent. In the present study also development was not arrested at any of the stages exposed to short and long day conditions. Ovchinnikova *et al.* (2016) conducted an experiment on *Harmonia axyridis*at 20°C under short (12 h) and long (18 h) dayconditions and evaluated that no diapause was induced under long day conditions but some females under short day conditions entered diapause.

*Effect of long and short photoperiod on Female laby bird beetle longevity and fecundity*

The female longevity was (54.16±0.75 and 55.34±0.90) days under long and short day photoperiod which was non-significantly different under both photoperiods.

The female lady bird beetle fecundity were (668.7±32.46 and 612.6±36.08) eggs under long and short day photoperiod, respectively. Although, the female fecundity was found maximum under long day photoperiod but statistically it was not significantly different from short day photoperiod.

*Effect of long and short photoperiod on eggs per female ladybird beetle per day*

The number of eggs per female beetle per day were (17.9± 0.98) under long photoperiod and (17.5± 0.37) eggs/day under short photoperiodic regemes. The results revealed that both photoperiods did not affect the number of eggs per female/day.

On the basis of the present results *H. dimidiata* should be reared under long day photoperiod, (16: 8) hours light dark during mass rearing program as compared to short day photoperiod (8:16) hours light dark , owing to shorter pre oviposition time leading to early reproductive maturation, longer oviposition time and comparatively high reproductive potential of adult female lady bird beetles.

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