

Temperature-dependent development and immature survival of an aphidophagous ladybeetle, *Propylea dissecta* (Mulsant)

Omkar and A. Pervez

Ladybird Research Laboratory, Department of Zoology, University of Lucknow, Lucknow – 226 007, India

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Abstract: Development and survival of the immature stages of an aphidophagous ladybeetle, *Propylea dissecta* (Mulsant) was investigated at five constant temperatures, viz. 20, 25, 27, 30 and 35°C, using *Aphis gossypii* Glover as prey. Developmental period of all the life stages were significantly affected with change in constant temperature and developmental rate increased with increase in temperature. Theoretical lower thermal threshold for complete development and thermal constant was 10.39°C and 465.11 Day-degrees, respectively. Of the various life stages, first instar larvae were most susceptible to mortality at temperatures between 20 and 30°C, whilst pre-pupae suffered least mortality. Egg-mortality was maximum at 35°C. Female biased sex ratios were obtained at all five temperatures tested with higher proportion of females at the extremes of temperature, thus suggesting that females are more thermal-tolerant. Lowest mortality of immature stages with maximum larval survival and adult emergence was recorded at 27°C, while reverse was the case at 35°C. Thus, 27°C may be considered best for the laboratory rearing of *P. dissecta*.

Key words: *Propylea dissecta*, Aphid, Coccinellidae, ladybeetle, temperature, thermal requirements

1 Introduction

The life of insects is influenced by various environmental factors and temperature is one of the most important and critical of the abiotic factors. It influences the dynamics of insect pests and their predators (HUFFAKER et al., 1999). The predators may be used as potential bioagents for the management of the pest populations. Temperature can act as a stressor and often affects the predation potential and reproduction of the insect predators by setting the limits of predator's biological activity (PONSONBY and COPLAND, 1996, 1998; HUFFAKER et al., 1999; JALALI et al., 1999; ROTT and PONSONBY, 2000; OMKAR and PERVEZ, 2002; ROY et al., 2002). The predators, such as, ladybeetles (Col.: Coccinellidae), have certain thermal limits and effective temperature range for their development and survival, which may vary between different developmental stages (HONEK and KOCOUREK, 1988), species (FRAZER and MCGREGOR, 1992; HONEK, 1996) and even the populations (LEE and ELLIOTT, 1998).

The change in constant temperature within a specific range affects the development and survival rates of ladybeetles, which may be described using specific rate functions (JERVIS and COPLAND, 1996) to predict the activity under field conditions (FRAZER and MCGREGOR, 1992) and to determine optimal conditions for their mass multiplication (RODRIGUEZ-SAONA and MILLER, 1999). Thus, prior to the augmentative release of a

ladybeetle, a study on the influence of temperature and its thermal requirements is of utmost importance.

Propylea dissecta (Mulsant) is an aphidophagous ladybeetle of Oriental region and an important predator of bottle-gourd aphid, *Aphis gossypii* Glover (OMKAR and PERVEZ, 2000a; PERVEZ, 2002). It is abundant in agricultural fields of Lucknow (India) (OMKAR and PERVEZ, 2000a) and is a polymorphic species, exhibiting three morphs, viz. pale, typical and intermediate (R. G. BOOTH, pers. comm.). Earlier, this ladybeetle was largely ignored and there were only the sporadic records of its occurrence in aphid colonies viz., *Aphis affinis* del Guercio and *Myzus persicae* (Sulzer) (SINGH and BALI, 1993) and coconut caterpillar, *Opsinia arenosella* Walker (PILLAI and NAIR, 1986). But recent studies on *P. dissecta* have proved it to be an important biocontrol agent of *A. gossypii* (OMKAR and PERVEZ, 2004; PERVEZ and OMKAR, 2004a). *P. dissecta* can withstand the stresses of prey deprivation by switching to natural foods (OMKAR and PERVEZ, 2003a). Temperature is known to significantly affect its age-specific fecundity and life table parameters (PERVEZ and OMKAR, 2004b). However, there is no information on temperature-dependent development, immature survival and thermal requirements of *P. dissecta*. Thus, the present study has been made to fulfil this gap of information using pale morphs of *P. dissecta* as an experimental model owing to its relative abundance in the field (> 60%).

2 Materials and Methods

2.1 Experimental design

Adults of pale morph of *P. dissecta* were collected from the agricultural fields of Lucknow, India and brought to laboratory. Stock culture was maintained in glass jars (height 15.0 cm × diameter 10.5 cm) containing infestations of aphid, *A. gossypii* on pieces of leaves of bottle-gourd, *Lagenaria vulgaris* Seringe at $25 \pm 1^\circ\text{C}$ and $65 \pm 5\%$ relative humidity (RH).

From the stock, one hundred eggs were selected in a Petri dish (diameter 11.0 cm × height 2.0 cm) and kept in an Environmental Test Chamber (ETC) (CH-6S, Remi Instruments, India) maintained at 20°C and 65% RH. On hatching, egg survival and incubation period was recorded. The first instars were counted and transferred to glass beakers (height 11.0 cm × diameter 8.5 cm). Larval instars were kept singly in each beaker and fed on sufficient amount of *A. gossypii* infested on pieces of leaves of *L. vulgaris*. All observations were made thrice daily and the aphids supply replenished. During larval development, cast exuviae were noted and removed daily in order to determine the number of moults, duration of each stage and the survival rates of various developmental stages. The pupal survival and duration were also recorded. The emerged adult beetles were sexed on the basis of differences in the black and white patches on head and pronotum (OMKAR and PERVEZ, 2000b). Similar experimental setup was followed at 25, 27, 30 and 35°C . Experiment was replicated ten times ($n = 10$).

2.2 Data analysis

The data pertaining to incubation, total larval, pupal and developmental periods were subjected to regression analysis. The developmental rate ($1/D$) of various life stages, i.e. egg, larva, pupa, and complete development was fitted with temperature to obtain the relationship in terms of $1/D = aT + b$, where D = developmental period, T = temperature, and a and b are the regression parameters obtained from the regression equation. Lower development threshold ($t = -b/a$) and thermal constant ($K = 1/a$) were calculated by extrapolating the linear portion of the temperature-developmental rate curve. The method of thermal summation, used here, is based on the linear portion of the temperature-developmental rate relationship for the middle range of temperature (MORRIS and FULTON, 1970). For a few ladybeetles, however, the linearly developed t and K values provide accurate basis for predicting development in the field

(OBRYCKI and TAUBER, 1982; PONSONBY and COPLAND, 1996). The lower developmental threshold (t -values) of different developmental stages were subjected to chi-square goodness-of-fit test. Sex ratio, mortality (%), larval survival (%) and adult emergence (%) were calculated. Data were subjected to one-way ANOVA and compared using Bonferroni's method on statistical software MINITAB.

3 Results

Developmental period of all the immature stages of *P. dissecta* was significantly temperature dependent (table 1). Incubation period of the eggs decreased significantly ($F = 281.33$; $P < 0.001$; d.f. = 4, 45) with increase in temperature. Durations of first ($F = 82.27$; $P < 0.001$; d.f. = 4, 45), second ($F = 85.09$; $P < 0.001$; d.f. = 4, 45), third ($F = 98.48$; $P < 0.001$; d.f. = 4, 45) and fourth ($F = 130.71$; $P < 0.001$; d.f. = 4, 45) instars along with those of pre-pupa ($F = 334.35$; $P < 0.001$; d.f. = 4, 45) and pupa ($F = 22.55$; $P < 0.001$; d.f. = 4, 45) decreased significantly with increase in temperature from 20 to 35°C . A significant decrease in total developmental period ($F = 260.65$; $P < 0.001$; d.f. = 4, 45) was recorded with increase in temperature from 20 to 35°C . The developmental rate of pre-imaginal stages of *P. dissecta* increases with increase in temperature from 20 to 35°C . This increase was linear (fig. 1) in the mid range of temperature with a regression equation, $Y = -0.0218 + 0.00215X$ ($r = 0.99$; $P < 0.001$). The regression equations to predict incubation, total larval, total pupal and total development periods were $Y = 4.680 - 0.085X$ ($r = -0.94$; $P < 0.001$), $Y = 16.000 - 0.310X$ ($r = -0.92$; $P < 0.001$), $Y = 7.500 - 0.146X$ ($r = -0.78$; $P < 0.001$) and $Y = 28.200 - 0.542X$ ($r = -0.91$; $P < 0.001$), respectively. The regression equations to predict the various developmental rates along with lower temperature threshold and thermal constants are mentioned in table 2. The t -values of different developmental stages were not found to be statistically significant ($\chi^2 = 0.95$; NS).

The survival of immature stages (eggs to adult emergence) increased from 37.6 to 62.3% with increase in temperature from 20 to 27°C and thereafter

Table 1. The effect of temperature on the developmental period of *Propylea dissecta*

Duration of development (in days)	Temperature					F-value
	20°C	25°C	27°C	30°C	35°C	
Incubation period	3.14 ± 0.03 a	2.43 ± 0.03 b	2.29 ± 0.05 c	2.04 ± 0.01 d	1.85 ± 0.02 e	281.33*
First instar	2.32 ± 0.05 a	1.73 ± 0.04 b	1.65 ± 0.07 b	1.45 ± 0.03 c	1.20 ± 0.02 d	82.27*
Second instar	2.44 ± 0.05 a	1.78 ± 0.05 b	1.76 ± 0.06 b	1.50 ± 0.04 c	1.28 ± 0.02 d	85.09*
Third instar	2.65 ± 0.05 a	1.81 ± 0.05 b	1.38 ± 0.06 d	1.57 ± 0.04 cd	1.51 ± 0.02 cd	98.48*
Fourth instar	3.13 ± 0.06 a	2.34 ± 0.05 b	2.15 ± 0.03 c	2.13 ± 0.03 c	1.67 ± 0.05 d	130.71*
Total larval period	10.55 ± 0.14 a	7.66 ± 0.08 b	6.94 ± 0.16 c	6.65 ± 0.07 c	5.66 ± 0.07 d	288.48*
Pre-pupal period	1.04 ± 0.02 a	0.56 ± 0.02 b	0.47 ± 0.01 c	0.45 ± 0.01 c	0.40 ± 0.01 d	334.35*
Pupal period	4.06 ± 0.04 a	2.82 ± 0.03 b	2.73 ± 0.04 c	2.51 ± 0.02 c	2.38 ± 0.02 d	22.55*
Total pupal period	5.11 ± 0.33 a	3.38 ± 0.03 b	3.20 ± 0.04 b	2.96 ± 0.02 bc	2.78 ± 0.03 c	41.62*
Total development	18.79 ± 0.40 a	13.47 ± 0.10 b	12.43 ± 0.19 c	11.64 ± 0.06 d	10.30 ± 0.08 e	260.65*

Values are mean ± SE.
 Values denoted by same letter in the row denote that the data are not significantly different.
 * Significant at $P < 0.001$.

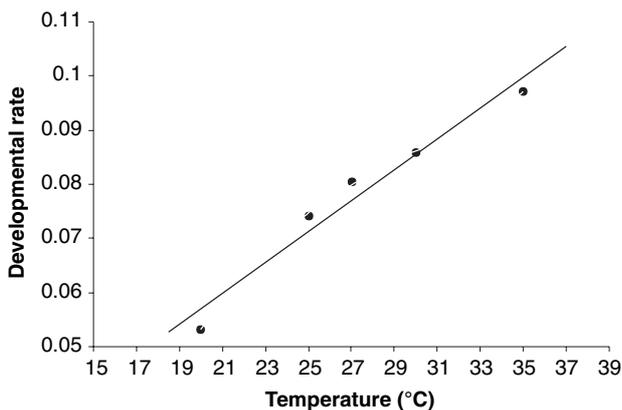


Fig. 1. Best-fit regression line for predicting the development in *Propylea dissecta* calculated from the developmental rates, when the immature predatory stages were fed on *Aphis gossypii* at different temperatures

Table 2. Lower development threshold (*t*), thermal constant (*K*) and regression equations of immature stages of *Propylea dissecta* at different temperatures (r^2 at $P < 0.001$)

Developmental rate (1/ <i>D</i>)	<i>t</i> (°C)	<i>K</i> (°Day)	Regression equation	r^2
Egg	8.29	55.25	$Y = -0.150 + 0.0181X$	0.99
Larva	9.97	262.47	$Y = -0.0380 + 0.0381X$	0.98
Pupa	10.54	119.90	$Y = -0.0879 + 0.00834X$	0.99
Complete development	10.39	465.11	$Y = -0.0218 + 0.00215X$	0.99

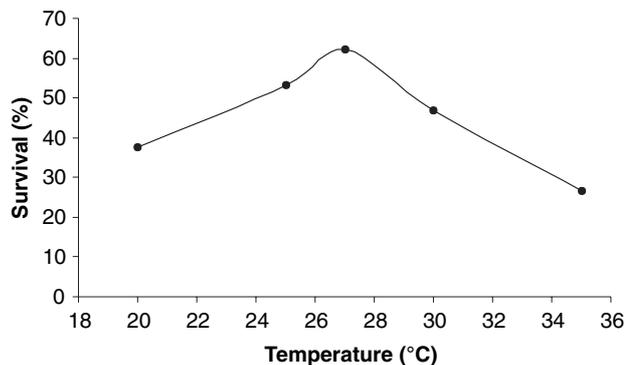


Fig. 2. Survival (%) from egg to adult emergence of *Propylea dissecta* at different constant temperatures, using *Aphis gossypii* as prey

decreased to 26.5% on further increase in temperature upto 35°C (fig. 2). First instar larva suffered highest mortality, while pre-pupal stage had the lowest at all constant temperatures, except at 35°C, where egg mortality was higher than that of first instars (table 3). Percent mortality of immature stages decreased with increase in temperature upto 27°C and thereafter increased with further increase in temperature. Female biased sex ratios were found at both the temperature extremes, i.e. 1 : 1.56 (male : female) at 20°C and 1 : 1.63 at 35°C (table 4). Both, larval survival ($F = 33.94$; $P < 0.001$), and adult emergence ($F = 8.09$; $P < 0.001$) were significantly affected by changes in constant temperatures (table 4) and the values were maximum at 27°C, whilst minimum at 35°C.

Table 3. Percent mortality of each immature stage of *Propylea dissecta* at various constant temperatures ($n = 1000$)

Stage	Temperature				
	20°C	25°C	27°C	30°C	35°C
Eggs	29.7 (703)	14.80 (852)	13.30 (867)	18.30 (817)	38.50 (615)
First instars	30.63 (487)	21.36 (670)	13.49 (750)	23.50 (625)	34.80 (401)
Second instars	7.19 (452)	6.72 (625)	6.40 (702)	8.16 (574)	12.22 (352)
Third instars	6.86 (421)	6.08 (587)	4.99 (667)	6.10 (539)	8.52 (322)
Fourth instars	4.99 (400)	4.43 (561)	3.00 (647)	5.01 (512)	8.39 (295)
Pre-pupae	1.25 (395)	1.96 (550)	0.77 (642)	2.34 (500)	3.73 (284)
Pupae	4.81 (376)	3.27 (532)	2.96 (623)	6.00 (470)	6.69 (265)

Data in parentheses denote the number of immature stages survived.

Table 4. Sex ratio, larval survival (%) and adult emergence (%) of *Propylea dissecta* at five temperatures

	Temperature					<i>F</i> -value
	20°C	25°C	27°C	30°C	35°C	
Sex ratio (male : female)	1 : 1.56	1 : 1.04	1 : 1.08	1 : 1.08	1 : 1.63	–
Larval survival (%)	61.53 ± 1.57 b	69.06 ± 2.18 c	77.10 ± 1.11 d	64.87 ± 1.08 b	51.51 ± 1.88 a	33.94*
Adult emergence (%)	86.87 ± 1.27 b	90.47 ± 0.95 cd	93.21 ± 0.79 d	88.72 ± 0.87 bc	84.03 ± 1.70 a	8.09*

Values are mean ± SE.
 Values denoted by same letter in the row denote that the data are not significantly different.
 * Significant at $P < 0.001$.

4 Discussion

Empirical data revealed that exposure to higher temperature reduces the developmental period of different immature stages and consequently increases the development rate, as also reported in other studies (OBRYCKI and TAUBER, 1982; ALI KHAN and YOUSUF, 1986; ORR and OBRYCKI, 1990; PONSONBY and COPLAND, 1996). Moreover, significant differences in the developmental period of each life stage suggest that all life stages of *P. dissecta* were highly sensitive to changes in constant temperatures. This, however, indicates the lesser ecological plasticity of *P. dissecta* as compared with successfully established ladybeetles, such as, *Coccinella septempunctata* Linnaeus (WHEELER and HOEBEKE, 1995). Whereas, the low values of developmental threshold (t -value) indicate the capability of immature stages to develop at low temperatures.

The t -value needed to incubate and hatch the eggs of *P. dissecta* was lowest ($t = 8.29^{\circ}\text{C}$) as compared with those required by other immature stages to develop. This value was even lower than that required (11.0 – 11.2°C) to undergo hatching of larvae of a closely related species, *Propylea quatuordecimpunctata* (Linnaeus) (BAUMGÄRTNER et al., 1987; HONEK and KOCOUREK, 1988). This suggests the possible development at lower temperatures with relatively lesser chilling effects. Theoretical lower threshold for complete development in *P. dissecta* was found to be lower ($t = 10.39^{\circ}\text{C}$) than those of other coccinellids studied viz., *Hippodamia convergens* Guerin Meneville (OBRYCKI and TAUBER, 1982), *P. quatuordecimpunctata* (BAUMGÄRTNER et al., 1987; HONEK and KOCOUREK, 1988) and *Chilocorus nigritus* (Fabricius) (PONSONBY and COPLAND, 1996; OMKAR and PERVEZ, 2003b).

There exists rate isomorphy, i.e. the proportion of total developmental time spent in a particular developmental stage does not change with temperature, in most insects and mites (JAROSIK et al., 2002; SRIVASTAVA and OMKAR, 2003). This could be the case only if the t -values are same for all developmental stages. In the present study, the differences in t -values for various developmental stages were non-significant, supporting rate isomorphy in *P. dissecta*. The existence of rate isomorphy could be of great practical importance, as it may determine the timing of life-history events and thermal requirements of development (JAROSIK et al., 2002).

Of the various stages of *P. dissecta*, the first instars, i.e. neonates, suffered highest mortality between 20 and 30°C with the extremes of temperature tending to be deleterious for their survival. Relatively high mortality of first instars as compared with other life stages was also reported in *C. nigritus* (PONSONBY and COPLAND, 1996), which was possibly because of their small size with thin and soft cuticle making them more vulnerable to physical stressors. Whereas, the increasingly thicker cuticles of later instars and thick pupal case possibly provide a shield, which protects the later stages from the extremes of temperature. Eggs suffered maximum mortality at higher extreme of temperature (35°C) and those failed to hatch, became flaccid and wrinkled. It appears that the egg-contents have been

denatured or even burnt as their colour changed from bright yellow to pale yellow.

High female-biased sex ratios were found at lower and higher temperature extremes, which suggests that females are more thermal tolerant than males. The female biased sex ratio was also reported in four Chilocerini and one Coccinellini species, which was ascribed to certain genetic factors in the ladybeetles that proved lethal to males when exposed to the stressors (HENDERSON and ALBRECHT, 1988).

The larval survival increased with increase in temperature from 20 to 27°C and thereafter decreased with further increase in temperature. This indicates that larvae were struggling to withstand the heat stress at upper extreme of temperature, when their internal body temperature possibly approaches the lethal point (JAMES, 2001). Adult emergence and immature survival also increased in the middle ranges of temperature with peaks at 27°C and decline at temperature extremes. Thus, 27°C may be regarded as the most suitable temperature for mass-rearing of the species.

It may be concluded that: (i) temperature significantly affected the developmental periods of various life stages of *P. dissecta*, (ii) low values for lower thermal threshold for development of various life stages suggest that they may develop at low temperatures with lesser chilling injuries, (iii) first instar was more vulnerable to low and mid-temperature ranges than other life stages, whilst eggs to upper extreme of temperature, (iv) adult females were more thermal-resistant than males, (v) 27°C is the optimum temperature to facilitate development and immature survival of *P. dissecta*. Thus, the findings encourage the biocontrol programmes to exploit *P. dissecta* for the management of aphid populations and to establish the predator in new zoogeographical habitats.

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Author's address: Omkar (corresponding author), Ladybird Research Laboratory, Department of Zoology, University of Lucknow, Lucknow – 226 007, India. E-mail: omkaar55@hotmail.com