



Short Communication

Temperature-dependent development of overwintering *Seriginus montela* Gray (Lepidoptera: Papilionidae) pupae and its validationKang-Woon Lee^a, Dong-Jae Lee^a, Jeong Joon Ahn^{b,*}^a Holoce Ecosystem Conservation Research Institute (HECRI), Gapchon-myon, Dunae-myon and Wuchon-myon, Hweongsung-gun, Gangwon-do, Republic of Korea^b Crop Protection Division, Department of Crop Life Safety, National Academy of Agricultural Science, 126 Suin-ro, Kweonseon-gu, Suwon, Gyeonggi-do 441-707, Republic of Korea

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ABSTRACT

The developmental time and survival of overwintering *Seriginus montela* Gray pupae were studied at four constant temperatures (15.0, 20.0, 25.0, and 30.0 °C), 40 ± 10% relative humidity, and 10:14 h light:dark cycle. The developmental time of both sexes decreased with increasing temperature between 15.0 °C (70.18 days for females and 55.28 days for males) and 30.0 °C (19.60 days for females and 13.78 days for males). The development periods of females were longer than those of males at each constant temperature. The relationship between the developmental rate and temperature was fitted by a linear model and a nonlinear developmental rate model (Lactin 1). The mortality of overwintered *S. montela* pupae was lowest at 25.0 °C (16.7%) and highest at 15.0 °C (36.7%). The lower developmental thresholds were 12.38 and 12.16 °C for females and males, respectively. The distribution of development completion for females and males was described by the two-parameter Weibull distribution equation ($r^2 = 0.87$ for females and $r^2 = 0.94$ for males). The date for the cumulative 50% adult emergence was within one or two days of that predicted using the Lactin 1 model. The temperature-dependent developmental model for *S. montela* could be applied to predict the timing of spring emergence in different geographical locations and will be helpful in developing a full-cycle phenology model for *S. montela*.

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Introduction

Insects are sensitive to temperature and respond rapidly to climate change with high mobility and adaptation (Angilletta, 2009). Among them, butterflies have been recognized as good model organisms in the study of coevolution (Gilbert, 1971), foraging behavior (Singer et al., 1994), metapopulation dynamics (Hanski, 2011), plasticity (Gotthard et al., 1999), and speciation (McMillan et al., 1997). Butterflies, typically, have characteristics, including high reproductive rates and short life cycles, which make them good candidates for understanding and predicting the biological outcomes of a changing climate (Walker, 1991; Crozier, 2002; Hellmann, 2002; Walker and Rotherham, 2010).

The dragon swallowtail *Seriginus montela* Gray (Lepidoptera: Papilionidae) is widely distributed from central China to the Amur and Ussuri regions (Igarashi, 2003). *S. montela* larvae feed on *Aristolochia contorta* (Aristolochiaceae) and *A. debilis*. The sexual polymorphism of *S. montela* adults is a good biological characteristic of the species. The body and wings of females are dark-brown and those of males are

whitish. It has three generations per year in temperate regions and overwinters as a pupa.

The overwintering of insects in diapause is a fascinating process involving behavioral, biochemical, and morphophysiological changes (Leather et al., 1993). Forecasting the emergence of overwintering populations would help to conserve and maintain the threatened species.

The purpose of this study was to investigate the temperature-dependent development and survival of overwintering *S. montela* pupae and to develop a temperature-dependent developmental model for this species.

Materials and methods

Description of study site

This experiment was conducted at Holoce Ecosystem Conservation Research Institute (HECRI) in Gapchon-myon, Dunae-myon, Wuchon-myon, Hweongsung-gun, and Gangwon-do, Korea. HECRI covers a relatively low, hilly district where 95% of the area is between 400 and 475 m above sea level (N37°30'–N37°31', E127°–E128°10'). The edges of the study site are adjacent to a stream or mountain. The study site was divided into three sections—whole site landscape,

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Table 1The mean adult emergence times (days) (mean $\pm$ SEM) and mortality (%) of overwintering *Seriginus montela* pupae at different constant temperatures.

	Temperature			
	15 °C	20 °C	25 °C	30 °C
Female	70.18 $\pm$ 4.088a	35.12 $\pm$ 4.774b	26.44 $\pm$ 3.260b	19.60 $\pm$ 5.102b
Male	55.28 $\pm$ 5.247a	32.33 $\pm$ 1.268b	15.07 $\pm$ 1.501c	13.78 $\pm$ 2.810c
Combined	64.39 $\pm$ 3.535a	33.45 $\pm$ 1.933b	19.52 $\pm$ 1.912c	16.84 $\pm$ 2.848c
M	36.7	26.7	16.7	26.7

M: mortality (%). Female: $F_{3, 34} = 18.82$, $p < 0.0001$, Male: $F_{3, 38} = 58.02$, $p < 0.0001$, Combined: $F_{3, 76} = 50.58$, $p < 0.0001$. Means with the same letter in row are not significantly different ($p < 0.05$, Tukey studentized range test).

microhabitats, and semi-wild insectariums, and these sections have been manipulated, managed, and conserved since 1997.

Insect colony

The overwintering pupae of *S. montela* were originally collected from HECRI microhabitats, which have been maintained for 16 years starting in 1997. Adults of *S. montela* obtained from the previous year's overwintering population were released to oviposit in the dome-type semi-wild insectariums (8 m in height $\times$ 20 m in diameter) and microhabitats. Newly hatched first instar larvae were transferred to cages (40 $\times$ 50 $\times$ 45 cm, width $\times$ length $\times$ height) and *A. contorta* was supplied for food. *S. montela* has three generations per year from April to October, and overwinters as a pupa from October to April.

Developmental experiment

Final instar larvae of *S. montela* were collected in insectariums and microhabitats from late September to early October and transferred to a cage (40 $\times$ 50 $\times$ 45 cm, width $\times$ length $\times$ height) for pupation. Detached pupae from each cage were randomly attached to a slanted

emergence plate from October to November. The developmental times of *S. montela* from overwintering pupa to adult emergence were investigated at temperatures of 15.0, 20.0, 25.0, and 30.0 °C (± 1.0 °C) and a photoperiod of 10:14 h (light:dark, L:D) from January 1 to March 15 in 2012. Sample sizes were greater than 15 pupae per treatment. Observations were made two times per day from 12:00 to 13:00 and from 22:00 to 23:00 because *S. montela* usually emerges on the same time schedule. The developmental times of each overwintering pupa were recorded and newly emerged adults were separated from the plate.

Data analysis

The developmental times of overwintering *S. montela* pupae were analyzed using the PROC GLM in SAS (SAS Institute, 2002). The statistical differences of developmental periods among the temperatures tested and between sexes were evaluated by two-way analysis of variance. Significant differences among multiple means of treatments were determined using Tukey's studentized range test.

The relationship between developmental rates (1/developmental periods) and temperatures was analyzed with linear and nonlinear functions. The linear model is $r(T) = a + bT$ where $r(T)$ is the

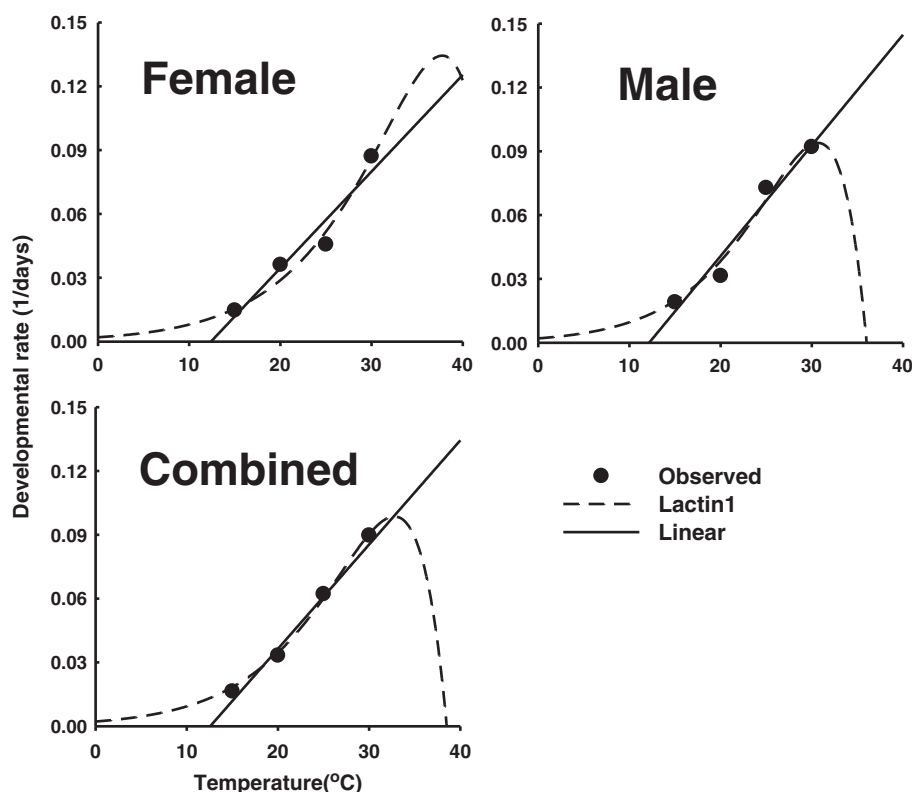


Fig. 1. Linear and nonlinear function fit to the data of developmental rates (day^{-1}) of *Seriginus montela* female, male and combined sexes, respectively.

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