果樹研究　Bull. Natl. Inst. Fruit Tree Sci. 10：1 ～ 8，2010

原著論文

Effect of constant temperatures on the development of peach fruit moth, *Carposina sasakii* (Lepidoptera: Carposinidae) [†1]

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Abstract

The temperature-dependent development of *Carposina sasakii* was investigated to estimate the lower developmental thresholds and thermal constants. *C. sasakii* was reared in immature apple fruits ('Fuji') for larval stage and in soil for pupal stage at 7 constant temperatures in the laboratory. Larvae and pupae survived well in fruits and soil at each temperature, except for pupae at 25°C. Developmental periods from egg to adult in female was longest at 15°C (90.3 ± 0.9 days) and shortest at 27.5°C (27.4 ± 0.2 days). The regression equations between developmental rates from egg to adult molt (y) and temperature (x) were estimated as y = - 0.0206 + 0.0021x (r^2=0.98) for females and y = - 0.0203 + 0.0020x (r^2=0.98) for males. The lower developmental thresholds and thermal constants in the immature stage were estimated as 10.0°C and 484.2 degree-days for females and 9.9°C and 488.0 degree-days for males, respectively.

Keywords: peach fruit moth, apple, development, lower developmental threshold, thermal constant

Introduction

The peach fruit moth, *Carposina sasakii* Matsumura, is one of serious pests of pome fruits in eastern Asia (Narita, 1986). Larvae of the moth injure pulp of the fruit and leave feces in the core of fruits during development. Growers cannot completely eliminate infested apples because infested apples sometimes do not show any signs on the surface of fruits. One of the significant signs is a hole created by the matured larvae when they escape from the fruits onto the ground. The matured larvae form two types of cocoons under the ground depending upon season and their voltinism

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（2008 年 11 月 6 日受付・2009 年 10 月 13 日受理）
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(Narita, 1986). They form the spindle-shaped cocoons for immediate pupation before mid summer and the compressed cocoons for overwintering at the late summer and autumn. In the bivoltine population, the mature larvae pupate immediately in the cocoons before mid summer, and adults emerge as the next generation and start oviposition on the fruits after mating (Kajino and Nakao, 1978; Narita and Takahashi, 1978; Narita, 1987). In the univoltine population and the second generation of the bivoltine population, mature larvae overwinter in the cocoons at the late summer and autumn, and pupate at the next spring. Since the larvae in the fruits and under the ground are hardly killed by insecticides, adults and eggs on the fruits are the target stage for control.

Insecticides are usually sprayed at the time of the peak of adult emergence. The emergence of *C. sasakii* is monitored by the number of adult males trapped on the sticky plates with sex pheromone. However, the adult males are not effectively attracted by the sex pheromone on the sticky plates when orchards are filled with sex pheromone for mating disruption. It is difficult to monitor the adult emergence in the orchards with mating disruption (Kydonieus and Beroza, 1982).

As an alternative method, the occurrence of the first generation of *C. sasakii* was predicted by the time of adult emergence in the green house (Kajino and Nakao, 1972). Quantification of the relationship between development and temperatures is useful to predict the seasonal occurrence. Developmental period from egg to adult was estimated by observing naturally infested fruits in the field (Suzuki and Kumakura, 1969), but this estimation may not be accurate because larvae do not develop well in the fruits on the trees (Ishiguri and Toyoshima, 2006). Recently, temperature-dependent development of *C. sasakii* was studied with immature apple fruits in the laboratory (Kim et al, 2001; Kawashima, 2008). In those studies, however, the survival rates of larval stage were low (24.2% to 53.0%, in the latter study), and the lower developmental thresholds and thermal constants estimated in those studies may not be reliable as basic developmental characteristics of *C. sasakii*. In this study, therefore, *C. sasakii* larvae were reared in the fresh immature fruits of apple to obtain data with high survival rates, and the temperature-dependent development was investigated for the estimation of the lower developmental thresholds and thermal constants.

Materials and Methods

A laboratory strain of *C. sasakii*, collected in Aomori Prefectural Agriculture and Forestry Research Center, had been reared in our laboratory at 25℃±1℃ with 16L8D photoperiod. Immature apple fruits ('Fuji'), collected during the end of June and the beginning of July, were used for maintaining culture.

Mated females were transferred individually from culture population into a Petri dish (9 cm diam., 2 cm height) where a paraffin paper with many pleats (Kawashima and Yamada, 1984) was set for a substance of oviposition. Humidity was not controlled but water (2 ml in a small lid) was supplied to avoid desiccation in the Petri dish. After 24 hr, the female was removed from the Petri dish, and eggs on the paper were reared at 7 different temperatures (15, 17.5, 20, 22.5, 25, 27.5 and 30 ºC) in the growth chamber with 16L8D photoperiod. Just before their hatch, the paper with eggs were cut into pieces with 1 or 2 eggs, and each piece was fixed with an adhesive tape on the surface of an immature fruit. Immature fruits were maintained in the plastic cup with a lid (6 cm diam., 8 cm height). Humidity in the plastic cups was not controlled. Sixty fruits were prepared as a unit for each temperature.

Observation was conducted daily to determine each developmental period. Egg hatch and larval penetration into fruits were observed under binocular microscope. When matured larvae escaped from the fruits, they were individually transferred onto the soil (Sakata-Soil-Mix, Sakata Seed Corporation) in the plastic cup (6 cm diam., 4 cm height) for cocooning. Their sexes were also checked after emergence.

Periods of larval and pupal stages were determined between the date of hatch and the date of escape from fruits, and between the date of the escape and the date of emergence, respectively. Shriveled eggs and larvae glued onto the adhesive tape were omitted from the results. Developmental periods of individuals that did not reach to adult were also omitted from analysis. The effect of temperature on the development was analyzed in each stage and sex with linear regression (using JMP® software, version 6.0.2, from SAS, Cary, NC, USA) to determine the lower developmental threshold (developmental zero, low-temperature threshold) and thermal constant (total accumulate temperature, degree-days). Developmental rates were expressed as the reciprocal of developmental periods (days) of eggs, larvae, pupae and total, respectively. The lines were obtained by regressing the developmental rates on temperature. The lower developmental thresholds and the thermal constants were estimated by solving the -intercept/slope and 1/slope of the fitted equation for each stage and total development, respectively.

Results

Survival rates are shown in Table 1. The smallest hatchability was shown at 25 °C (73.0%) and the highest at 30 °C (94.9%). Survival rates of larvae and pupae were ranged from 75.8 % to 96.3 %, and from 48.1 % to 98.1 %, respectively. Less than two larvae were glued onto the adhesive tape and less than three eggs were shriveled in each temperature, except for the 13 eggs squashed in the rearing unit of 30 °C. Only 34 % of individuals completed their development at 25 °C. Hatchability and survival rate of pupae were high at 20 °C and 30 °C. Sex ratios (female ratio = ♀/(♀+♂) were ranged from 39.6 % to 51.9 %, but were not significantly different from the even sex ratio (female : male = 1:1) (Pearson's *chi*-square test, p>0.05 for each temperature).

Developmental periods of each stage and total (from egg to adult emergence) are shown in each sex in Table 2. Periods were not different in sex (F=0.30, p=0.59) but significantly different by temperatures (F=7557.56, p<0.01). The developmental periods decreased with increasing temperatures, except for larval stage. The egg developmental periods ranged from 18.9 days at 15°C to 5.3 days at 30°C. The larval periods decreased from 38.3 days at 15°C to 12.1 days at 27.5°C, and increased to 13.8 days at 30°C. The pupal periods ranged from a maximum of 33.1 days at 15°C to minimums of 9.1 days at 27.5°C and 9.2 days at 30°C. Under the constant thermal condition, female and male moths reached adult within a month (minimum) at 27.5°C, and for 3 months (maximum) at 15°C. Larval period was slightly longer than pupal period, and was twice as long as egg period in both sexes and all temperatures.

Removing the developmental rate at 30°C, the developmental rates of six temperatures were fit on the regression line (Fig. 1), and the regression equations of developmental rates (y) against temperatures (x) were statistically significant (Table 3). The lower developmental thresholds in female were 8.0°C (r^2=0.95) for egg, 10.3°C (r^2=0.93) for larval, 10.5°C (r^2=0.98) for

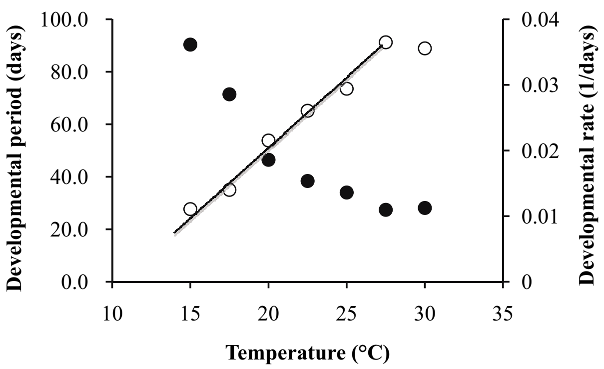

Fig. 1. Developmental periods (●) and developmental rates (○) of females of *Carposina sasakii*. The lower developmental t hreshold and thermal constant in female were estimated as 10.0°C and 484.2 degree-days by the regression equation of developmental rates from egg to adult (y) against temperatures (x) y = -0.0206 + 0.0021x (r^2=0.98).

pupal developmental stages, and 10.0°C (r^2=0.98) for total developmental period. The thermal constants of egg, larva and pupa in female were 118.8, 209.9, and 157.7 degree-days, respectively. The thermal constants from egg to adult were 484.2 degree-days for females and 488.0 degree-days for males, respectively.

Discussion

In the codling moth, *Cydia pomonella*, which is a serious pest of apples (Dorn et al., 1999) the effects of temperature on development and behavior were intensively investigated (Howell and Neven, 2000; Kuhrt et al., 2005 and 2006). However, there are only two studies about the temperature-dependent development of *C. sasakii* in the laboratory (Kim et al., 2001; Kawashima, 2008), even though *C. sasakii* is also serious pest of apples in Asia. In comparison with the two previous studies, *C. sasakii* developed well in immature apple fruits at each temperature in this study, except for the survival rate of pupa at 25°C. Survival rates in this study (Table 1) were higher than those in the previous study, which showed that the survival rates of larva were 24.2% (the lowest rate) at 14°C and 34.8% at 20°C (Kawashima, 2008). Larval periods in this study (Table 2) were shorter than those in the previous studies, which showed that the larval periods were 27.4 ±3.8 days (n=17) in female and 26.1±3.4 days (n=28) in male at 20°C (Kawashima, 2008), and were 26.02 ± 0.22 (n=128) in both sexes at 20°C (Kim et al., 2001). Pupal periods in this study were also shorter than those in the previous studies, although the egg period and hatchability in this study were similar to those. As a result, the thermal constants in this study (Table 3) were smaller than those of larval and pupal stages in the previous study (Kawashima, 2008).

The cause of low survival rate of pupa at 25°C was not clarified in this study. Although more males than females emerged when more than 10 individuals were reared together in a fruit (Kawashima, 2008), sex specific mortality and delay of development were not detected when larvae were reared individually (Tables 1 and 2). Female larvae may be sensitive to the crowded condition, or males may be competitive against females.

Developmental characteristics of *C. sasakii* obtained in this study are useful to investigate the factors affecting the mortality of larvae and the delay of development in the field. The developmental rate linearly increased with increasing temperature except for 30°C (Fig.1). This trend is similar to that of *C. sasakii* in Korea (Kim et al., 2001), but is different from that of C. pomonella. The developmental rate of *C. pomonella* increased with

Table 1. Survival rates of *Carposina sasakii* when reared with immature apple fruits ('Fuji') under constant thermal condition

Temperature (°C)	n[z]	Hatchability (%)	Survival rate of larva[y] (%)	Survival rate of pupa[x] (%)	Survival rate from egg to adult emergence (%)	Sex ratio[w] (♀/♀+♂)(%)
15	70	78.6	87.3	72.9	50.0	42.9
17.5	70	88.6	75.8	83.0	55.7	43.6
20	70	91.4	85.9	90.9	71.4	51.1
22.5	78	76.9	90.0	98.1	67.9	39.6
25	74	73.0	96.3	48.1	33.8	48.0
27.5	75	80.0	90.0	98.1	70.7	51.9
30	50	94.0	80.9	94.7	72.0	47.2

[z] The number of individuals without the eggs shriveleld on the paraffin paper and the larvae glued onto the adhesive tape.
[y] The rates divided the number of matured larvae by the number of larvae entered into fruits.
[x] The rates divided the number of adults by the number of matured larvae.
[w] Sex ratios were not significantly different from the even sex ratio (female : male = 1:1) (Pearson's *chi*-square test, p>0.05 for each temperature).

Table 2. Developmental periods of *Carposina sasakii* when reared with immature apple fruits ('Fuji') in the laboratory

Sex	Temp. (°C)	n^z	Egg	Larva^y	Pupa^x	Total
Female	15	15	18.9 ± 0.2	38.3 ± 1.1	33.1 ± 0.4	90.3 ± 0.9
	17.5	17	12.9 ± 0.1	35.2 ± 0.5	23.3 ± 0.2	71.4 ± 0.5
	20	24	9.0 ± 0.0	20.9 ± 0.3	16.5 ± 0.1	46.5 ± 0.4
	22.5	21	8.2 ± 0.1	16.9 ± 0.3	13.3 ± 0.1	38.4 ± 0.2
	25	12	7.9 ± 0.1	15.0 ± 0.4	11.1 ± 0.4	34.0 ± 0.6
	27.5	27	6.0 ± 0.0	12.1 ± 0.2	9.2 ± 0.1	27.4 ± 0.2
	30	17	5.3 ± 0.1	13.8 ± 0.3	9.1 ± 0.2	28.1 ± 0.5
Male	15	20	18.9 ± 0.1	38.2 ± 0.8	33.5 ± 0.4	90.5 ± 0.6
	17.5	22	13.0 ± 0.0	33.8 ± 0.3	23.6 ± 0.2	70.4 ± 0.2
	20	23	9.0 ± 0.0	20.3 ± 0.2	16.9 ± 0.2	46.2 ± 0.3
	22.5	32	8.1 ± 0.0	16.8 ± 0.2	13.4 ± 0.1	38.3 ± 0.1
	25	13	8.0 ± 0.2	14.5 ± 0.4	11.4 ± 0.3	33.8 ± 0.5
	27.5	25	6.0 ± 0.0	11.8 ± 0.2	9.7 ± 0.1	27.5 ± 0.1
	30	19	5.1 ± 0.1	12.8 ± 0.3	9.5 ± 0.1	27.4 ± 0.3

Mean ± SE. ANOVA was conducted for sex and temperature. The factor 'sex' was not significant (F=0.30, p=0.59), but 'temperature' was significant (F=7557.56, p<0.0001).
^{z} The number of individuals reached to adult.
^{y} Periods of larval stage were determined between the date of hatch and the date of escape from fruits,
^{x} Periods of pupal stage were determined between the date of escape and the date of emergence.

Table 3. Regression equations, lower developmental thresholds and thermal constants of each developmental stage of *Carposina sasakii*

Sex	Stage	Regression equations^z	Fs^y	r^y	T_0 (°C)^x	K(degree-days)^w
Female	egg	y = -0.0676 + 0.0084x	2005.73	0.95	8.0	118.8
	larva	y = -0.0488 + 0.0048x	1600.15	0.93	10.3	209.9
	pupa	y = -0.0665 + 0.0063x	4903.00	0.98	10.5	157.7
	egg to adult	y = -0.0206 + 0.0021x	4960.55	0.98	10.0	484.2
Male	egg	y = -0.0696 + 0.0085x	2402.22	0.95	8.2	117.7
	larva	y = -0.0506 + 0.0049x	2700.83	0.95	10.3	204.2
	pupa	y = -0.0603 + 0.0060x	5731.39	0.98	10.1	167.6
	egg to adult	y = -0.0203 + 0.0020x	8161.18	0.98	9.9	488.0

^{z} Regression equations were estimated by developmental rates (y) against temperatures (x).
^{y} Variance ratio. All regressions are statistically significant (df=1, p<0.01).
^{x} Lower developmental threshold.
^{w} Thermal constant.

temperature and reached a maximum at approximately 30°C (Howell and Neven, 2000). *C. sasakii* may be more sensitive to high temperature during summer than *C. pomonella*. Larval survival of *C. sasakii* in apple fruits was low (0% - 20.8%) in the field, irrespective of the oviposition date between early June and late August (Kim and Lee, 2002; Ishiguri and Toyoshima, 2006). If the fruits were picked from trees, larvae developed well in the fruits even under the field condition (Ishiguri and Toyoshima, 2006). Physical and physiological factors of fruits on trees affecting development of larvae must be investigated in the laboratory and the field.

Seasonal prevalence of *C. sasakii* is variable depending on the area in Japan (Chiba and Kobayashi, 1985), and is usually described as bivoltine, univoltine, or mixed types. The strain we used in this study was discriminated as the bivoltine type (Ishiguri and Shirai, 2004). The development of other types should be compared with those in this study to clarify the relationship between the response to temperature and the seasonal prevalence in the field.

Acknowledgments

We thank Mr. Y. Ishiguri for his kindness to provide C. sasakii strain and for his valuable comments to this manuscript. This research was supported by a Grant-in-Aid for Research Program (the research project for utilizing advanced technologies in agriculture, forestry and fisheries, No. 1911) from the Ministry of Agriculture, Forestry and Fisheries of Japan.

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モモシンクイガの発育に及ぼす恒温条件の影響

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摘　　要

発育を完了した個体のデータを元にモモシンクイガの発育零点と有効積算温量を推定するため，収穫した幼果（‘ふじ’）と園芸培養土でモモシンクイガを飼育して７種の恒温条件における発育特性を調査した．25℃における蛹ステージを除いて，幼虫と蛹はよく発育した．卵から羽化までの発育期間は，15℃でもっとも長く（90.3 ± 0.9 日），27.5℃ でもっとも短かった（27.4 ± 0.2 日）．温度（x）に対する卵から羽化までの発育速度（y）の回帰式は，雌で y = -0.0206 + 0.0021x（r^2=0.98），雄で y = -0.0203 + 0.0020x（r^2=0.98）であった．全発育間における発育零点と有効積算温量は，雌で 10.0℃と 484.2 温日度，雄で 9.9℃と 488 温日度であった．