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The effect of temperature on development and fecundity of ***Phyllocnistis citrella*** Stainton (Lepidoptera: Gracillaridae)*

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Summary

Development and fecundity of ***Phyllocnistis citrella*** Stainton (Lepidoptera: Gracillaridae) were recorded at five constant temperatures ranging from 15 to 35±1°C in 5°C increments and a varying temperature of 25-35°C. Developmental time (egg to adult) of ***P. citrella*** decreased with increasing temperatures, ranging from 51.7 days at 15°C to 10.1 days at 35°C. The immature mortality was highest at 15°C and lowest at 30°C. No eggs were deposited at 15°C. At all temperatures studied the females lived statistically longer than the males. Net reproductive rate (R_o) was the highest at 30°C with 50.19 female/ female. The highest intrinsic rate of increase (r_m) were 0.258 and 0.260 females/female/day at 30°C and 35°C, respectively, but they found at the same statistical group. Results concluded that a temperature of 30°C is optimal for ***P. citrella.***

Key words: Citrus leafminer, ***Phyllocnistis citrella***, development, fecundity, life tables
Anahtar sözcükler: Turunçgil yaprak galerigüvesi, ***Phyllocnistis citrella***, gelişme süresi, üreme gücü, yaşam çizelgeleri

Introduction

Since the first appearance of the Citrus leafminer, ***Phyllocnistis citrella*** Stainton (Lepidoptera: Gracillaridae) in the east Mediterranean region of Turkey in summer 1994

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(Uygun et al., 1995; 2000), it continues to be one of the most serious pests of citrus trees younger than 5-6 years. As in the other countries in which the pest was detected for the first time, studies in Turkey have primarily concentrated upon the control of this pest (Uygun et al., 1996 a; 1996 b; Yumruktepe et al., 1996). In spite of the wide distribution and apparent economic importance of ***P. citrella***, there is limited information on its biology and ecology. This knowledge would prove useful in improvement of control programs directed against the pest in citrus orchards.

The present study was designed to study the effect of different temperatures on development and fecundity of ***P. citrella*** under controlled laboratory conditions.

Material and Methods

Insect rearing

Phyllocnistis citrella was obtained from citrus orchards in east Mediterranean region and reared on ***Citrus aurantium*** at 27±1°C, 80±5 % RH and a photoperiod of 16 hours artificial light in a climatic room. New plants, suitable for the pest to lay eggs upon, were replaced twice weekly whilst those exhibiting hatched pupae were removed.

Development and mortality rate of immature stages

The saplings with fresh leaves were covered with tulle cages and 20-30 ***P. citrella*** adults were released into the cages. Every other day, the adults were suctioned and the leaves were examined under the binocular microscope. Individual eggs were kept on each leaf and numbered. The duration and the mortality at different developmental stages were recorded until eclosion by daily observations at all temperatures (15, 20, 25, 30, 35±1°C constant and 25-35°C varying temperatures).

Longevity and fecundity

Following eclosion, the same individuals were kept at specific temperatures during the immature stages and remained in those conditions in the same tulle cages. To determine the daily number of eggs laid the adults were taken from the cages with a suction trap and released to new saplings everyday. Dead individuals were collected and the sex ratio was determined after preparation of genitalia. The number of eggs laid per female was calculated by dividing the number of eggs per cage by the number of females. The duration of preoviposition, oviposition and postoviposition periods, longevity and the number of eggs per cage were recorded through daily observation until all adults had died.

Data analysis

The data obtained from daily observation were used to construct life tables according to Southwood (1978) and Sokal and Rohlf (1981).

Differences in developmental time, longevity and fecundity were tested by analysis of variance (ANOVA) and Fisher's Least Significant Difference test. A linear technique was employed to compute the lower developmental thereshold of the developmental stages (egg to adult) by using the growth rate data as a dependent variable and temperature as an indipendent variable. The lower development thereshold was determined as the intercept of the regression line with the X-axis. The degree-day (DD) requirements were calculated as the reciprocal value of the slope of the linear regression line. The confidence of the differences between the intrinsic rates of increase (r_m) values were tested by STUDENT's t-test.

Results

Development and mortality rate of immature stages

The length of egg, larval and pupal periods of ***P. citrella*** decreased with increasing temperatures (Table 1). The total developmental time from egg to adult ranged from 51.7 days at 15°C to 10.1 days at 35°C and the differences found were to be statistically significant at all temperatures studied.

Table 1 Mean duration of immature stages of ***Phyllocnistis citrella*** at different temperatures (days) (mean ± SE)

Temperature	Duration of immature stages (days)							
(°C)	n	Egg	n	Larva	n	Pupa	n	Total
15	80	7.3±0.20 a* (6-12)**	62	21 0±5 62 a (14-33)	62	23.9±0.53 a (11-30)	62	51 7±0 79 a (46-60)
20	72	5.4±0 12 b (4-7)	62	7 2±1 56 b (6-9)	58	14 0±0 16 b (12-17)	58	28 1±0 21 b (22-30)
25	90	3 0±0 09 c (2-4)	84	5.3±1 55 c (3-6)	82	7 7±0 09 c (7-9)	82	15 7±0 30 c (14-16)
30	105	2.8±0.04 cd (2-3)	102	4 8±1 04 cd (3-6)	98	4 6±0 10 e (3-7)	98	12.2±0 07 e (11-13)
35	137	1.9±0 04 e (1-7)	129	4 1±0 75 d (4-7)	125	4 0±0 07 f (3-6)	125	10 1±0 08 f (10-13)
25-35	108	2.6±0 18 d (2-3)	104	4 6±1 38 cd (3-11)	100	6.3+0 20 d (4-8)	100	14 1±0 20 d (13-18)

* Means in columns followed by the same letter are not statistically different according to LSD test (p=0 05)

** Values in paranthesis are minimum and maximum values

A linear regression analysis was applied to the developmental stages within a 15-35°C temperature range. The developmental rate of ***P. citrella*** decreased linearly with increasing temperature ($y = 0.0041x-0.0429$, $R^2 = 0.9918$) (Figure 1). The regression analysis showed that the developmental threshold was estimated as 10.4°C.

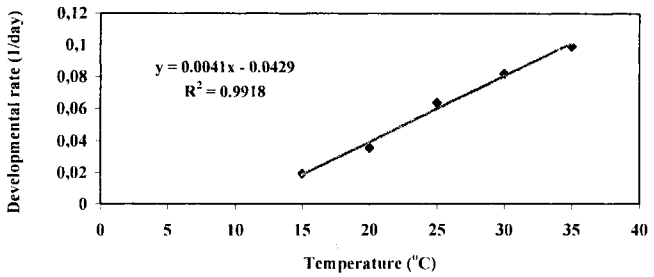

Figure 1 Rate of total development (egg to adult) of ***Phyllocnistis citrella*** Line represents the linear regression of development rate on temperature within the range of 15-35°C

The mortality rate of eggs ranged from 8.75 % at 15°C to 0.95 % at 30°C (Table 2). During the larval stages the mortality was higher than in the egg stage. No mortality was conducted at the pupal stage. Total mortality was high at 15°C (22.5 %) and at 20°C (19.44 %), but relatively low at the other temperatures.

Table 2 Mortality rate of egg and immature stages of ***Phyllocnistis citrella*** at different temperatures (%)

Temperature (°C)	Mortality of egg and immature stages (%)						Total mortality in number (Egg to adult) n		Mortality (%) (Egg to adult)
	n	Egg	n	Larva	n	Pupa			
15	80	8.75	73	15.18	62	-	80	18	22 50
20	72	6 94	67	13.42	58	-	72	14	19.44
25	90	3.33	87	5 67	82	-	90	8	8.88
30	105	0 95	104	5 07	98	-	105	7	6 67
35	137	1 46	135	7 44	125	-	137	12	8 75
25-35	108	1 85	106	5 60	100	-	108	8	7 40

Longevity and fecundity

The preoviposition and postoviposition periods were shorter than one day at temperatures above 20°C. The oviposition period shortened with increasing temperature (8.42 days at 20°C and 5.52 days at 35°C). Both females and males lived longer at 15°C (female, 14.5 days; male, 11.3 days) and shorter at 35°C (female; 5.6 days, male; 4.7 days) than the other temperatures studied. At all temperatures females lived longer than males. The highest oviposition rate was recorded at 30°C and at 25-35°C (average of 17.6 and 15.1 eggs per female per day, respectively), the lowest was at 20°C (7.2 eggs per female per day), whereas no eggs were deposited at 15°C. The overall fecundity was significantly lower at 20°C (29.6 eggs/female) than at the other temperatures tested and higher at 30°C and at 25-35°C (average of 57.1 and 54.8 eggs per female, respectively) which were found at the same statistical group (Table 3).

Table 3. Mean duration of preoviposition, oviposition, postoviposition, longevity and fecundity of ***Phyllocnistis citrella*** at different temperatures (mean ± SE)

Temperature (°C)	Duration of different periods (days)				Longevity (days)				No of eggs per female	
	n	Preovipositio	Oviposition	Postoviposition	n	Female	n	Male	per day	total
15	34		No eggs laid		34	14.5±0.21 a*	26	11.3±0.16 a	-	-
						(10-17)**		(8-13)		
20	31	1.1	8.42±1.48 a	1.4	31	11.2±0.14 b	25	9.4±0.12 b	7.2±1.12 c	29.58±6.57 c
		(1-2)	(5-12)	(0-4)		(7-14)		(6-11)	(0-9)	(21-47)
25	44	<1	6.28±1.72 b	<1	44	7.3±0.10 c	34	6.2±0.08 c	14.7±2.84 ab	51.81±15.91 ab
		(0-1)	(2-9)	(0-2)		(3-11)		(3-9)	(0-20)	(10-88)
30	56	<1	5.85±1.80 bc	<1	56	6.6±0.04 e	38	5.3±0.01 e	17.6±2.93 a	57.10±23.08 a
		(0-1)	(1-9)	(0-2)		(2-9)		(2-7)	(0-25)	(0-81)
35	65	<1	5.52±1.13 c	<1	65	5.6±0.07 f	54	4.7±0.02 f	14.4±2.43 ab	48.62±19.03 ab
		(0-1)	(2-7)	(0-2)		(1-7)		(1-6)	(0-22)	(0-74)
25-35	51	<1	6.08±1.41 b	<1	51	6.8±0.05 d	42	5.7±0.09 d	15.1±2.87 a	54.84±14.56 a
		(0-1)	(3-9)	(0-2)		(2-10)		(1-9)	(0-23)	(0-96)

* Means in columns followed by the same letter are not statistically different according to LSD test (p=0.05)
** Values in paranthesis are minimum and maximum values

The sex ratio of the offspring in each experiment was 1.3:1.0; 1.2:1.0; 1.3:1.0; 1.5:1.0; 1.2:1.0, and 1.2:1.0 females: males at 15, 20, 25, 30, 35 and 25-35°C, respectively.

According to the life tables, the mortality started at the developmental stages and was highest at 20°C. Survival rates of ***P. citrella*** adults decreased mainly after the oviposition period usually at older individuals in a short period at all temperatures (Figure 2). The oviposition period was 12 and 7 days at 20°C and 35°C and 9 days at 25, 30 and 25-35°C, respectively. Temperatures above 20°C resulted in shorter generation times (T) and increased net reproductive rates (R_o). The highest net reproductive rate was observed at 30°C with 50.19 females/female. The intrinsic rate of increase (r_m) showed significant differences at different temperatures being highest at 35°C with 0.260 females/female/day and 0.258 females/female/day at 30°C which were found at the same statistical group.

Discussion

Until 1986, the existent literature references cite ***P. citrella*** in many countries of Africa, Asia, Australia and Pacific Islands, remaining confined in these areas, however, an expansion occured in 1993 when ***P. citrella*** was detected in Florida (Heppner, 1993). Following the invasion in Florida, the studies concentrated primarily on the control of the pest. For an integrated management of a pest some biological and ecological aspects must be known.

There is limited information available in the literature on developmental time, mortality rate, longevity and fecundity of ***P. citrella*** at different temperatures under laboratory conditions. The egg, larval and pupal periods of ***P. citrella*** decreased with increasing temperatures. The total developmental time from egg to adult ranged from 51.7 days at 15°C to 10.1 days at 35°C and the differences were found to be statistically significant at all temperatures studied. Because varying temperatures reflect the conditions in the field better than the constant ones, the varying temperatures between 25 and 35°C resulted in a longer developmental time than in the averege temperature of 30°C. This seems to be mainly due to longer developmental time of the pupae; the other stages develop under this regime as fast as at a constant 30°C. Pandey and Pandey (1964) recorded an incubation period of 2-10 days, a larval period of 5-10 days, a pupal period of 6-20 days and a total life cycle of 13-52 days in the field during November- January in India with no indication of the temperature. Although these results describe studies in the field, they match well the results that we found in the laboratory. Wilson (1991), reported the egg, larval and pupal periods and the total life cycle as 2-6, 6-7, 6-7 and 14-18 days, respectively at 33°C. Wilson's results are similar to our results for 30 and 35°C. Ba-Angood (1977), found these periods as 2-6, 7-8, 8-9 and 18 days, respectively at the field with a changing temperature of 22-27°C. Radke and Kandalkar (1987) observed the egg, larval and pupal periods as 2.00 (1-3), 5.19 (5- 6)

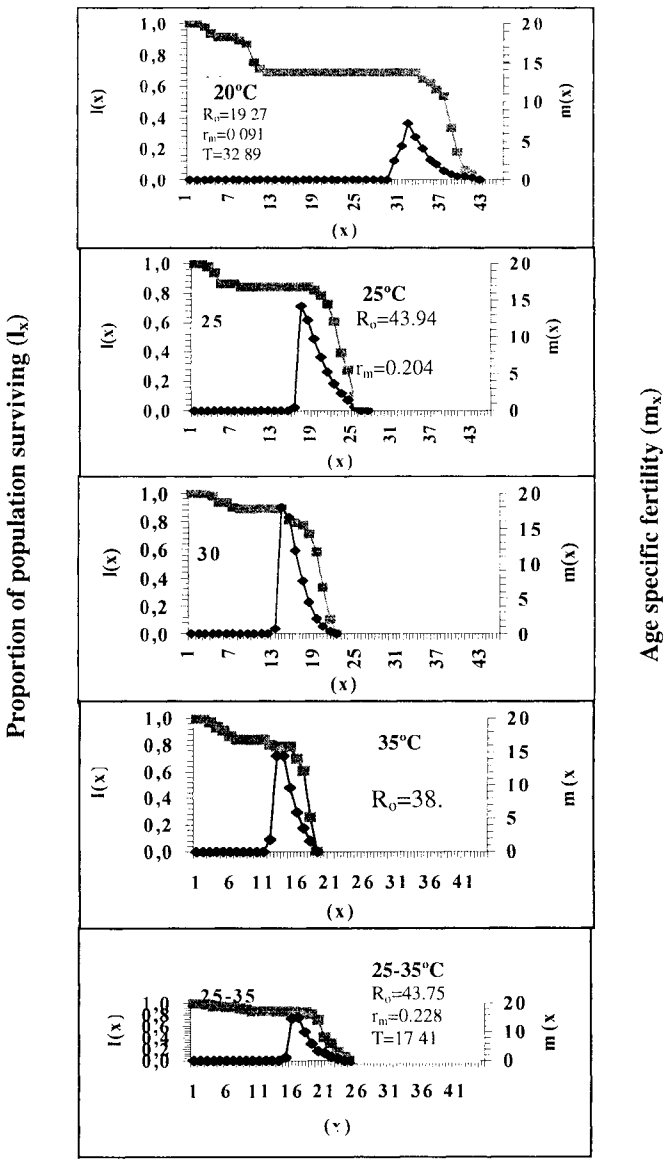

Figure 2 Survivorship curve (l_x) and age specific fecundity rate (m_x) of ***Phyllocnistis citrella*** at different temperatures [(x): Age (days), $l(_x)$: Proportion of population surviving, $m(_x)$: Age specific fertility]

and 6.24 (6-7) days, respectively. Huang et al. (1989) reported these periods as 0.5- 10.5, 3.0-49.5 and 3.5-17.0 days, whereas Beattie and Smith (1993) detailed 1-10, 5-6, 6-22 days, respectively. Although the temperatures are not given in these studies the differences might be attributed to that these studies were done at the field at different ecological conditions.

At all temperatures studied the mortality was low in the egg period and higher in the larval periods whereas no mortality was detected at the pupal period. Whether in the egg period (8.75 % at 15°C and 6.94 % at 20°C) or in the larval periods (15.2 % at 15°C and 13.4 % at 20°C), the mortality was higher at the lower temperatures. It was concluded that lower temperatures are suboptimal for these stages. At 30°C the mortality was at its minimum. The slightly higher mortality at 35°C may be the consequence of the faster development of the leaves which developed a hard leaf layer and became unsuitable for the larvae that could not feed on them and died. Some larvae developed as fast that they had consumed the leaf before it was fully developed and suffered from starvation. In both the field and laboratory observations it was concluded that death was also caused by not crossing the existing mines and not feeding when meeting with ones own or an other larvaes' mines. Siu-King and Ren-Guang (1980), Heppner (1993) and Stansly and Rouse (1993) also state that larvae generally do not cross existing mines and cannot feed when they meet their own or other larvaes' mines. This results in death was probably due to the protection of the pupae by the pupal cell.

Until now, the mortality of ***P. citrella*** has not been studied under laboratory conditions, but there are some field studies. Ba-Angood (1977) stated that in the field the mortality was higher when the temperature was high. Radke and Kandalkar (1987) stated that 19 % of the eggs laid by the pest in the field died. Wilson (1991) recorded that only 5.2 % of the larvae developed into pupa. The total mortality during development amounted to 96 %. Mari et al. (1996) found a 60-80 % mortality of the larval instars. These figures are much higher than the values we found in the laboratory. The direct impact of temperature seems to be not a major mortality factor, and so other factors must be responsible for the high mortality in the field.

Our results demonstrated that the preoviposition and postoviposition periods were shorter than one day at temperatures above 20°C. Ba-Angood (1977) stated that copulation takes place 9 to 11 hours after emergence and generally at night which matches well with our results. The author also noted that the unmated males and females lived as long as 6 days, while those mated had much shorter life which varied from 24 to 36 hours. In our study at 30°C, 35°C and 25-35°C it was observed that some of the mated females lived 1-2 days post copulation or post emergence. The oviposition period shortened with increasing temperature (8.42 days at 20°C and 5.52 days at 35°C). Both females and males lived longer at 15°C (female; 14.5 days, male; 11.3 days) and shorter at 35°C (female; 5.6 days, male;

4.7 days) than the other temperatures studied. At all temperatures the females lived longer than the males. Radke and Kandalkar (1987) found a longevity of 2.37 and 3.75 days for males and females, respectively and 24.87 (21-28) eggs per female. The longevity of both sexes and the number of eggs are lower in Radke and Kandalkar's study than in the present one. Huang et al. (1989) reported without a temperature a longevity of 1.0- 22.5 days and 1.0-7.5 days for females and males, respectively and 7- 108 eggs per female. Beattie and Smith (1993) observed that a female could live 5-10 days and lays 20 eggs per night and more than 50 eggs per its life which matches, more or less, similar to our results with the constant temperatures of 25°C, 30°C and varying temperature of 25-35°C. Heppner (1993) and Knapp et al., (1993) stated that adults live for a few days. Knapp (1995) observed an adult longevity of 2-12 days which could increase to 20 days and a fecundity of 48 (36-76) eggs during its life which is close to the rate observed in our data.

At all temperatures the females predominated over the males in number and the highest ratio was at 30°C with 1.5:1.0 (female:male). Huang et al. (1989) stated a female: male ratio of 1.1:1.0. Ba-Angood (1977) observed that the females slightly surpassed the males in number which is similar to our results for all temperatures studied.

The results of the life tables showed that ***P. citrella*** at 30°C displayed the highest net reproduction rate (R_o =50.19 females/female) and highest intrinsic rate of increase (r_m= 0.258 females/female/day), although at 35°C, it was marginally but statistically insignificant higher (r_m=0.260 females/female/day) than those of 30°C. At 30°C the female: male ratio was highest and mortality was lowest compared to the other temperatures studied. It is concluded that 30°C is the optimal temperature for the pest to grew.

Özet

Sıcaklığın Turunçgil yaprak galerigüvesi, *Phyllocnistis citrella* Stainton (Lepidoptera: Gracillaridae)'nın gelişme ve üremesine etkisi

Beş sabit (15, 20, 25, 30, 35°C) ve bir değişken (25-35°C) sıcaklığın Turunçgil yaprak galerigüvesi, ***Phyllocnistis citrella*** Stainton (Lepidoptera: Gracillaridae)'nın gelişme ve üremesi üzerine etkileri araştırılmış ve bu sıcaklıklarda zararlının yaşam çizelgeleri oluşturulmuştur. Ergin öncesi toplam gelişme süresinin sıcaklık arttıkça kısaldığı, en uzun 15°C'de 51.7 gün en kısa da 35°C'de 10.1 gün olduğu belirlenmiştir. En fazla ölüm 15°C'de olmuş ve bu sıcaklıkta yumurtlamanın olmadığı saptanmıştır. En az ölüm ise 30°C'de olmuştur. Çalışılan bütün sıcaklıklarda dişi bireyler erkek bireylerden istatistiki olarak daha uzun yaşamışlardır. Net üreme gücü (R_o), en yüksek 50 19 dişi/dişi ile 30°C'deki bireylerde saptanmıştır. Kalıtsal üreme kapasitesi (r_m) 30 ve 35°C'lerde sırasıyla 0.258 dişi/dişi/gün ve 0.260 dişi/dişi/gün olarak belirlenmiş ancak aynı istatistiki grupta yer almışlardır. Elde edilen sonuçlara göre ***P. citrella*** için optimum sıcaklığın 30°C olduğu kanaatine varılmıştır.

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