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Fertility Life Table of *Leucoptera coffeella* (Guérin-Mèneville) (Lepidoptera: Lyonetiidae) at Seven Temperatures in Coffee

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Abstract: Coffee leaf miner *Leucoptera* coffeella (Guérin-Mèneville, 1842) (Lepidoptera: Lyonetiidae) it is a one biggest insect pest of coffee in American continent coffee plantations, due to its generalized occurrence, since all coffee varieties are susceptible to attack, and the economic damage caused, which can involve production losses up to 50%. The present study investigated biological aspects, including the fertility life table and the ideal conditions for reproduction and development of this pest, at 7 constant temperatures (18, 22, 25, 28, 30, 32, 35°C) under laboratory conditions. The results showed that the mean biological development time of *L. coffeella* was affected by temperature, completing its life cycle more rapidly in increased temperatures until the 32°C, but low survivor of all *L. coffeella* stages was observed from 30°C. The preoviposition period decreased with increasing temperatures, the total fecundity was significantly affected by the temperature and the longevities of males and females were affected by the temperature. The highest net reproductive rate (22.23), finite rate of increase (0.15) and viability were observed at 28°C, indicating that this temperature was the most suitable for development. The information obtained can be used in integrated pest management programs, to forecast *L. coffeella* outbreaks and population growth, and to study the behavior of this insect under different climate conditions

Keywords: Coffee Leaf Miner, Biology, Coffee Var. Obatã

1. Introduction

In Brazil, *Leucoptera coffeella* (Lepidoptera: Lyonetiidae) is a major insect pest of coffee [1, 2]. It occurs over a wide area of the country and attacks all coffee varieties [3]. The pest causes significant economic damage [4], with production losses up to 50% [5].

The biology of this insect pest has been studied in several different coffee varieties [4, 6]. Egg-adult development varies with the coffee variety, temperature conditions, relative humidity, and rainfall [1, 4]. Temperature is the most important abiotic factor, affecting fecundity, reproduction, development time, emergence rate, and longevity [7, 8]. Climate changes may potentially affect many aspects of insect life cycles and ecology, especially those controlled by variables related to the availability of energy, e.g., degree-days. Birth and mortality rates of an insect population are determined by different biotic and abiotic factors [9].

The fertility life table is an appropriate method to study arthropod population dynamics because it allows estimation of parameters related to the growth potential of these populations [10]. Parra *et al.* [11] studied the fertility life table of *L. coffeella* at three constant temperatures (20, 27 and 30°C) in the *Mundo Novo* coffee variety and defined 27°C as the optimal temperature for maximum population growth rate.

The present study evaluated the effect of seven constant temperatures on the development of *L. coffeella* on leaves of the *Obatã* coffee variety, determining the fertility life table and optimal conditions for reproduction and development of this pest. This study provided new information on biological parameters of the coffee leaf miner on this coffee variety, over a wide range of temperatures.

2. Materials and Methods

Coffee leaves of the *Obatã* variety with larvae and pupae of coffee leaf miners were collected in the experimental area at the Department of Entomology and Acarology, ESALQ-USP, Piracicaba (22°42'51.0366"S, 47°37'41.556” W, 548 m), São Paulo state, Brazil.

For insect rearing, a method developed by Parra [4] was used. Infested leaves were taken to the laboratory and maintained in conical cages until adult emergence. Emerging adults were separated by sex based on external characteristics of the last abdominal segment. The adults were placed in rearing cages (40 × 40 × 50 cm) covered with a voile cloth. Coffee leaves on the third and fourth internodes (higher photosynthetic activity) were used as the oviposition substrate. The leaves were maintained in floral foam moistened with deionized water and covered with voile. Leaves were added, eggs counted, and dead insects removed daily. Pairs were maintained in a climate-controlled chamber at 25±2°C, 65±10% RH and a photoperiod of 14:10 h [L:D].

2.1. Biology of L. coffeella in Different Temperatures

The biology of *L. coffeella* was studied in climate-controlled chambers regulated at temperatures of 18, 22, 25, 28, 30, 32 and 35 ±1°C, 65±10% RH and photoperiod 14:10 h [L:D].

The study was initiated with a colony composed of 2100 eggs, divided into 7 treatments with 6 replicates each. Each replicate per treatment consisted of 50 eggs. Leaves with eggs were maintained in conical cages on the bottom with moistened filter paper. After the larvae hatch, they damage the upper epidermis of the leaf and burrow into the mesophyll, feeding on parenchyma cells. The larvae were maintained in conical cages during all larval and pupal stages until adult emergence. Developmental metrics included the duration and viability of the egg-adult period, duration of the preoviposition and oviposition periods, fecundity, and adult longevity. After adult emergence, 30 pairs of moths from each temperature were placed in a single-pair mating system at the same temperature. As described by Parra [4], the pairs were maintained in a conical cage (13.5 × 3 × 4.5 cm) (height × radius of upper base × radius of lower base) with coffee leaves of the *Obatã* variety, closed with voile. To feed the adults, a braided cotton roll soaked in 10% sucrose solution was placed on the voile [4]. Coffee leaves were changed and the number of eggs was counted daily, using a stereomicroscope (10×). The mortality of males and females was also determined. The experiment was concluded when all adults died.

Data from all tests were submitted to a Shapiro-Wilk test (P<0.05) to verify the normality, as well as a Bartlett's test (P<0.05) to verify the homoscedasticity. The data for the egg-adult period, viability and number of eggs per female were submitted to the analysis of variance by the GLM procedure and the means were compared by Tukey test at 5% significance [12]. The values for sex ratio, sr = (number of females)/(number of females + number of males), were analyzed by the chi-square test (P < 0.05) in R [12].

2.2. Fertility Life Table of L. coffeella at Different Temperatures

In order to obtain the fertility life tables at each temperature, the number of eggs per female (m_x) on each day (x) and the proportion of the number of females surviving to the next day were calculated. Then, the fertility life table was constructed according to Maia *et al.* [10]. The following parameters were estimated: generation time (T) (eq. 1), net reproductive rate (R_0) (eq. 2), intrinsic rate of increase (r_m) (eq. 3) and finite rate of increase ($ʎ$) (eq. 4).

$$Ro = \Sigma\, l_x . m_x \quad (1)$$

$$r_m = \frac{\ln(Ro)}{T} \quad (2)$$

$$T = \frac{\ln(\mathrm{Ro})}{\mathrm{r}_m} \quad (3)$$

$$ʎ = \mathrm{e}^{\mathrm{r}_m} \quad (4)$$

The bootstrap technique was used to calculate the variance of the estimated parameters, according to Meyer *et al.* [13], using R version 3.2.3 [12].

3. Results and Discussion

3.1. Biology of Leucoptera coffeella at Different Temperatures

Temperature significantly influenced the incubation period of eggs, which varied from 3.1 days at 35°C to 10.3 days at 18°C ($F_{6,33} = 168.11, P < 0.001$) and stabilized at temperatures higher than 25°C (Figure 1). Although the incubation periods were progressively shorter at higher temperatures, egg viability was significantly reduced only at 35°C (Table 1), as also observed by Parra [4]. Authors such as Notley [14] and Magalhães *et al.* [15] have noted that differences in the viability of this stage may be associated with other environmental factors besides the temperature, e.g., with genotypes of *Coffea arabic*a.

The mean duration of larval development was 7.1 days at 32°C to 21.6 days at 18°C ($F_{5,30} = 178.3, P < 0.001$) and stabilized at temperatures higher than 25°C, as also found by Parra [4] (Figure 1). Larvae failed to complete their development at 35°C. The mean durations of larval development were longer than those reported by Parra [4], probably because that study used a different coffee variety (*Mundo Novo*). According to Magalhães *et al.* [15], the development time of the coffee leaf miner varies because of the chemical compounds in different varieties of Brazilian coffee. At temperatures above 28°C, larval viability decreased (Table 1), as also observed by Parra [4]; Katiyar and Ferrer [16].

Temperature significantly influenced the pupal development ($F_{5,30} = 109.91, P < 0.001$), which varied from 3.5 days at 32°C to 14.7 days at 18°C (Table 1). Pupal development was not observed at 35°C. As in the other stages, the higher the temperature, the shorter the development time, with a more stable behavior at higher temperatures (Figure 1). Pupal development times were similar to those reported by Parra [4] and Katiyar and Ferrer [16]. Pupal viability was highest at 25°C, decreasing above and below this temperature.

At temperatures of 30°C and above, pupal malformations were observed; only 80% of the individuals at 30°C and 66% at 32°C showed the "X" shape of normal pupae. Individuals that failed to produce a cocoon (20% at 30°C and 34% at 32°C) dried out in the high temperatures, since the cocoons provide a proteinaceous shelter produced from the labial glands, to protect the insects from environmental stress [17].

The development time of the egg-adult period was significantly affected by the temperature between 18°C and 32°C ($F_{5,30}$ =7224; P<0.001) (Table 1), ranging from 11.53 days at 32°C to 46.62 days at 18°C. Viability did not differ significantly from 18°C to 28°C, ranging from 50% at 32°C to 70% at 18°C, with a peak at 25°C (89%), similar to the variation found by Notley [14] in coffee leaf miners on Mount Kilimanjaro in Africa.

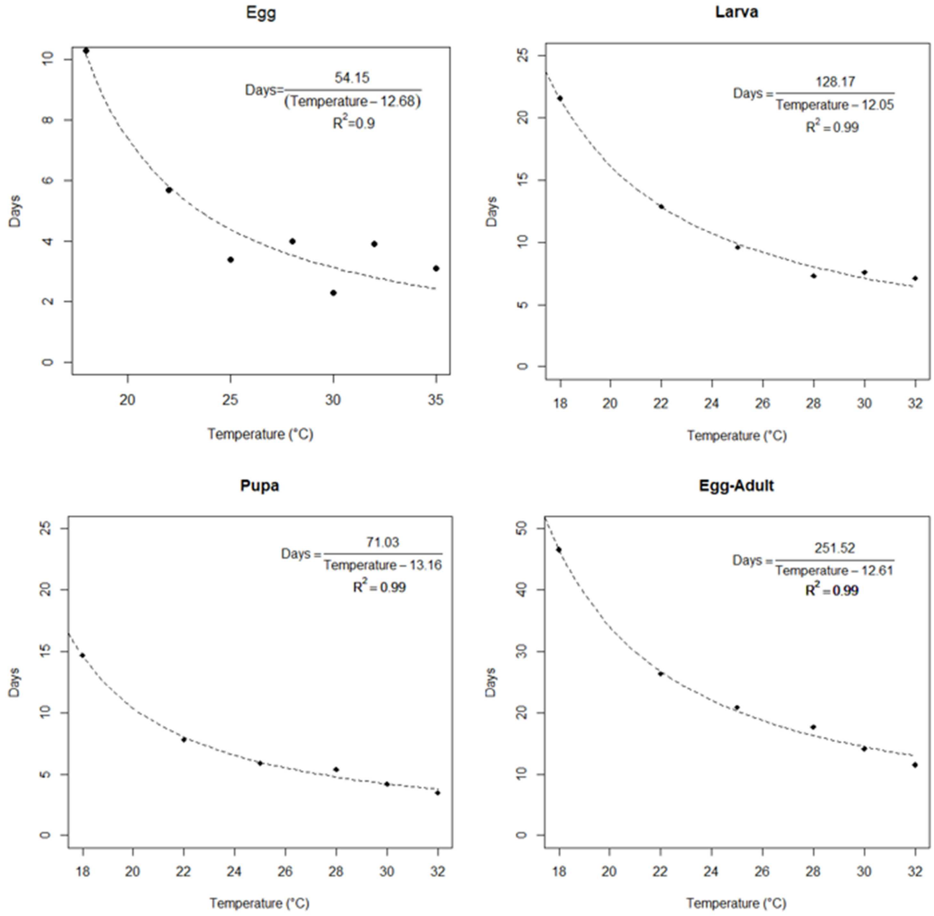


Figure 1. *Development of Leucoptera coffeella at seven constant temperatures on coffee leaves of the Obatã variety. RH 65±10% and photoperiod of 14:10 h (L:D).*

In the present study, although the viability of the egg-adult period decreased as the temperature moved away from 25°C, enough individuals survived at all temperatures (except 35°C) to allow construction of a fertility life table. The temperature did not significantly influence the sex ratio (Table 1) (X^2 = 0.98, p = 0.5718), which varied from 0.55 to 0.57, as found by Parra [4]. The preoviposition period decreased with increasing temperatures ($F_{5,28}$ =115.3; P<0.001) (Table 2). Females of *L. coffeella* initiated oviposition 12.4 days after adult emergence at 18°C, but only 1.4 days after emergence at 32°C. Total fecundity (mean number of eggs laid per female) was significantly affected by the temperature ($F_{5,28}$ =74.5; P<0.001), and was highest at 25 and 28°C, with 64.9 and 60.3 eggs per female, respectively. The negative relationship between oviposition and temperature (Table 2) was similar to the findings of Notley [14] for populations in Africa. On the Obatã coffee variety, coffee leaf miners laid the most eggs at 25 and 28°C.

Table 1. *Mean duration and viability of the egg, larval, pupal and egg-adult periods, and sex ratio (±SE[a]) of Leucoptera coffeella, at seven constant temperatures on coffee leaves of the Obatã variety. RH 65±10% and photoperiod of 14:10 (L:D).*

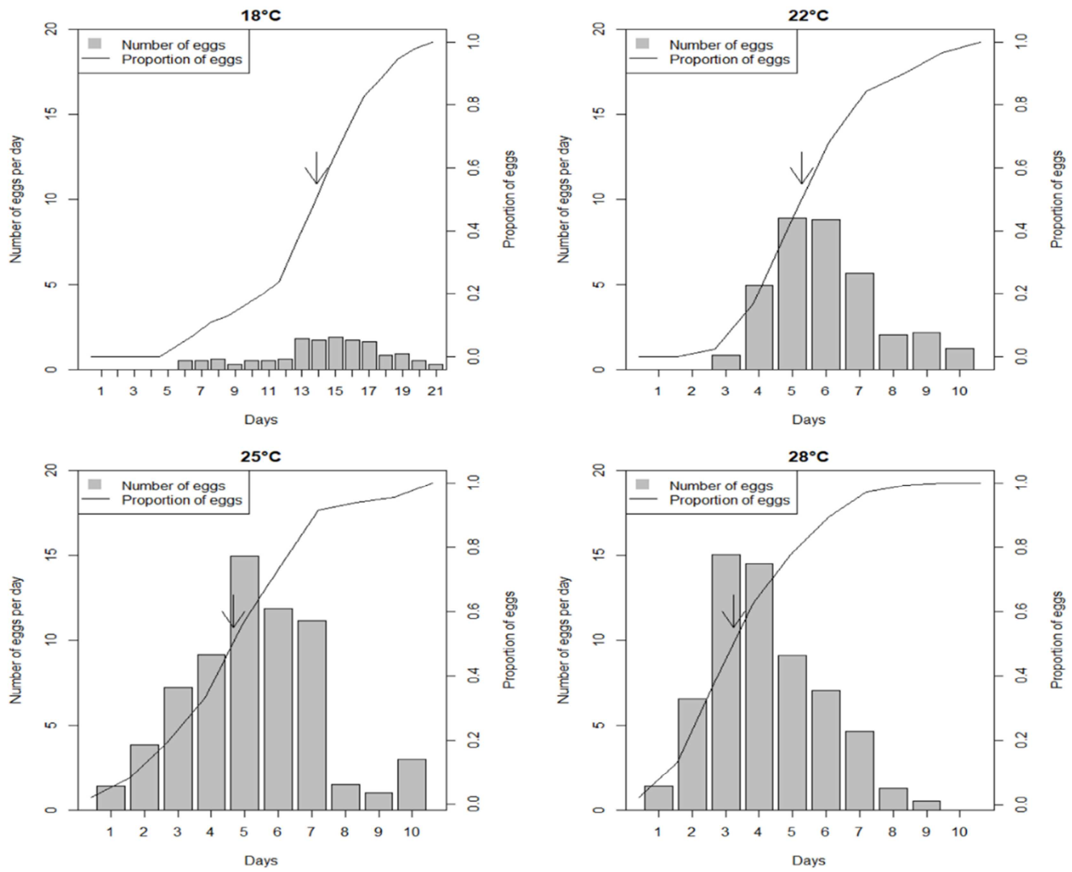
Temperature (°C)	**Duration (days) ***							
	n	**Egg**	**N**	**Larva**	**n**	**Pupa**	**n**	**Egg-adult**
18	300	10.3±0.2a	272	21.6±0.8a	246	14.7±0.3a	209	46.6±1.3 a
22	300	5.7±0.0b	280	12.9±0.2b	261	7.8±0.1b	236	26.4±0.3b
25	300	3.4±0.2c	291	9.6±0.3bc	263	5.9±0.2c	248	20.9± 0.7c
28	300	4.0±0.1c	282	7.3±0.2c	262	5.4±0.4c	229	17.7±0.7c
30	300	2.3±0.1c	265	7.6±0.4c	214	4.2±0.3cd	208	14.2±0.2cd
32	300	3.9±0.3c	257	7.1±0.6c	208	3.5±0.3d	167	11.5±0.1d
35	300	3.1±0.4c	105	-	-	-	-	

Table 1. *Continued.*

Temperature (°C)	**Viability (%)**				**Sex ratio****
	Egg	**Larva**	**Pupa**	**Egg-adult**	
18	91±1.6a	88±3.6a	85±3.0a	70±5.9a	0.56a
22	93±3.9ab	90±5.6ab	90±4.6ab	75±4.7ab	0.57a
25	97±2.7b	97±2.6b	95±3.4ab	89±7.1b	0.56a
28	94±1.8b	93±3.8b	87±4.4a	76±6.9ab	0.57a
30	88±2.3a	81±5.8c	78±6.7c	56±6.0c	0.57a
32	86±3.4a	81±6.8c	73±10.6c	50±8.9c	0.57a
35	35±8.9c	-	-	-	-

Means followed by the same letter in a column do not differ from one another (Tukey, $p<0.05$* and $X^2<0.05$**). N = number of individuals used in each treatment. [a] Standard error.

The number of eggs per day varied according to the temperature. At the lowest temperature (18°C), the highest number of eggs were laid between days 13 and 17, whereas the oviposition peak was observed at 22 and 25°C on day 5 and at 28, 30 and 32°C on day 3 (Figure. 2). Fifty percent of the total eggs were laid in the first six days (Figure 2), except at 18°C, when 50% were laid by day 14.



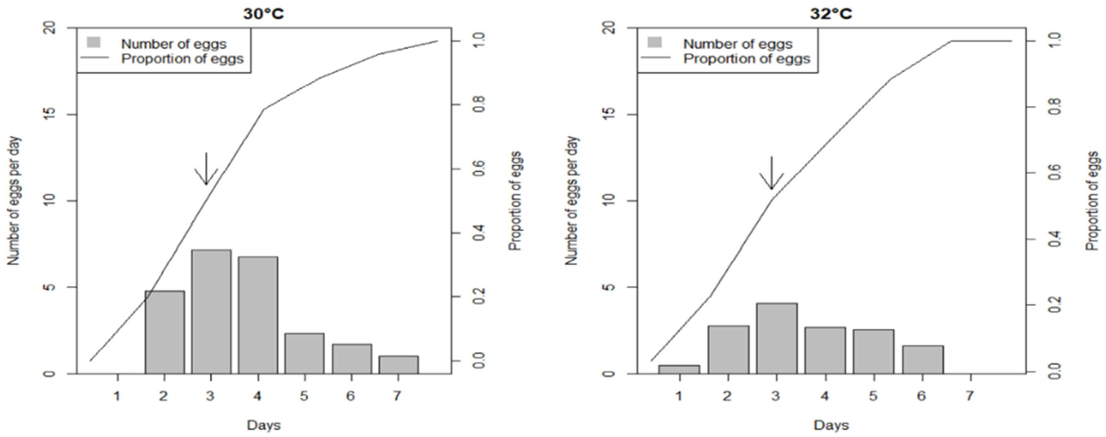


Figure 2. Mean number of eggs/female. day^{-1} (±SE) (histogram) and cumulative proportion of eggs laid by females of Leucoptera coffeella at six constant temperatures on coffee leaves of the Obatã variety. RH 65±10% and photoperiod of 14:10 h [L:D]. Arrow indicates when the female laid 50% of the total eggs.

Table 2. Preoviposition period and fecundity of Leucoptera coffeella at six constant temperatures on coffee leaves of the Obatã variety. RH 65±10% and photoperiod of 14:10 h [L:D].

Parameter	Temperature°C					
	18	22	25	28	30	32
Pre-oviposition period (days)	12.4±0.6a	7.2±0.5b	3.4±0.2c	2.9±0.3cd	1.6±0.2d	1.4±0.1d
Mean number of eggs per female	10.2±1.6a	34±4.2b	64.9±3.6c	60.3±3.8c	23±2.1b	14.5±1.4a

Means followed by the same letter are not significantly different (Tukey, p<0.05)

3.2. Adult Longevity

The longevities of males ($F_{5,98} = 99.32$; $P < 0.001$) and females ($F_{5,98} = 102.9$; $P < 0.001$) were significantly affected by the temperature. For males, longevity varied between 3.7 and 17.9 days, and for females, between 4.3 and 21.5 days, at 32°C and 18°C respectively (Table 3). This pattern resulted from the higher metabolism associated with an increase in temperature.

Table 3. Male and female longevities (±SE) of Leucoptera coffeella at six constant temperatures on coffee leaves of the Obatã variety. RH 65±10% and photoperiod of 14:10 h [L:D].

Temperature (°C)	n	Period (days) Mean ± SE	
		Males	Females
18	100	17.9±1.3a	21.5±1.3a
22	100	11.5±0.5b	14±0.7b
25	100	6.2± 0.4c	7.6±0.5c
28	100	6±0.2c	8.6±0.4c
30	100	3.9±0.2cd	6.3±0.3cd
32	100	3.7±0.2d	4.3±0.3de

Means followed by the same letter in a column are not significantly different (Tukey, p<0.05). n = number of individuals used in each treatment

3.3. Fertility Life Table of Leucoptera Coffeella at Six Constant Temperatures

The highest population growth rate was observed at 28°C, when the net reproductive rate (R_0) was equal to 22.23 (14.7–32.3) (Table 4). At 35°C, it was not possible to construct a fertility life table since adults were not obtained at this temperature. The R_0 is defined as the average number of females that a female individual in a population will produce in its oviposition period, and is related to increases, decreases or stability in a population in each generation (18; 9). Parra *et al*. [11] found the highest value of R_0 at 27°C (29.82), higher than the value obtained in the present study. The lowest values of R_0 were obtained at the extreme temperatures (18°C and 32°C), indicating that these temperatures are not suitable for the insect's reproduction.

The intrinsic rate of increase (r_m) was positive in all temperatures, indicating that the population of coffee leaf miners increases in the temperature range 18–32°C, varying from 0.03 (0.00–0.05) at 18°C to 0.15 (0.09–0.19) at 28°C (the highest r_m observed) (Table 4). In Brazil, the largest infestations have been observed in dry periods, which usually extend from April to October. During rainy periods, populations are smaller [19].

The value of r_m reflects the rate at which a population increases in size [9]. At the extreme temperatures (18 and 32°C), the survival of immature stages and the fecundity both decreased. At 18°C, the observed value can be associated with the long development times of the immature stages (Table 1) and pre-oviposition periods. At 32°C, although the development time was shorter than at the other temperatures, the viability and fecundity were low, affecting the intrinsic rate of increase in the life table (Table 1).

The generation time (T) corresponded with the biological data, since an inverse relationship between the development time and the temperature was also observed in the 18 to 30°C range (Table 4).

The values obtained for the finite rate of increase (λ) ranged between 1.03 (0.99–1.05) at 18°C and 1.16 (1.09–1.21) at 28°C (Table 4). This parameter indicates the growth potential of an insect population and is therefore frequently used in insect control programs [20]. Based on the data for this parameter, populations of *L. coffeella* can potentially increase at all the temperatures (18–32°C) studied here.

Table 4. *Mean values of the parameters (95% CI) of the fertility life table of Leucoptera coffeella, at six constant temperatures on coffee leaves of the Obatã variety. RH 65±10% and photoperiod of 14:10 h [L:D].*

Parameter	**Temperature°C**					
	18	**22**	**25**	**28**	**30**	**32**
R_0	5.12 a (0.75–9.76)	14.26 b (11.28–25.6)	18.91 b (15.29–22.78)	22.23 b (14.76–32.3)	7.08 ab (0.9–15.23)	4.09 a (0.77–7.78)
r_m	0.03 a (0.00–0.05)	0.08 b (0.04–0.11)	0.11 b (0.06–0.14)	0.15 b (0.09– 0.19)	0.14 ab (0.00–0.19)	0.10 ab (0.00–0.15)
T	48.80 a (47.45–50.51)	31.15 b (29.55–32.69)	23 c (22.33–25.68)	20.54 c (22.48–48.87)	13.86 d (12.5–15.67)	13.79 d (12.40–15.37)
Λ	1.03 a (0.99–1.05)	1.09 a (1.04–1.12)	1.13 a (1.07–1.15)	1.16 a (1.09–1.21)	1.15 a (1.00–1.21)	1.10 a (0.98–1.16)

R_0 = net reproductive rate, r_m = intrinsic rate of increase, T = generation time, λ = finite rate of increase. Means followed by the same letter in a line do not differ significantly (95% CI). Parameters compared by bootstrap analysis (10,000 repetitions).

4. Conclusion

The results of this study confirmed the effect of temperature over the range of 18 to 32°C on the biology, reproductive and population parameters of *L. coffeella*. In coffee leaf miner increasing temperatures reduces preoviposition period and eclosion time of the laid eggs, key biological factors to consider when new pest populations are arriving to new crop systems.

In the same line, *L. coffeella* showed an optimal developmental and biological fitness at temperature of 28°C with fecundity, time of 50% of emerged eggs, growth population rate, intrinsic rate of increase and finite rate of increase expressing the highest values among the evaluated temperatures. Special attention is suggested on field crops grown at this temperature, because the insect pest will have a high potential to affect the coffee crop with high intensity.

Whole the information obtained is fundamental for growers, researchers, epidemiologists and agronomist to develop trust and precise monitoring and prediction schemes as well as for designing integrated pest management programs for *L. coffeella,* based on the growth population to a regional scale.

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