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Temperature Effects on *Trichogramma pretiosum* Riley and *Trichogrammatoidea annulata* De Santis

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ABSTRACT
*The influence of temperature on lifetime attributes of Trichogramma pretiosum Riley and Trichogrammatoidea annulata De Santis (Hymenoptera: Trichogrammatidae) was evaluated at four constant temperatures (15, 20, 25, and 30°C), RH 70 ± 10%, photophase 14 h. Anagasta kuehniella (Lepidoptera: Pyralidae) eggs were used as hosts. Developmental times of both parasitoid species were similar when exposed to 20, 25, or 30°C. T. annulata, however, developed slightly faster than **T.** pretiosum at 15°C. Emergence rates of both species were above 89%. The temperature threshold for **T.** pretiosum and T. annulata was 11°C and the number of degree-days required for their development was 126.9 and 122.3, respectively. Parasitization was maximal at 25°C. T. annulata, however, parasitized significantly more hosts than T. pretiosum in the entire temperature range. Temperature had no effect in brood size. T. annulata progeny consisted predominantly of males, except at 15°C, whereas in T. pretiosum it consisted predominantly of females, except at 30°C. Parental females lived longer than males.*

Key words: Insecta, biological control, avocado fruit borer, temperature

INTRODUCTION

The avocado fruit borer, *Stenoma catenifer* (Wals.) (Lepidoptera: Elachistidae) has become a key pest on avocado, *Persea americana* (L.) in Brazil in the last decades. When infestations are high, the yield are low, which in pesticide free groves may reach 100% (Hohmann and Meneguim, 1993). As control measures have relied mainly on highly toxic insecticides, studies were initiated to evaluate other methods to reduce pest damage on avocados (Hohmann et al., 2000; Hohmann et al., 2001). The studies also included the identification of parasitoids in the family Trichogrammatidae. This family comprises the most studied and used egg parasitoids of lepidopterans worldwide (Hassan, 1988; Li, 1994). Two species were reared from *S. catenifer* eggs: *Trichogramma pretiosum* Riley and *Trichogrammatoidea annulata* De Santis (Hymenoptera: Trichogrammatidae) (Hohmann and Meneguim, 1993; Hohmann et al., 2001). Their broad distribution and parasitization rates in some areas suggested that these species could be considered as a tool to reduce *S. catenifer* damage on avocados. However, before any attempt is made to mass rear and release these egg parasitoids, the factors that may affect their distribution and abundance must be understood. In this regard one important factor to be considered is temperature (Andrewartha and Birch, 1954). The knowledge of the thermal requirements and the effects of temperature are among many attributes that influences the outcome of biological control projects (Butler and Lopez,

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1980, Chihrane et al., 1993; Bernal, 1995). Therefore, the aim of this study was to estimate the developmental times and the thermal requirements, and evaluate temperature effects on prasitism capacity, sex ratio, and longevity of *T. pretiosum* and *T. annulata.*

MATERIAL AND METHODS

Parasitoid's Culture Origin and Maintenance

T. pretiosum and *T. annulata* cultures were initiated with parasitized *S. catenifer* eggs collected on avocado by C. L. Hohmann, in Arapongas (Lat. 23° 25' 00' S, Longit. 51° 26" 00' W - GR) PR, Brazil, in 1990. At emergence, each parasitoid species was exposed to cards (9 X 10 cm) containing a large supply of sterilized (UV light) *A. kuehniella* eggs inside of polyethylene containers (10 X 13 cm). The host has been mass-reared in the Laboratory of Entomology of the Instituto Agronômico do Paraná – IAPAR, in Londrina, PR, Brazil for more than five years. Parasitoid cultures were initiated at different days to provide wasps whenever they were needed to start the experiments. The host as well as the parasitoid cultures were maintained at 25 ± 2°C, RH 70 ± 20%, and photophase of 14 h.

Experimental Conditions

The development times and the effect of temperature on parasitism capacity and longevity were measured at 15, 20, 25, and 30°C inside temperature cabinets (Fanem, Model 347). Relative humidity and photoperiod were 70 ± 10% and 14L: 10D, respectively. To determine developmental times and thermal constants, fifteen females of each species (< 12 h old) were individually placed in rearing units (Hohmann et al., 1988) containing 100 ± 10 *A. kuehniella* eggs (< 24 h old) during 24 h. The females were then removed and the vials placed inside of the respective environment chamber, and held for parasitoid development. The number and sex of the wasps were recorded daily during their period of emergence.
To determine the effect of temperature on parasitism capacity and longevity the same protocol described above was followed, except that the host eggs were replaced daily until all females died. Honey was provided as food source. The number of parasitized eggs, emergence, brood size and parental female's longevity were recorded.

Statistical Analysis

The variables developmental times, parasitoid's emergence, parasitization rates, longevity, and sex ratio between treatment groups were compared using a one-way analysis of variance (ANOVA) (SAS Institute Inc. 1994) followed by Duncan's Multiple Range Test at 0.05 level. Data on emergence, and sex ratio were transformed (log x). Sex ratio was expressed as % of females [(no. of females/ no. of males + no. of females) x 100].
The development thresholds (*t*) and thermal constants (K) were estimated using the hyperbole method (Haddad and Parra, 1984).

RESULTS AND DISCUSSION

The developmental times of *T. pretiosum* and *T. annulata* were similar at 20, 25, and 30°C (Table 1). At 15°C, however, *T. annulata* developed slightly faster than *T. pretiosum* ($P<0.05$). The developmental times increased from 6.6 days at 30°C to 29.7 days at 15°C in *T. pretiosum*, and from 6.4 days at 30°C to 29.0 days at 15°C in *T. annulata*. This relationship has been consistent for various species of *Trichogramma* (Russo and Voegelé, 1982; Harrison et al., 1985; Bleicher and Parra, 1989; Cônsoli and Parra, 1995; Hohmann and Luck, 2000).
Emergence rates were high (> 89%) for both parasitoid species in the entire temperature range. Similar results have been reported for other species of *Trichogramma* (Harrison et al., 1985; Cônsoli and Parra, 1995).
The estimated temperature threshold of both species was ca. 11°C and the number of degree-days for *T. pretiosum* and *T. annulata* was 126.9 and 122.3, respectively (Table 2). The lower temperature thresholds were very similar to that reported for *T. pretiosum* reared on *Sitotroga cerealella* Olivier (Lepidoptera: Gelechiidae) eggs (Goodenough et al., 1983), and for *Trichoplusia ni* Hübner (Lepidoptera: Noctuidae) eggs (Butler and Lopez, 1980).

Table 1 - Developmental times (days) of *T. pretiosum* and *T. annulata* reared on *A. kuehniella* eggs at four constant temperatures. RH 70 ± 10% and 14L:10D.

Temperature (°C)	Parasitoid spp.	
	T. pretiosum	*T. annulata*
15	29.7A a	29.0 B a
20	14.7A b	14.3A b
25	9.0A c	8.5A c
30	6.6A d	6.4A d

Means followed by the same upper case letter in the row or by the same lower case letter in the column were not significantly different at 0.05 level (Duncan's Multiple Range Test).

Table 2 - Developmental threshold (*t*) and thermal constants (K) of *T. pretiosum* and *T. annulata* reared on *A. kuehniella* eggs. RH 70 ± 10% and 14L:10D.

Parasitoid spp	*t* (ºC)	K (degree-days)	R^2
T. pretiosum	11.2	126.9	0.99
T. annulata	11.3	122.3	0.99

Table 3 - Number of *A. kuehniella* eggs parasitized by *T. pretiosum* and *T. annulata* at four constant temperatures. RH 70 ± 10% and 14L:10D.

Temperature (°C)	Parasitoid spp.	
	T. pretiosum	*T. annulata*
15	43.3A a	70.4A b
20	79.2 B a	144.0 B b
25	122.2 CD a	210.2 C b
30	106.4 D a	154.4 B b

Means followed by the same upper case letter in the column or by the same lower case letter in the row were not significantly different.

Table 4 - Percent of females of *T. pretiosum* and *T. annulata* reared on *A. kuehniella* eggs at four constant temperatures. RH 70 ± 10% and 14L:10D.

Temperature (°C)	% of females	
	T. pretiosum	*T. annulata*
15	73AB a	61A b
20	82A a	33 B b
25	91 CD a	31 B b
30	52 B a	24 B b

Means followed by the same upper case letter in the column or by the same lower case letter in the row were not significantly different.

Trichograma pretiosum parasitized fewer eggs than *T. annulata* in all temperatures tested (Table 3) (P<0.05). Parasitization was maximal at 25°C, but no difference was detected between the number of eggs parasitized by *T. pretiosum* at 25 and 30°C. The number of eggs parasitized by *T. annulata* at 25°C differed significantly from that at 15, 20, and 30°C. These results are in accordance with studies carried out with other species of *Trichogramma* (Lund, 1938; Russo and Voegelé, 1982; Pak and Oatman, 1982; Harrison et al., 1985; Pak and van Heiningen, 1985).

Offspring sex ratio varied with parasitoid species and temperature (Table 4). In *T. pretiosum* the offspring was female-biased throughout the entire temperature range, being less abundant at 15°C (73%) and 30°C (52%) ($P<0.05$). Harrison et al. (1985) studying the temperature effects on *T. pretiosum* sex ratio reported that females were slightly less abundant at lower and upper thresholds for development. *T. annulata* offspring was mostly male-biased, except at the lowest temperature (15°C) ($P<0.05$). Similar results were revealed by Hutchison et al. (1990) in *Trichogrammatoidea bactrea* Nagaraja. According to the authors, the % of daughters ranged from 51.6% at 30°C to 77.1% at 15°C.

As temperature increased, longevity decreased accordingly (Table 5). *T. pretiosum* parental females lived significantly longer than *T. annulata* within the 20-30°C range, but no difference occurred at 15°C. Females lived significantly longer than their conspecific males at comparable temperatures, regardless of species, the differences being greater at the lower temperature range (Table 5).

Temperature effect on longevity was independent of the parasitoid species, indicating that the higher parasitism capacity of *T. annulata* was not a longevity artifact as it could be speculated, but rather, it resulted from its higher fecundity.

Table 5 - Longevity of *T. pretiosum* (*T. a*) and *T. annulata* (*T. p*) reared on *A. kuehniella* males and females at four constant temperatures. RH 70 ± 10% and 14L:10D.

Temperature (°C)	Males		Females	
	T. p	*T. a*	*T. p*	*T. a*
15	15.0A a	15.2A a	32.9A a	30.5A a
20	13.0A a	12.1A a	29.1A a	27.3 B a
25	7.6 B a	8.4 B a	21.3 B a	18.3 C a
30	3.1 C a	3.3 C a	13.4 C a	11.6 D a

Means followed by the same upper case letter in the column or by the same lower case letter in the row, among conspecifics, were not significantly different.

Longevity of *T. pretiosum* and *T. annulata* males and females changed with temperature. As temperature increased longevity decreased accordingly (Table 5). This result is in agreement with studies conducted with other *Trichogramma* species (Lund, 1938; Pak and Oatman, 1982; Harrison et al., 1985; Bleicher and Parra, 1989).

The present study revealed significant differences between *T. pretiosum* and *T. annulata* with respect to temperature requirements, and temperature effects on development and survival. *T. annulata* developed faster at 15°C and required less heat units to complete development than *T. pretiosum.* They also confirmed their higher parasitism rate compared to *T. pretiosum* when exposed to *A. kuehniella* eggs in all temperatures tested (Hohmann et al., 2001).

T. pretiosum lived longer than *T. annulata* in the 20 to 30°C temperature ranges. This result, however, might not have a practical relevance unless *Trichogramma* females are able to find food after emergence. In the absence of a carbohydrate source they would not be able to survive more than two days (Hohmann et al., 1988).

Despite the statistical significance of the differences found, it is not certain whether these differences would be biologically important to give either species an advantage or not. Therefore, the results above need to be confirmed with more detailed laboratory and field studies, using the natural host *S. catenifer*.

The outcome of these and other studies (Maceda et al., 1994; Hohmann et al., 2001), when added up to information on parasitoids' spatial and seasonal distribution and abundance, may provide the tools to understand the major factors that are affecting *T. pretiosum*and *T. annulata* reproductive success. Moreover, all this information will be crucial to determine their potential role as biological control agents of the *S. catenifer* on avocados.

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RESUMO

A influência da temperatura sobre parâmetros biológicos de *Trichogramma pretiosum* Riley e *Trichogrammatoidea annulata* De Santis (Hymenoptera: Trichogrammatidae) foi avaliada à 15, 20, 25 e 30°C. Foi usado como hospedeiro ovos de *Anagasta kuehniella* (Lepidoptera: Pyralidae). A progênie de ambos parasitóides apresentou um desenvolvimento similar quando submetidas a 20, 25 ou 30°C. *T. annulata*, contudo, apresentou um desenvolvimento ligeiramente mais rápido que *T. pretiosum* à 15°C. A porcentagem de emergência foi elevada para ambas espécies em todas as temperaturas, não havendo influência destas sobre o número de parasitóides emergindo por hospedeiro. O número de graus dia para desenvolvimento de *T. pretiosum* e *T. annulata* foi 126,9 e 122,3, respectivamente, e a temperatura base de desenvolvimento 11°C. A taxa de parasitismo aumentou com o aumento da temperatura, sendo máxima a 25°C. Fêmeas de *T. annulata*, entretanto, parasitaram um número significativamente maior de hospedeiros que *T. pretiosum* nas quatro temperaturas. A progênie de *T. annulata* foi constuída principalmente por machos, exceto a 15°C, enquanto que a de *T. pretiosum* por fêmeas, exceto a 30°C. A longevidade de ambas espécies de parasitóides diminuiu com o aumento da temperatura sendo que as fêmeas viveram por um periodo significativamente maior que os machos.

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