

## TEMPERATURE EFFECT ON THE BIOLOGY AND REPRODUCTIVE PERFORMANCE OF THE EGG PARASITOID *Trissolcus basalis* (WOLL.)

Beatriz S. Corrêa-Ferreira<sup>1</sup> e Flávio Moscardi<sup>1</sup>

### RESUMO

Efeito da Temperatura na Biologia do Parasitóide de Ovos *Trissolcus basalis* (Woll.)

O efeito de quatro temperaturas (18°, 22°, 26° e 30°C), no desenvolvimento e performance reprodutiva de *Trissolcus basalis* (Woll.) foi determinado usando ovos de *Nezara viridula* (L.) como hospedeiro. O tempo de desenvolvimento da oviposição à emergência dos adultos foi inversamente proporcional à temperatura, com períodos médios para machos e fêmeas variando de 8,7 e 10,2 dias à 23,6 e 26,7 dias, respectivamente. As percentagens de desenvolvimento, de emergência e de sobrevivência dos parasitóides foram elevadas (96,9% a 100%) em todas as temperaturas estudadas, não se constatando, entretanto, influência sobre estes parâmetros biológicos. A produção diária de descendentes por fêmea variou significativamente nas diferentes temperaturas, com a maior produção de descendentes/dia (34,8) obtida a 30°C. Em todas as temperaturas, o pico de produção ocorreu no primeiro e segundo dia de vida adulta, com um total máximo de 194,9 ovos por fêmea de *T. basalis*, obtido a 22°C. Entretanto, a produção total de ovos não diferiu significativamente nas diferentes temperaturas.

PALAVRAS-CHAVE: Insecta, controle biológico, parasitismo de ovos, *Nezara viridula*.

### ABSTRACT

The effect of four temperatures (18°, 22°, 26° and 30°C) on the development and reproductive performance of *Trissolcus basalis* (Woll.) was assessed using eggs of *Nezara viridula* (L.) as host. The developmental time from oviposition to adults emergence was inversely proportional to the temperature in which the insect was reared, with average developmental times for males and females varying from 8.7 and 10.2 days to 23.6 and 26.7 days, respectively. Parasitoid development, emergence and survival of the parasitoids were high (96.9% to 100%) at each studied temperature which seemed not to influence these biological parameters. However, daily production of offspring per females varied significantly with the temperature, with greatest production of offspring per day (34.8) occurring at 30°C.

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<sup>1</sup> Centro Nacional de Pesquisa de Soja, EMBRAPA, Caixa postal 1061, 86001-970, Londrina, PR.

At all temperatures, the production peak of offspring occurred in the first and second days of adult life, with a maximum total of 194.9 eggs per female of *T. basalis*, at 22°C. Total egg production did not differ significantly among the different temperature regimes.

KEY WORDS: Insecta, biological control, egg parasitoid, *Nezara viridula*.

## INTRODUCTION

*Trissolcus basalis* (Woll.) is an important egg parasitoid of *Nezara viridula* (L.) and several other species of Pentatomidae associated with soybean (Corrêa-Ferreira 1980, 1991). It has shown considerable potential as a control agent of these pest populations in different crops. From Egypt this parasitoid was introduced into Australia in 1933 for *N. viridula* control (Wilson 1960) and after into various countries for biological control programs, such as New Zealand in 1949 (Cumber 1951), Hawaii in 1962 (Davis 1964) and more recently (1980-1982) Argentina and Taiwan (Crouzel & Saini 1983, Su & Tseng 1984), with outstanding success in certain regions (Caltagirone 1981, Clarke 1990). *T. basalis* was found for the first time in Brazil in 1979, parasiting *N. viridula* eggs in Londrina, Paraná region (Corrêa-Ferreira 1980). This Brazilian race was introduced into Easter Island, Chile, in 1987, to control *N. viridula* (Corrêa-Ferreira, unpublished).

Biological and reproductive rates of a natural enemy is important in assessing its potential efficiency as a biological control agent. Noble (1937), Kamal (1937), Wilson (1961) and Ganesalingam (1966) studied aspects of the reproductive biology of *T. basalis*. Several others researched the influence of different biotic factors on the developmental cycle of this parasitoid (Powell *et al.* 1981, Powell & Shepard 1982, Orr *et al.* 1985, Foerster & Ghezzi 1988, Corrêa-Ferreira & Zamataro 1989, Awan *et al.* 1990).

Considering the great diversity of factors which influence the biology and behavior of egg parasitoids and the importance of these biological characteristics for the success of these agents in the control of host populations (Caltagirone 1985), this study was set out to quantify and compare various of these different parameters. The ideal temperatures for the expression of *T. basalis* maximum parasitism potential towards its preferred host *N. viridula* were also studied. These data can be of assistance for the appropriate use of this agent in biological control programs for the stink bug complex in soybean under Brazilian conditions.

## MATERIAL AND METHODS

A Brazilian breed of *T. basalis*, originally collected in soybean fields in Londrina, Paraná region, was used for the biological and behavioral tests. Eggs of *N. viridula* from a colony maintained in the laboratory were used as hosts, according to the methodology described by Corrêa-Ferreira (1985). The time of development and the percentual emergence of *T. basalis* adults were assessed at constant temperatures of 18°, 22°, 26° and 30°C, in B.O.D. type incubators with 14 hours photophase and relative humidity of 65% ± 10%, with 15 replications per temperature. Newly emerged *T. basalis* females were separated in 8.0 cm x 2.5 cm glass tubes and honey fed. Two days after the emergence, 40 *N. viridula* eggs were offered to female parasitoids for four hours, at each temperature. After this period the eggs were separated in Petri dishes with adequate humidity and maintained under the temperature conditions in which the immatures developed.

During the period of adult emergence the eggs were observed daily, at same fixed time, to record the date of emergence and the sex of each adult parasitoid. Those eggs that remained intact for several days after the emergence period were dissected to check their contents. Only eggs that produced a recognizable parasitoid were considered parasitized. Eggs which did not show evidence of the immature phases of the parasitoid were not included in the calculations since it was not possible to determine whether they were parasitized, sterile or damaged.

The following data were recorded for each temperature under study: developmental time from oviposition to adult emergence; percentage of developed adults (number of adults which developed but did not necessarily emerged, relatively to the total number of parasitized eggs); percentage of adult emergence (number of emerged adults relatively to the number of eggs in which the parasitoids developed as far as the adult stage); percentage of survival from egg to adult emergence (number of adult parasitoid that emerged from the total number of parasitized eggs), and sex ratio (number of females relatively to males and females produced).

To better identify where temperature is more influent this experiment was repeated, at a second stage, following the methodology already described. Now, after the exposure of the eggs to the parasitoids, they were separated in Petri dishes and maintained at constant temperature of 26°C during the development period until complete emergence of the adults. The observations and analyses were identical to those carried out in the first phase of the test. The reproductive performance of the *T. basalis* females when subjected to the different temperatures, was assessed for a period of 12 days. Shortly after emergence and mating, each female was separated in 8.0 cm x 2.5 cm glass tubes and offered an average of 60 *N. viridula* eggs. These were removed after 24 hours and replaced with a new batch of eggs. Eggs stored at 8°C for a period of one to thirty days were used for this test.

The eggs that had been exposed to the parasitoids were placed in Petri dishes with adequate moisture and kept at laboratory environmental conditions. *N. viridula* nymphs which emerged from non-infested eggs were removed to avoid possible damage to the non-emerged parasitoids. After the emergence and death of the adult parasitoids, they were counted and sexed. The remaining eggs were dissected and their content analyzed. Developmental time, emergence of the adults, incidence of parasitism and females reproductive capacity at different temperatures were established in ten replications, in a completely randomized design. An analysis of variance was carried out (ANOVA) and the means compared by the Duncan test (5%).

## RESULTS AND DISCUSSION

**Time of Development and Emergence.** Developmental time, from oviposition to adult emergence of *T. basalis* in eggs of *N. viridula*, was significantly different among the various temperature regimes (Table 1). Developmental time for *T. basalis* males and females was inversely proportional to the temperatures, with longer periods at lower temperatures (Table 1). The results also show that the average developmental time was shorter for *T. basalis* males than for females. This was also found in other Scelionidae species such as *Telenomus podisi* Ashm. (Yeargan 1980) and *Trissolcus euschisti* (Ash.) (Yeargan 1982). Male emergence began one to three days before female emergence, depending on the temperature conditions (Fig. 1). The average period needed for male development at the temperatures 26°, 22° and 18°C was, respectively, 2.1, 6.3 and 14.9 days greater than at 30°C. Likewise females kept at 30°C had shorter developmental time than those females kept at 26°C, 22°C and 18°C.

At each temperature, the percentage of developed adults, emerged adults and percentage

Table 1. Developmental time of *Trissolcus basalis* from oviposition to the adult emergence at four temperatures.

Temperature	Development egg to adult (days) <sup>1</sup>			
	Male		Female	
	X ± SE	Range	X ± SE	Range
18°C	23,6 ± 0,3a	21-26	26,7 ± 0,1a	24-32
22°C	15,0 ± 0,1b	14-16	17,1 ± 0,1b	16-21
26°C	10,8 ± 0,1c	10-12	12,1 ± 0,1c	11-14
30°C	8,7 ± 0,2d	8-10	10,2 ± 0,1d	9-11

<sup>1</sup> Means followed by the same letter do not differ at the 5% level according to Duncan's multiple range test.

of survival (from egg to adult) in *T. basalis* were high (96.9% to 100%). Thus, the influence of temperature on these parameters was not significant (Table 2). Similar behavior was observed when the eggs were parasitized at the different temperatures and maintained at 26°C for development (Table 3). These values were slightly higher than those recorded by Orr *et al.* (1985) for a breed of *T. basalis* in Louisiana, at the same temperature intervals. These authors, however, noted significant reduction in these parameters for temperatures of 15° and 36°C.

The difficulty in determining the real number of parasitized eggs has been discussed for other species. Obviously, the number of emerged adults supplies only a partial indication of the total number of parasitized eggs as non-emerged parasitoids are often present in immature

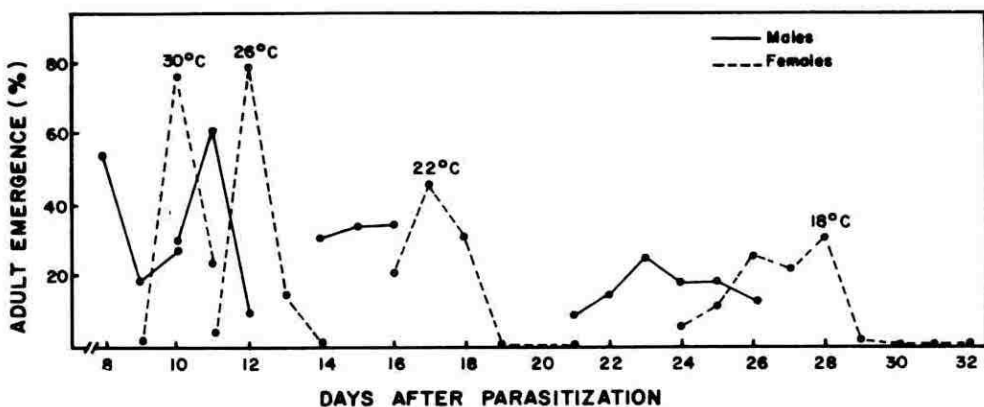


Figure. 1. Percentage of emergence of *Trissolcus basalis* adults from eggs of *Nezara viridula* submitted to different temperatures.

Table 2. Percent of adult development, adult emergence, survival, egg parasitism, and the sex ratio of *Trissolcus basalis* in *Nezara viridula* eggs at four temperatures.

Parameters	Temperature <sup>1</sup>			
	30°C	26°C	22°C	18°C
Adult development(%)	99.1 <sup>NS</sup>	100.0 <sup>NS</sup>	98.7 <sup>NS</sup>	100.0 <sup>NS</sup>
Adults emerged(%)	97.8 <sup>NS</sup>	100.0 <sup>NS</sup>	99.8 <sup>NS</sup>	100.0 <sup>NS</sup>
Survival (egg-adult)(%)	96.9 <sup>NS</sup>	100.0 <sup>NS</sup>	98.5 <sup>NS</sup>	100.0 <sup>NS</sup>
Egg parasitism(%)	53.5 c	99.7 a	72.9 b	82.5 b
Sex ratio	0.86 b	0.93 a	0.92 a	0.91 ab

<sup>1</sup> Means followed by the same letter, in the line, do not differ at the 5% level according to Duncan's multiple range test.

<sup>NS</sup> = not significant.

<sup>N</sup> = 600 (total number in each temperature).

forms. In this study, only eggs from which *T. basalis* adults emerged, or in which perceptible immature forms of the parasitoid were identified, were considered as parasitized eggs. The highest level of parasitism (99.7%), obtained at 26°C, was significantly higher than the levels of 83.5%, 72.9% and 53.5% observed at 18°, 22° and 30°C, respectively (Table 2).

The higher temperature effect was observed at 30°C with only 53.5% of the eggs parasitized. This result differs from that of Foerster & Ghezzi (1988) where the smallest proportion of parasitized eggs (77.0%) was found at 22°C. Temperature influence was reduced when development took place at a more suitable temperature (26°C). In this study 26°C was the temperature at which the adults performed best, with 98.2% of egg parasitism. This level of parasitism, however, did not differ from that obtained at 22°C (95.5%) (Table 3). At this temperature regime there was a substantial increase in egg parasitism, when the eggs were transferred to a more suitable temperature for parasitoid development (Table 3). Here the lowest parasitization was also found at 30°C (60.5%).

The average sex ratio in the *T. basalis* offspring was similar at 26°, 22° and 18°C, showing values of 0.93, 0.92 and 0.91, respectively (Table 2). These sex ratios were statistically superior to the ratio of 0.86 obtained at 30°C, corresponding to one male per six females. However, when the eggs were parasitized at the different temperatures and development took place at 26°C, the difference between the sex ratios in the offspring were smaller (Table 3). Sex ratios of 0.91, 0.93, 0.94 and 0.93 were obtained for the temperatures of 30°, 26°, 22° and 18°C, respectively.

Table 3. Percent of adult development, adult emergence, egg parasitism, and the sex ratio of *Trissolcus basalis* in *Nezara viridula* eggs, when parasitoids were submitted at different temperatures during oviposition and kept at 26°C up to adult emergence.

Parameters	Temperature <sup>1</sup>			
	30°C	26°C	22°C	18°C
Adult development(%)	94.8 <sup>NS</sup>	99.4 <sup>NS</sup>	98.9 <sup>NS</sup>	100.0 <sup>NS</sup>
Adults emerged(%)	100.0 <sup>NS</sup>	100.0 <sup>NS</sup>	99.8 <sup>NS</sup>	100.0 <sup>NS</sup>
Survival (egg-adult)(%)	95.2 <sup>NS</sup>	99.4 <sup>NS</sup>	98.7 <sup>NS</sup>	100.0 <sup>NS</sup>
Egg parasitism(%) <sup>2</sup>	60.5 c	98.2 a	95.5 ab	88.6 b
Sex ratio	0.91 b	0.93 a	0.94 a	0.93 ab

<sup>1</sup> Means followed by the same letter, in the line, do not differ at the 5% level according to Duncan's multiple range test.

<sup>NS</sup> = not significant.

<sup>N</sup> = 600 (total number in each temperature).

<sup>2</sup> Transformed data by arc sen  $x/100$ .

Under these conditions, at 30°C one male was produced for 10.3 females, a superior proportion to that obtained when the parasitism and development took place at a constant temperature of 30°C.

**Reproductive Capacity.** Table 4 shows the average total offspring generated during the first twelve days of adult life and the daily offspring production by *T. basalis* females in different temperatures. Daily offspring production per female varied at different temperatures. The largest average number of offspring generated per female and per day, 34.8, took place at 30°C. This value was significantly higher than those observed at other temperatures. However, when the total offspring production was compared there were no significant differences between the temperatures tested. The largest absolute values occurred at 22° and 26°C with 194.9 and 187.1 offsprings, respectively.

From the curve of offspring generated per day by the female throughout the first twelve days of adult life (Fig. 2) it was observed that, for each temperature studied, the production peak took place on the first and second days of life. This result agrees with those of Corrêa-Ferreira & Zamataro (1989) and Awan *et al.* (1990) for *T. basalis* and with those of Yeargan (1982) for *T. podisi* and *T. euschisti*, with the exception that, for *T. basalis*, at 22°C the maximum production occurred on the sixth day.

Fertility of female *T. basalis* has been studied by various authors, and variable values have

Table 4. Total egg production<sup>1</sup> and daily reproductive capacity of *Trissolcus basalis* females, parasitizing *Nezara viridula* eggs, at four temperatures.

Temperature	Total offspring ( $\bar{X} \pm SE$ )	Offspring/female/day ( $\bar{X} \pm SE$ )
18°C	138.8 ± 15.7 a <sup>2</sup>	18.9 ± 1.0 c
22°C	194.9 ± 27.5 a	21.0 ± 1.7 c
26°C	187.1 ± 31.3 a	25.7 ± 0.6 b
30°C	173.2 ± 21.9 a	34.8 ± 2.2 a

<sup>1</sup> For determination of total number of eggs, counts were made up the 12th day after adult emergence.

<sup>2</sup> Means followed by the same letter, in the coluns, do not differ at the 5% level according to Duncan's multiple range test.

been found according to the methodologies and breeds used. The values obtained in this study were superior to those of Noble (1937), Wilson (1961) and Ganesalingam (1966) and lower than those of Corrêa-Ferreira & Zamataro (1989) and Awan *et al.* (1990). The smaller fertility rate of this study, comparatively to that of the latter authors, can be explained by the lower exposure time of the host eggs (twelve days) to parasitization and by the heterogeneity of the

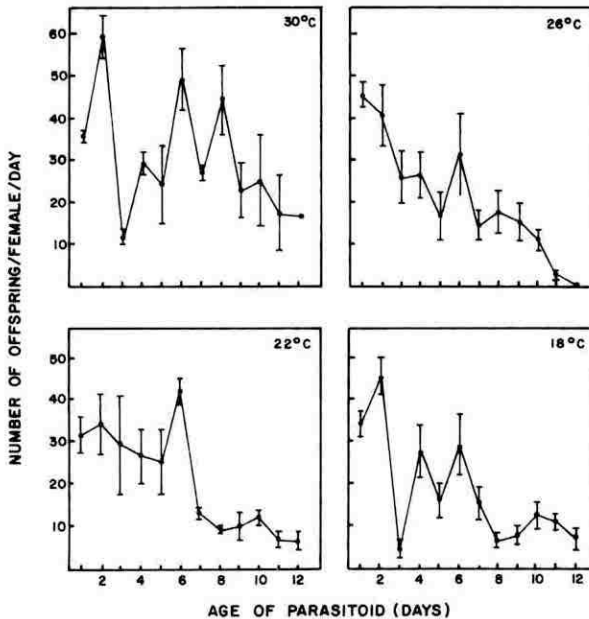


Figure 2. Daily production of offspring by *Trissolcus basalis* females at different temperatures.



eggs used in these experiments.

Development and reproductive cycle of the *T. basalis* were already studied by several authors (Noble 1937, Wilson 1961, Lee 1979, Power & Shepard 1982, Foerster & Ghezzi 1988). The results of this study, however, allowed a better understanding of the behavior of *T. basalis* breed adapted to the Brazilian conditions, with data that will be very useful for the biological control program of soybean stink bugs across of massal breeding in the laboratory and *T. basalis* releases in the field.

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