Telenomus podisi parasitism on *Dichelops melacanthus* and *Podisus nigrispinus* eggs at different temperatures

Érica Ayumi Taguti[1], Jaciara Gonçalves[1], Adeney de Freitas Bueno[2,], and Suelhen Thais Marchioro[3]*

Abstract

Dichelops melacanthus (Dallas) (Heteroptera: Pentatomidae) is the most important stink bug species that feeds on maize in South America, and it is frequently controlled with chemical pesticides. As an alternative, more sustainable management strategies can be applied, among which the egg parasitoid *Telenomus podisi* Ashmead (Hymenoptera: Scelionidae) stands out. However, *T. podisi* can have the undesired effect of parasitizing the predator and biocontol agent *Podisus nigrispinus* (Dallas) (Hemiptera: Pentatomidae). Therefore, the study of *T. podisi* parasitism on eggs of both *D. melacanthus* and *P. nigrispinus* is of theoretical and practical interest. Individual 48-h-old parasitoids were offered a card with host eggs. Parasitism was allowed for 24 h at 25 ± 2 °C. Afterwards, the eggs were exposed to temperatures of 15, 20, 25, or 30 ± 2 °C to test the effects of temperature on the development of immature stages of the parasitoid (egg-to-adult period, emergence (%), and sex ratio). In addition, eggs were offered on a daily basis to parasitoid females exposed to 15, 20, 25, or 30 ± 2 °C. Parasitism was allowed for 24 h before eggs were replaced with fresh ones (≤ 24 h) and stored at 25 ± 2 °C until emergence. This allowed us to study the effects of temperature on adult parasitism capacity (daily parasitism, accumulated percentage of parasitism, parental female longevity, and total number of parasitized eggs per female). Our results show that temperature significantly influenced duration of the egg-to-adult period, emergence, sex ratio, total number of parasitized eggs, and parental female longevity of *T. podisi* on both host species. Development time of the parasitoid was reduced with increasing temperature. Emergence above 80% was observed at temperatures of 20 and 25 °C in eggs of *D. melacanthus*, and at 20, 25, and 30 °C in eggs of *P. nigrispinus*. In both hosts, the ratio of females (sex ratio) was highest at the lowest temperature (15 °C). In both host species, daily parasitism and total number of parasitized eggs decreased with time, and longevity of females was inversely proportional to an increase in temperature. These results allow us to conclude that extreme temperatures of 15 and 30 °C are not favorable for *T. podisi* parasitism, even though parasitism was still observed. Therefore, in regions where those extreme temperatures are common, additional studies are necessary to explore the need for a higher number of parasitoids for successful field releases. Even though the release of *T. podisi* in the field may negatively impact the predator *P. nigrispinus*, it is problaly still safer than the use of chemical insecticides, which would be the alternative measure to *T. podisi* in the control of stink bugs.

Key Words: biological control; egg parasitoid; temperature effects

Resumen

Dichelops melacanthus (Dallas) (Heteroptera: Pentatomidae) es la especie de chinche más importante que ataca el cultivo del maíz en América del Sur, con frecuencia es controlada con insecticidas químicos. Entre las estrategias de manejo sostenibles, se destaca el control biológico con el parasitoide de huevos, *Telenomus podisi* Ashmead (Hymenoptera: Scelionidae). Sin embargo, *T. podisi* también puede parasitar al predador *Podisus nigrispinus* (Dallas) (Hemiptera: Pentatomidae), aspecto considerado no deseable. Por lo tanto, el estudio del parasitismo de *T. podisi* en huevos de *D. melacanthus* y *P. nigrispinus* es de interés teórico y práctico. Huevos de estas especies fueron ofrecidos a parasitoides de 48 h de edad, en forma individual. El parasitismo fue permitido durante 24 h a 25 ± 2 °C. Posteriormente, estos huevos se expusieron a una temperatura de 15, 20, 25, o 30 ± 2 °C, para verificar el efecto de la temperatura en el desarrollo de estados inmaduros del parasitoide fueron examinados el período de huevo a adulto, emergencia (%) y proporción de sexos. Además, los huevos se ofrecieron diariamente a parasitoides hembras expuestas a 15, 20, 25, o 30 ± 2 °C. Se permitió el parasitismo durante 24 h a esas temperaturas y luego se reemplazaron los huevos por otros frescos (≤ 24 h) y se mantuvieron a 25 ± 2 °C hasta la emergencia. Por lo tanto, se estudiaron los efectos de la temperatura en la capacidad de parasitar de los adultos (parasitismo diario, porcentaje acumulado de parasitismo, longevidad parental femenina y número total de huevos parasitados por hembra). Nuestros resultados muestran que la temperatura influyó sobre la duración del período de huevo a adulto, en la emergencia, en la proporción de sexos, en el número total de huevos parasitados y en la longevidad parental femenina expuesta a huevos de *D. melacanthus* y *P. nigrispinus*. El tiempo de desarrollo del parasitoide se redujo con el aumento de la temperatura. Emergencia superior al 80% se observó en temperaturas de 20 y 25 °C en huevos de *D. melacanthus*, y a 20, 25, y 30 °C en huevos de *P. nigrispinus*. En ambos huéspedes, el porcentaje de hembras (proporción de sexos) fue mayor a la temperatura más baja (15 °C). La capacidad de parasitismo, la longevidad de las hembras parentales y el número total de huevos parasitados variaron con la temperatura en ambas especies hospedadoras, lo que mostró que el parasitismo diario y el número total de huevos parasitados disminuyeron con el tiempo y que la longevidad de las hembras fue inversamente proporcional al aumento de la temperatura. Estos resultados permiten concluir que las temperaturas extremas de 15 y 30 °C no fueron favorables para el parasitismo de *T. podisi*, a pesar de que hubo parasitismo. Por lo tanto, en las regiones donde esas temperaturas extremas pueden ser comunes, se deben realizar estudios adicionales para verificar la posibilidad de liberar un

[1]Programa de Pós-Graduação em Entomologia, Departamento de Zoologia, Setor de Ciências Biológicas, Universidade Federal do Paraná, Curitiba, Paraná , Brazil; E-mail: ericataguti@hotmail.com (E. A. T.), jaciarag@gmail.com (J. G.)
[2]Embrapa Soja, Rodovia Carlos João Strass, s/n, Caixa postal 231 - Distrito de Warta, 86001-970 Londrina, Paraná, Brazil; E-mail: adeney.bueno@embrapa.br (A. D. F. B.)
[3]Universidade Federal da Fronteira Sul, Campus Laranjeiras do Sul, Rodovia BR 158 - km 405, 85301-970 Laranjeiras do Sul, Paraná, Brazil;
E-mail: suelhenmarchioro25@hotmail.com (S. T. M.)
*Corresponding author; E-mail: adeney.bueno@embrapa.br

mayor número de parasitoides en estas circunstancias. A pesar de que la liberación de *T. podisi* en el campo podría afectar negativamente al predador *P. nigrispinus*, esta estrategia de control de plagas es probable que sea aún más seguro que el uso de insecticidas químicos, que sería la medida alternativa a *T. podisi* en el control de las chinches.

Palabras Claves: control biológico; parasitoide de huevo; efectos de la temperatura

Dichelops melacanthus (Dallas) (Heteroptera: Pentatomidae), the most important stink bug species feeding on maize in South America, is controlled frequently with chemical pesticides (Bueno et al. 2015). Pest outbreaks are favored by a "green bridge," i.e., by cultivating 2 crops per yr (soybean in summer, maize in fall and winter) (Bianco 2005) when an increase in the *D. melachanthus* population is observed during the soybean season (Chiesa et al. 2016).

The early presence of stink bugs in maize development frequently has triggered an overuse of insecticides. Current pest management adopted in the field still primarily depends on chemicals (van Lenteren & Bueno 2003). In spite of this overuse of chemical insecticides, stink bug outbreaks tend to occur earlier and with greater intensity in soybean and maize each yr (Bueno et al. 2015). Therefore, a more efficient and sustainable pest management approach is urgently needed. One of the most sustainable pest management strategies of integrated pest management, which has been applied increasingly worldwide, is augmentative biological control (van Lenteren et al. 2018).

Egg parasitoids are the most important group of biocontrol agents used in stink bug augmentative biological control management, because pests are controlled at a stage prior to plant damage (Koppel et al. 2009). *Telenomus podisi* Ashmead (Hymenoptera: Scelionidae) is an egg parasitoid available for field releases in augmentative biological control that acts as a biocontrol agent of different pentatomid stink bug species. However, the family Pentatomidae includes pests as well as biocontrol agents, such as the predator *Podisus nigrispinus* (Dallas) (Hemiptera: Pentatomidae), which also is parasitized by *T. podisi* (Torres et al. 1996; Medeiros et al. 1997; Pacheco & Corrêa-Ferreira 2000; Koppel et al. 2009; Margaría et al. 2009).

An important consideration for *T. podisi* parasitism in the field is the possible impact of abiotic conditions, especially temperature, which directly influences development and survival of insects (Wilson & Barnett 1983), affecting sex ratio, emergence, and other biological characteristics (Canto-Silva et al. 2005). Therefore, *T. podisi* parasitism is directly associated with its ability to adapt to different hosts and climatic conditions. To this end, the objective of this work was to study the effects of temperature on *T. podisi* parasitism on eggs of *D. melacanthus* and *P. nigrispinus*.

Materials and Methods

PARASITOID AND HOST COLONIES

Telenomus podisi females as well as the studied hosts, *D. melacanthus* and *P. nigrispinus*, originated from insect colonies kept at Embrapa Soybean, Londrina, Paraná, Brazil. Colonies were kept under controlled environmental conditions inside biochemical oxygen demand climate chambers (ELETROLab®, model EL 212, São Paulo, São Paulo, Brazil) set at 80 ± 10% humidity, temperature of 25 ± 2 °C, and a 14:10 h (L:D) photoperiod. Hosts and parasitoids were reared according to the methodologies described by Peres and Corrêa-Ferreira (2004) for *T. podisi*, by Panizzi et al. (2000) for *D. melacanthus*, and by Denez et al. (2014) for *P. nigrispinus*, and are briefly summarized in the following paragraph.

Telenomus podisi was collected originally from soybean fields in Londrina, Paraná, Brazil. The population has been maintained in the laboratory for approximately 10 yr. It is reared on eggs of *Euschistus heros* (Fabricius) (Hemiptera: Pentatomidae) glued to pieces of cardboard (2 cm × 8 cm), and introduced into tubes together with eggs already parasitized by *T. podisi* close to parasitoid emergence. Small drops of honey are placed inside these tubes to provide food for the adults when they emerge. The tubes are then closed, and the eggs allowed to be parasitized for 24 h. Adults that emerge from these eggs are used for trials as well as for colony maintenance.

Dichelops melacanthus and *P. nigrispinus* were collected from soybean plants in Londrina, Paraná, Brazil. Those populations were kept in the laboratory for approximately 2 yr during which new field insects were introduced each yr to maintain colony quality. *Dichelops melacanthus* was fed with beans (*Phaseolus vulgaris* L.; Fabaceae), soybeans (*Glycine max* L. Merr.; Fabaceae), peanuts (*Arachis hypogaea* L.; Fabaceae), sunflower seeds (*Helianthus annuus* L.; Asteraceae) and privet fruits (*Ligustrum lucidum* Aiton; Oleaceae). *Podisus nigrispinus* was fed with third to fifth instars of *Anticarsia gemmatalis* Hübner (Lepidoptera: Erebidae) and *Spodoptera frugiperda* (Smith) (Lepidoptera: Noctuidae) from the laboratory colony. *Dichelops melacanthus* and *P. nigrispinus* were kept in cages (20 cm × 20 cm sides × 24 cm tall) made of plastic screen and lined with filter paper. A Petri dish with a cotton wad soaked in distilled water (9 cm diam) was added to each cage. Three times per wk, cages were cleaned, stink bug food was replaced, and egg masses collected. After collection, some egg masses were transferred to acrylic boxes (11 cm × 11 cm × 3.5 cm) (Gerbox, Adria laboratórios Ltda., Londrina, Paraná, Brazil) lined with filter paper moistened with sterile, distilled water. After eclosion, second instars were transferred to new cages identical to those previously described. Egg masses not used for stink bug colony maintenance were stored in gallon containers of liquid nitrogen at −196 °C for later use in the parasitoid experiments.

BIOASSAYS

Four bioassays were independently conducted, 2 of which were used to study the biological characteristics of *T. podisi* developing in eggs of *D. melacanthus* and *P. nigrispinus* at different temperatures (1 trial for each host species). The other 2 bioassays were designed to study the parasitism capacity of adult *T. podisi* on eggs of *D. melacanthus* and *P. nigrispinus* at different temperatures (1 trial for each host species).

Before beginning the experiments, *T. podisi* was reared for 1 generation on eggs of the respective experimental host species in order to eliminate a possible pre-imaginal conditioning by rearing them on the alternative host (*E. heros*). Thus, experiments were carried out with the second generation of the parasitoid reared on the host eggs.

BIOLOGICAL CHARACTERISTICS OF *TELENOMUS PODISI* DEVELOPING IN EGGS OF *DICHELOPS MELACANTHUS* (TRIAL 1) AND *PODISUS NIGRISPINUS* (TRIAL 2) AT DIFFERENT TEMPERATURES

The bioassays were conducted in a completely randomized design with 4 treatments (15, 20, 25, and 30 ± 2 °C) and 7 replicates (N = 28). Each replicate consisted of 3 individual females in plastic microtubes of 12 mm diam and 75 mm height (7 replicates × 3 females per repli-

cate = 21 females evaluated per treatment) following the methodology used by Queiroz et al. (2017) for other species of the genus *Telenomus*. Rather than using a single female parasitoid, a group of females was used per replicate: female insect egg parasitoids are small and fragile, and thus vulnerable to tiny injuries during experimental manipulation, which could affect their behavior. Using a set of parasitoids for each replicate can mitigate this potential negative effect to some extent.

Telenomus podisi females (48 h old) were placed individually into glass tubes (12 mm diam × 75 mm height). A cardboard card (10 mm × 70 mm) containing 40 eggs of each host (*D. melacanthus* for trial 1, and *P. nigrispinus* for trial 2) was exposed to each female. A droplet of honey was offered to the females as food.

Parasitism was allowed for 24 h inside a biochemical oxygen demand climate chamber at 25 ± 2 °C, RH 80 ± 10%, and a 14:10 h (L:D) photoperiod. Afterwards, parasitoids were removed from the tubes and the eggs of each species were transferred to climate chambers at temperatures of 15, 20, 25, and 30 ± 2 °C, with 80 ± 10% relative humidity (RH), and a 14:10 h (L:D) photoperiod. The following biological traits were evaluated: duration of egg-to-adult period, sex ratio (sex ratio = number of females/[number of females + number of males]), and parasitoid emergence (%). The emergence of *T. podisi* was observed daily to determine the duration of the egg-to-adult period. Parasitoid emergence was evaluated under a stereoscope by counting the host eggs with an exit orifice that resulted from adults emerging from the egg.

PARASITISM CAPACITY OF *TELENOMUS PODISI* IN EGGS OF *DICHELOPS MELACANTHUS* (TRIAL 3) AND *PODISUS NIGRISPINUS* (TRIAL 4) AT DIFFERENT TEMPERATURES

The bioassays were performed in a completely randomized design with 4 treatments (15, 20, 25, and 30 ± 2 °C) and 22 replicates containing 1 *T. podisi* female for each host species. Eggs of both *D. melacanthus* and *P. nigrispinus* stored in liquid nitrogen were removed daily from storage to be used in the experiments.

Forty-eight-h-old females were individually placed into Duran tubes (1.5 mL) (Dovil Ltda., São Paulo, São Paulo, Brazil) containing cardboard cards (0.8 cm × 5 cm) with approximately 40 eggs of *D. melacanthus* (trial 3) or *P. nigrispinus* (trial 4). A droplet of honey was placed on the wall of the tube for feeding, and tubes were then sealed with plastic film.

The tubes were kept inside biochemical oxygen demand climate chambers set to the respective treatment temperature, 80 ± 10% RH, and a 14:10 h (L:D) photoperiod. The eggs were exposed to parasitism for 24 h, and the cards were replaced daily until the females died. The cards containing parasitized eggs were stored in plastic bags (4 cm × 23 cm) and kept inside a biochemical oxygen demand climate chamber at 25 ± 2 °C, 80 ± 10% RH, and a 14:10 h (L:D) photoperiod until emergence and death of adults. The parameters evaluated were daily parasitism, cumulative percentage of parasitism, total number of eggs parasitized per female, and longevity of parental females.

DATA ANALYSIS

The results obtained in the experiments were submitted to exploratory analysis to evaluate the normality assumptions of the residuals (Shapiro & Wilk 1965), homogeneity of variance of treatments, and additivity of the model to allow the application of ANOVA (Burr & Foster 1972). Egg to adult period (trial 1) and longevity of parental females (trial 3) were square-root transformed. Emergence of *T. podisi* from *P. nigrispinus* eggs (trial 2) was transformed into arcsine $\sqrt{X/100}$. Sex ratio of *T. podisi* emerged from *P. nigrispinus* eggs (trials 2 & 4) were transformed into to perform ANOVA. Then, averages were compared by the Tukey test at a 5% error probability, with the statistical analysis program SAS (SAS Institute 2009).

Results

BIOLOGICAL CHARACTERISTICS OF *TELENOMUS PODISI* DEVELOPING IN EGGS OF *DICHELOPS MELACANTHUS* (TRIAL 1) AND *PODISUS NIGRISPINUS* (TRIAL 2) AT DIFFERENT TEMPERATURES

Egg to adult periods of *T. podisi* in eggs of *D. melacanthus* (trial 1) and *P. nigrispinus* (trial 2) were inversely related to an increase in temperature (10.8 and 10.9 d at 30 °C, and 58.8 and 67.2 d at 15 °C, respectively). Thus, an increase in temperature from 15 to 30 °C caused a reduction of the egg to adult period of *T. podisi* in eggs of *D. melacanthus* by 48.0 d, and in eggs of *P. nigrispinus* by 56.3 d (Table 1).

The emergence (%) of *T. podisi* offspring from *D. melacanthus* and *P. nigrispinus* eggs was influenced by temperature, with the highest parasitoid emergence from *D. melacanthus* eggs at 20 °C, and from *P. nigrispinus* eggs at 25 and 30 °C (Table 1). Emergence from eggs of *D. melacanthus* was ≥ 80% at 20 and 25 °C (97.4 and 80.5%, respectively), but much lower at 15 °C (27.5%). Emergence (%) from eggs of the predator *P. nigrispinus* was ≥ 80% at temperatures of 20, 25, and 30 °C (87.7, 98.4, and 97.7%, respectively), and also lower at 15 °C (3.8%) (Table 1).

Temperature influenced sex ratio (sex ratio = number of females/[number of males + number of females]), which decreased with increasing temperature in both hosts (*D. melacanthus* and *P. nigrispinus*) (Table 1). However, sex ratio was always above 0.5 at all evaluated temperatures, both in pest and predator eggs (Table 1). The highest sex ratio was found when parasitoids were exposed to 15 °C on both host species. The lowest value was recorded at 25 °C in *D. melacanthus* eggs and at 30 °C in *P. nigrispinus* eggs (Table 1).

PARASITISM CAPACITY OF *TELENOMUS PODISI* IN EGGS OF *DICHELOPS MELACANTHUS* (TRIAL 3) AND *PODISUS NIGRISPINUS* (TRIAL 4) AT DIFFERENT TEMPERATURES

Parasitism capacity of *T. podisi* was affected by temperature in both hosts (Figs. 1 & 2). The number of parasitized eggs per d varied depending on temperature and host, with the highest parasitism always observed on the first d of each experiment. Cumulative parasitism (%) of *D. melacanthus* eggs reached 80% on the 24th, 20th, 14th, and 8th d of adult lifespan at 15, 20, 25, and 30 °C, respectively (Fig. 1). Similarly, cumulative parasitism (%) of *P. nigrispinus* eggs reached 80% on the 18th, 17th, 14th, and 12th d of adult lifespan at 15, 20, 25, and 30 °C, respectively (Fig. 2).

The number of eggs laid per d by parasitoid females (daily parasitism) decreased constantly in both host species over the adult parasitoid lifespan (Figs. 1 & 2). In *D. melacanthus* eggs, parasitism was highest within the first 24 h at all tested temperatures, when average numbers of parasitized eggs were 4.1, 10.6, 10.9, and 12.3 at 15, 20, 25, and 30 °C, respectively (Fig. 1). In *P. nigrispinus* eggs, parasitism was highest during the first 24 h only at 20 and 25 °C, whereas at 15 °C (Fig. 2A) and 30 °C (Fig. 2D), maximum parasitism was observed on the second and fourth d, with 0.5, 8.2, 9.7, and 10.8 eggs parasitized at temperatures of 15, 20, 25, and 30 °C, respectively (Fig. 2).

Longevity of parental females of *T. podisi* was inversely proportional to the increase in temperature on both hosts (Table 2). Reared on *D. melacanthus* eggs, the maximum longevity of *T. podisi* parental females was 89.0 d at 15 °C, and decreased to 41.7, 30.8, and 13.9 d

Table 1. Biological trait values of *Telenomus podisi* Ashmead (Hymenoptera: Scelionidae) on eggs of *Dichelops melacanthus* (Dallas) (Hemiptera: Pentatomidae) and *Podisus nigrispinus* (Dallas) (Hemiptera: Pentatomidae) at 80 ± 10% RH and a 14:10 h (L:D) photoperiod.

Host	Temperature (°C)	Egg to adult (d)[1]	Emergence (%)[1]	Sex ratio[1]
D. melacanthus (trial 1)	15	58.8 ± 0.8 a[2]	27.5 ± 3.2 c[3]	0.90 ± 0.05 a
	20	22.1 ± 0.1 b	97.4 ± 1.2 a	0.71 ± 0.06 ab
	25	12.6 ± 0.2 c	80.5 ± 6.6 b	0.50 ± 0.05 b
	30	10.8 ± 0.3 d	69.7 ± 6.8 b	0.57 ± 0.05 b
	CV (%)	2.12	17.69	20.47
	P	< 0.0001	< 0.0001	0.0002
	df_{error}	24	24	23
	F	2,727.13	29.33	10.44
P. nigrispinus (trial 2)	15	67.2 ± 0.5 a[2]	3.8 ± 1 c[3]	1.00 ± 0.00 a[4]
	20	21.1 ± 0.1 b	87.7 ± 1.4 b	0.92 ± 0.01 b
	25	14.1 ± 0.1 c	98.4 ± 0.4 a	0.89 ± 0.03 b
	30	10.9 ± 0.2 d	97.7 ± 0.8 a	0.60 ± 0.01 c
	CV (%)	1.31	6.92	1.41
	P	< 0.0001	< 0.0001	< 0.0001
	df_{error}	22	23	18
	F	6,962.25	470.90	97.86

[1]Means ± SE followed by the same letter (separately for each host) did not differ significantly (Tukey's test, $P > 0.05$).
[2]Original means followed by statistics performed on $\sqrt{X}$ transformed data.
[3]Original means followed by statistics performed on arcsine $\sqrt{X/100}$ transformed data.
[4]Original means followed by statistics performed on $\sqrt{x+0.5}$ transformed data.

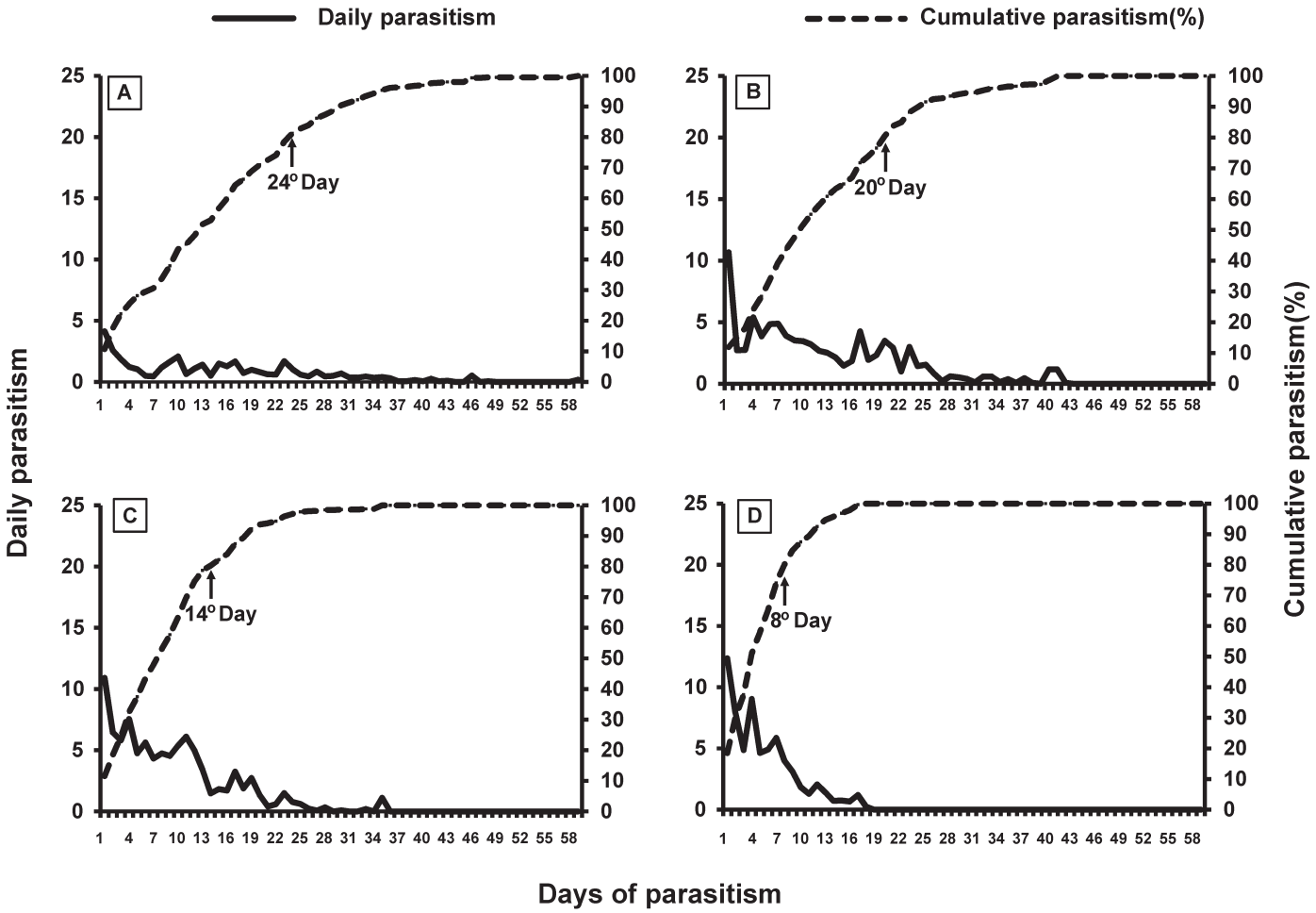

Fig. 1. Distribution of lifetime parasitism of *Telenomus podisi* Ashmead (Hymenoptera: Scelionidae) in *Dichelops melacanthus* eggs (Dallas) (Hemiptera: Pentatomidae) at different temperatures. (A) 15 °C, (B) 20 °C, (C) 25 °C, (D) 30 °C at 80 ± 10% RH and a 14:10 h (L:D) photoperiod. Arrows indicate parasitism of 80%.

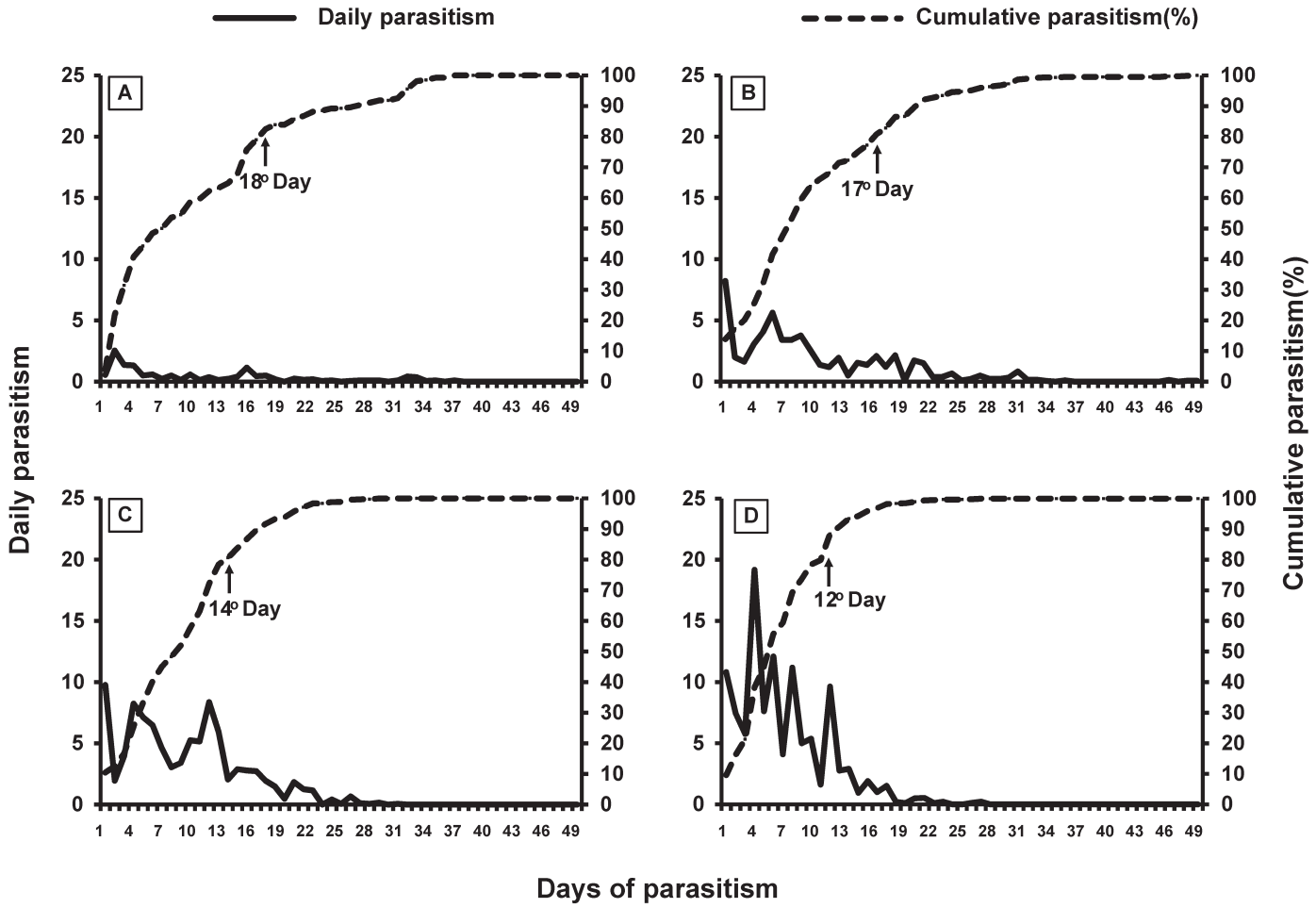

Fig. 2. Distribution of lifetime parasitism *Telenomus podisi* Ashmead (Hymenoptera: Scelionidae) in *Podisus nigrispinus* eggs (Dallas) (Hemiptera: Pentatomidae) at different temperatures. (A) 15 °C, (B) 20 °C, (C) 25 °C, (D) 30 °C at 80 ± 10% RH and a 14:10 h (L:D) photoperiod. Arrows indicate parasitism of 80%.

at temperatures of 20, 25, and 30 °C, respectively. Reared on *P. nigrispinus* eggs, the maximum longevity of *T. podisi* was 133.5 d at 15 °C followed by 59.7, 42.5, and 31.8 d at temperatures of 20, 25, and 30 °C, respectively (Table 2).

The total number of eggs parasitized per female throughout development varied with temperature and host species. Reared on *D. melacanthus* eggs, the number of parasitized eggs did not differ between 20 and 25 °C, with a mean of 101.5 and 100.0 eggs, respectively. At temperatures of 15 and 30 °C, the mean number of parasitized eggs was 40.8 and 67.5, respectively. In eggs of *P. nigrispinus*, the total number of parasitized eggs differed between all tested temperatures (15, 20, 25, and 30 °C, with 13.6, 61.6, 100.9, and 82.6 parasitized eggs, respectively) (Table 2).

Discussion

Telenomus podisi parasitism was highly influenced by both temperature and host species, indicating a possible effect of these parameters on the success of a biocontrol program with this egg parasitoid. Our results are of theoretical and practical interest and can contribute significantly to a successful *T. podisi* release for the management of *D. melacanthus*.

The inverse relationship between increasing temperatures and shorter egg to adult periods is one of the consequences of a higher parasitoid metabolic activity at higher temperatures (Hernández & Diáz 1996; Bueno et al. 2009). Higher metabolic activity implies a higher parasitoid population growth rate, which can be an advantage for biological control as new adults will emerge earlier. Although faster population growth is favorable, an increase in temperature also can have negative effects on *T. podisi* parasitism. At extremely high temperatures, optimal development time is impaired and mortality increases (Bueno et al. 2008). We observed a similar relationship when rearing *T. podisi* on *D. melacanthus* eggs at 30 °C. However, increased larval mortality was not observed in *T. podisi* on *P. nigrispinus* eggs. This might be associated with size differences between *D. melacanthus* (smaller) and *P. nigrispinus* (larger) eggs, as previously observed for other parasitoids (Smith 1996). Host egg size may affect host suitability for a parasitoid. Besides size, other differences between host eggs include egg surface and chorion structure, which might directly impact loss of water and nutrients at 30 °C. The impact of host egg characteristics on parasitoid survival and development has been pointed out already by Cônsoli et al. (1999).

Temperature impacts larval development and survival, as well as several adult biological traits. At 30 °C, parental female longevity was reduced, thus shortening the period of parasitism. It contributed to lowering the total number of parasitized eggs, which is an undesirable effect in biological control. The impact of temperature on the longevity of *T. podisi* adults may be due to an incapability for lipogenesis, as observed for most parasitoid species (Visser & Ellers 2008). However, as an ectothermic insect, the metabolic rate and lipid consumption of *T. podisi* (Huey & Berrigan 2001) will depend

Table 2. Parasitism capacity of *Telenomus podisi* Ashmead (Hymenoptera: Scelionidae) on eggs of *Dichelops melacanthus* (Dallas) (Hemiptera: Pentatomidae) and *Podisus nigrispinus* (Dallas) (Hemiptera: Pentatomidae) at 80 ± 10% RH and a 14:10 h (L:D) photoperiod.

Host	Temperature (°C)	Longevity of parental females (d)[1]	Total number of parasitized eggs per female[1]
D. melacanthus (trial 3)	15	89.0 ± 6.3 a[2]	40.80 ± 2.17 c[2]
	20	41.7 ± 2.4 b	101.58 ± 3.96 a
	25	30.8 ± 2.5 c	100.00 ± 3.98 a
	30	13.9 ± 0.9 d	67.50 ± 4.14 b
	CV (%)	17.50	10.89
	P	< 0.0001	< 0.0001
	df_{error}	76	72
	F	98.32	67.96
P. nigrispinus (trial 4)	15	133.5 ± 5.6 a[2]	13.68 ± 1.96 d[3]
	20	59.7 ± 3.2 b	61.65 ± 4.05 c
	25	42.5 ± 2.2 c	100.94 ± 3.57 a
	30	31.8 ± 1.5 d	82.65 ± 4.95 b
	CV (%)	10.94	15.83
	P	< 0.0001	< 0.0001
	df_{error}	73	76
	F	182.52	123.16

[1]Means ± SE followed by the same letter (separately for each host) did not differ statistically (Tukey's test, $P > 0.05$).
[2]Original means followed by statistics performed on $\sqrt{X}$ transformed data.
[3]Original means followed by statistics performed on $\sqrt{x + 0.5}$ transformed data.

on temperature. Thus, allocation of lipids acquired during the larval stage will determine the adult lifespan and fecundity of *T. podisi* (Visser & Ellers 2008) and, therefore, its lifetime reproductive success (Huey & Berrigan 2001).

Considering that the shortened egg to adult period observed at 30 °C may have reduced the amount of lipids acquired during the *T. podisi* larval development, it probably led to reduced adult longevity and a smaller number of parasitized eggs. Parasitoid foraging decisions are commonly affected by the availability of lipid reserves, and the number of mature eggs (Godfray 1994) in females. Lipids carried over from the larval stage can be allocated to either egg production or to adult lipid reserves, leading to a constant trade-off between reproduction and lifespan (Pexton & Mayhew 2002).

Not only extremely high temperatures, but extremely low temperatures also can impair *T. podisi* parasitism. The low *T. podisi* emergence observed at 15 °C from *D. melacanthus* eggs (27.5%) and *P. nigrispinus* eggs (3.8%) may be related to the proximity of this value to the lower lethal temperature of *T. podisi*. Previous reports support this hypothesis. Yeargan (1980) observed low emergence of *T. podisi* on eggs of *Podisus maculiventris* (Say) (Hemiptera: Pentatomidae) at 15.5 °C, and also Nakama & Foerster (2001) and Torres et al. (1997) who reported a viability of less than 50% when *T. podisi* was exposed to 15 °C. However, it is important to consider that extremely low or high temperatures usually do not occur over long periods in the field and, therefore, field trials which help to understand the real effects of extreme weather conditions on *T. podisi* still need to be conducted.

Another important trait to be considered in biological control is sex ratio (number of females/[number of females + number of males]), since only females parasitize and, in consequence, control the target host in the field. Thus, it is desirable to increase the production of females (Bueno et al. 2009). In general, an increase in temperature was related to a decrease in sex ratio. While parasitism occurred at 25 °C, and subsequent larval development took place at several temperatures (trials 1 and 2), the higher number of females emerging at 15 °C suggests their higher cold tolerance compared with males. This might be due to larger size or larger fat body of females, a hypothesis supported by Yeargan (1980), who reported that at 15.5 °C only females emerge. Likewise, Doetzer & Foerster (2007) reported a higher occurrence of females under natural conditions during the coldest months in the off-season of soybean in southern Paraná, Brazil.

It is also important to analyze the distribution of *T. podisi* lifetime parasitism because the active time of parasitoid females might vary related to temperature (Reznik & Vaghina 2006), hosts (Reznik et al. 2001), and parasitoid species (Pratissoli & Parra 2000), and may directly influence parasitoid use in the field. For example, whether parasitoid activity is higher in the first d of life or is evenly distributed throughout adulthood is important when choosing the best parasitoid release strategy (Bueno et al. 2010) for various reasons.

The sooner the parasitoid reaches 80% of its lifetime parasitism, the better, because parasitoids would be less exposed to mortality factors under field conditions. In practice, those factors could be pesticide spraying necessary for crop management, or an abrupt change in weather conditions that may kill the egg parasitoid (Carmo et al. 2010; Denis et al. 2011). On the other hand, one of the factors that can reduce field efficiency of egg parasitoids is the lack of synchronization between the occurrence of the most susceptible stage of the target host with the period of greater adult parasitism activity (Cingolani et al. 2014). Thus, a longer *T. podisi* lifetime parasitism distribution would be preferable.

Taking into account that synchronization between parasitoid and host is an important challenge in augmentative biological control, *T. podisi* exhibited favorable traits for a prospective successful biocontrol agent. As opposed to other parasitoids such as *Trichogramma pretiosum* Riley (Hymenoptera: Trichogrammatidae), which reached over 80% lifetime parasitism (cumulative parasitism) already on the 12th, 10th, 5th, and 7th d of adulthood when exposed to 18, 20, 25, and 30 °C, respectively (Bueno et al. 2012). *Telenomus podisi* reached this parasitism level only at the 24th, 20th, 14th, and 8th d of parasitoid adulthood at 15, 20, 25, and 30 °C on *D. melacanthus* eggs, respectively. Similarly, *T. podisi* reached 80% lifetime parasitism on the 18th, 17th, 14th, and 12th d of parasitoid adulthood at 15, 20, 25, and 30 °C on *P. nigrispinus* eggs, respectively. Therefore, a much longer period is available for synchronization of the parasitoid with its target pest, reducing the chance for release failures in the field.

It is important to point out that even though temperature is considered one of the most important factors in augmentative biological control success, it is not the only factor responsible for changes in

development and survival of egg parasitoids. Other biotic and abiotic factors, such as photoperiod, relative humidity, interspecific and intraspecific competition, may interfere with biological control (Bueno et al. 2012). However, *T. podisi* generally was influenced by temperature in eggs of both *D. melacanthus* and *P. nigrispinus*. The extreme temperatures of 15 and 30 °C were unfavorable for *T. podisi* parasitism (although it still occurred) because traits were impaired at those temperatures, especially parasitism and adult longevity. In regions where those extreme temperatures may occur commonly, additional studies are necessary to investigate the possible need for increased parasitoid numbers during field releases. Even though the release of *T. podisi* in the field may negatively impact the predator *P. nigrispinus*, it is probably still safer than the use of chemical insecticides, which would be the alternative measure to *T. podisi* in the control of stink bugs.

Acknowledgments

The authors thank "Empresa Brasileira de Pesquisa Agropecuária" (Embrapa Soybean); "Coordenação de Aperfeiçoamento de Pessoal de Nível Superior" (CAPES); "Conselho Nacional de Desenvolvimento Científico e Tecnológico" (CNPq) (grants 303779/2015-2 and 402797/2016); and "Universidade Federal do Paraná" (UFPR) for funding that supported this research.

References Cited

Bianco R. 2005. Manejo de pragas do milho em plantio direto, pp. 8–17 *In* Anais IX Reunião Itinerante de Fitossanidade do Instituto Biológico. Emopi, Campinas, São Paulo, Brazil.

Bueno AF, Corrêa-Ferreira BS, Roggia S, Bianco R. 2015. Silenciosos e daninhos. Revista Cultivar Grandes Culturas 196: 25–27.

Bueno RCO, Parra JRP, Bueno AF. 2009. Biological characteristics and thermal requirements of a Brazilian strain of the parasitoid *Trichogramma pretiosum* reared on eggs of *Pseudoplusia includens* and *Anticarsia gemmatalis*. Biological Control 51: 355–361.

Bueno RCOF, Carneiro TR, Bueno AF, Pratissoli D, Fernandes OA, Vieira SS. 2010. Parasitism capacity of *Telenomus remus* Nixon (Hymenoptera: Scelionidae) on *Spodoptera frugiperda* (Smith) (Lepidoptera: Noctuidae) eggs. Brazilian Archives of Biology and Technology 53: 133–139.

Bueno RCOF, Carneiro TR, Pratissoli D, Bueno AF, Fernandes OA. 2008. Biology and thermal requirements of *Telenomus remus* reared on fall armyworm *Spodoptera frugiperda* eggs. Ciência Rural 38: 1–6.

Bueno RCOF, Parra JR, Bueno AF. 2012. *Trichogramma pretiosum* parasitism of *Pseudoplusia includens* and *Anticarsia gemmatalis* eggs at different temperatures. Biological Control 60: 154–162.

Burr IW, Foster LA. 1972. A test for equality of variances. Mimeo series, no. 282. Purdue University, West Lafayette, Indiana, USA.

Canto-Silva CR, Romanowski HP, Redaelli LR. 2005. Effect of temperature on the development and viability of *Gryon gallardoi* (Brethes) (Hymenoptera: Scelionidae) parasitizing *Spartocera dentiventris* (Berg) (Hemiptera: Coreidae) eggs. Brazilian Journal of Biology 65: 415–421.

Carmo EL, Bueno AF, Bueno RCOF. 2010. Pesticide selectivity for the insect egg parasitoid *Telenomus remus*. BioControl 55: 455–464.

Chiesa ACM, Sismeiro MNS, Pasini A, Roggia S. 2016. Tratamento de sementes para manejo do percevejo-barriga-verde na cultura de soja e milho em sucessão. Pesquisa Agropecuária Brasileira 51: 301–308.

Cingolani M, Greco F, Nancy M, Liljesthrom GG. 2014. Effect of *Telenomus podisi*, *Trissolcus urichi*, and *Trissolcus basalis* (Hymenoptera: Platygastridae) age on attack of *Piezodorus guildinii* (Hemiptera: Pentatomidae) eggs. Environmental Entomology 43: 377–383.

Cônsoli FL, Kitajima EW, Parra JRP. 1999. Ultrastructure of the natural and factitious host eggs of *Trichogramma galloi* Zucchi and *Trichogramma pretiosum* Riley (Hymenoptera: Trichogrammatidae). International Journal of Insect Morphology and Embryology 28: 211–229.

Denez MD, Bueno AF, Pasini A, Bortolotto OC, Stecca CS. 2014. Biological parameters of *Podisus nigrispinus* (Hemiptera: Pentatomidae) fed with different soybean insect pests. Annals of the Entomological Society of America 107: 967–974.

Denis D, Pierre JS, van Baaren J, van Alphen JJM. 2011. How temperature and habitat quality affect parasitoid lifetime reproductive success - a simulation study. Ecological Modelling 222: 1604–1613.

Doetzer AK, Foerster LA. 2007. Efeito da temperatura no desenvolvimento, longevidade e reprodução de *Trissolcus basalis* (Wollaston) e *Telenomus podisi* Ashmead (Himenóptera: Scelionidae). Neotropical Entomology 36: 233–242.

Godfray HCJ. 1994. Parasitoids: Behavioural and Evolutionary Ecology. Princeton University Press, Princeton, New Jersey, USA.

Hernández D, Díaz F. 1996. Efecto de la temperatura sobre el desarrollo de *Telenomus remus* Nixon (Hymenoptera: Scelionidae) parasitoide de *Spodoptera frugiperda* (Smith) (Lepidoptera: Noctuidae). Boletin de Entomologia Venezolana 11: 149–153.

Huey RB, Berrigan D. 2001. Temperature, demography, and ectotherm fitness. American Naturalist 158: 204–210.

Koppel AL, Herbert Jr DA, Kuhar TP, Kamminga K. 2009. Survey of stink bug (Hemiptera: Pentatomidae) egg parasitoids in wheat, soybean, and vegetable crops in southeast Virginia. Environmental Entomology 38: 375–379.

Margaría CB, Loiácono MS, Lanteri AA. 2009. New geographic and host records for scelionid wasps (Hymenoptera: Scelionidae) parasitoids of insect pests in South America. Zootaxa 2314: 41–49.

Medeiros MA, Shimidt FVG, Loiácono MS, Carvalho VF, Borges M. 1997. Parasitismo e predação em ovos de *Euschistus heros* (Fab.) (Heteroptera: Pentatomidae) no Distrito Federal, Brasil. Anais da Sociedade Entomológica do Brasil 26: 397–401.

Nakama PA, Foerster LA. 2001. Efeito da alternância de temperatura no desenvolvimento e emergência de *Trissolcus basalis* (Wollaston) e *Telenomus podisi* Ashmead (Hymenoptera: Scelionidae). Neotropical Entomology 30: 269–275.

Pacheco DJP, Corrêa-Ferreira BS. 2000. Parasitismo de *Telenomus podisi* Ashmead (Hymenoptera: Scelionidae) em populações de percevejos pragas da soja. Anais da Sociedade Entomológica do Brasil 29: 295–302.

Panizzi AR, Parra JRP, Santos CH, Carvalho DR. 2000. Rearing the southern green stink bug using artificial dry diet and artificial plant. Pesquisa Agropecuária Brasileira 35: 1709–1715.

Peres WAA, Corrêa-Ferreira BS. 2004. Methodology of mass multiplication of *Telenomus podisi* Ashmead and *Trissolcus basalis* (Hymenoptera: Scelionidae) on eggs of *Euschistus heros* (Hemiptera: Pentatomidae). Neotropical Entomology 33: 457–462.

Pexton JJ, Mayhew PJ. 2002. Siblicide and life-history evolution in parasitoids. Behavioral Ecology 13: 690–695.

Pratissoli D, Parra JRP. 2000. Desenvolvimento e exigências térmicas de *Trichogramma pretiosum* Riley, criados em duas traças do tomateiro. Pesquisa Agropecuária Brasileira 35: 1281–1288.

Queiroz AP, Bueno AF, Pomari-Fernandes A, Grande MLM, Bortolotto OC, Silva DM. 2017. Quality control of *Telenomus remus* (Hymenoptera: Platygastridae) reared on the factitious host *Corcyra cephalonica* (Lepidoptera: Pyralidae) for successive generations. Bulletin of Entomological Research 107: 791–798.

Reznik SY, Umarova TY, Voinovich ND. 2001. Long-term egg retention and parasitization in *Trichogramma principium* (Hymenoptera, Trichogrammatidae). Journal of Applied Entomology 125: 169–175.

Reznik SY, Vaghina NP. 2006. Temperature effects on induction of parasitization by females of *Trichogramma principium* (Hymenoptera, Trichogrammatidae). Entomological Review 86: 133–138.

SAS Institute. 2009. SAS/STAT: user's guide. Version 9.2. Cary, North Carolina, USA.

Shapiro SS, Wilk MB. 1965. An analysis of variance test for normality (complete samples). Biometrika 52: 591–611.

Smith SM. 1996. Biological control with *Trichogramma*: advances, successes, and potential of their use. Annual Review of Entomology 41: 375–406.

Torres JB, Pratissoli D, Zanuncio JC. 1997. Exigências térmicas e potencial de desenvolvimento dos parasitóides *Telenomus podisi* Ashmead e *Trissolocus brochymenae* (Ashmead) em ovos de percevejo predador *Podisus nigrispinus* (Dallas). Anais da Sociedade Entomológica do Brasil 26: 339–342.

Torres JB, Zanuncio JC, Cecon PR, Gasperazzo WL. 1996. Mortalidade de *Podisus nigrispinus* (Dallas) por parasitoides de ovos em áreas de eucalipto. Anais da Sociedade Entomológica do Brasil 25: 463–472.

van Lenteren JC, Bueno VHP. 2003. Augmentative biological control of arthropods in Latin America. Biocontrol 48: 123–139.

van Lenteren JC, Bolckmans K, Köhl J, Ravensberg WJ, Urbaneja A. 2018. Biological control using invertebrates and microorganisms: plenty of new opportunities. BioControl 63: 39–59.

Visser B, Ellers J. 2008. Lack of lipogenesis in parasitoids: a review of physiological mechanisms and evolutionary implications. Journal of Insect Physiology 54: 1315–1322.

Wilson LT, Barnett WW. 1983. Degree-days: an aid in crop and pest management. California Agriculture 37: 4–7.

Yeargan KV. 1980. Effects of temperature on development rate of *Telenomus podisi* (Hymenoptera: Scelionidae). Annals of the Entomological Society of America 73: 339–342.