

# Functional response of *Telenomus remus* (Hymenoptera: Scelionidae) to different egg densities of *Spodoptera frugiperda* (Lepidoptera: Noctuidae)

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## ABSTRACT

The parasitoid *Telenomus remus* Nixon (Hymenoptera: Scelionidae) is a natural enemy of lepidopteran pests, and several studies have been conducted to evaluate its efficiency on eggs of *Spodoptera frugiperda* (J. E. Smith) (Lepidoptera: Noctuidae). Functional response is an important parameter for assessing the potential of parasitoids. The objective of this work was to evaluate the harvesting behavior and the functional response of *T. remus* on eggs of *S. frugiperda*. We conducted the experiment in an acclimatized chamber at  $25 \pm 2^\circ\text{C}$ , relative humidity  $70\% \pm 10\%$  and photophase of 12 hours. For harvesting behavior, we observed that a female takes 2.4 minutes, from the placement to the first cleaning, which is 44.7 seconds, to parasitize one egg. The value of the quadratic coefficient was negative ( $P_1 < 0.0005$ ), so it can be assumed that the functional response of *T. remus* to eggs of *S. frugiperda* was type III. The results of attack rate ( $a'$ ), handling time ( $T_h$ ), and number of eggs parasitized at 24 hours by *T. remus* were, respectively, 0.00664 and  $0.5649 \cdot \text{h}^{-1}$ . The sex ratio of the parasitoid was influenced by host densities, with a ratio of 0.5 at 10 and 30 egg densities, but values were not differentiated from the density of 15 eggs. Our results can be helpful for developing field studies, enhancing the knowledge and possibilities for integrated pest management.

**Keywords:** parasitoid; biological control; behavior.

Biological control is one of the pillars of integrated pest management, and in some cases, it is as effective as the use of insecticides. For example, the mass rearing of natural enemies and subsequent flood releases tend to reduce the damage caused by pests to crops, preventing their population from evolving quickly and with minimal damage to the environment. Agronomists and farmers can visualize this strategy as they consider the possible replacement of agrochemicals with an equally effective action plan (Parra et al., 2002).

Parasitoids are one of the main groups of biological control agents, being used on a large scale in biological control programs. The adoption of this strategy occurs mainly due to the need to reduce insecticide residues in food and in the environment, resulting from the improper use of such products. Among the parasitoid species in which they stand out in Brazil, *Telenomus remus* Nixon (Hymenoptera: Scelionidae) is one of the main ones, since this insect parasitizes eggs of important Lepidoptera, such as the fall armyworm *Spodoptera frugiperda* (J. E. Smith, 1789) (Lepidoptera: Noctuidae) (Parra; Coelho Jr., 2019).

The fall armyworm, a cosmopolitan and polyphagous pest, is one of the main pests in Brazil, reportedly resistant to various forms of control, especially Bt transgenic plants (Huang, 2021). The control of this pest is often carried out with chemical products with little monitoring criteria, which can be harmful to numerous non-target organisms, such as other arthropods, fish, birds, and mammals. In addition, the successive use of insecticides can cause the pest to resist the mechanisms of action employed, especially if they are not properly managed (Berg; Plessis, 2022).

Using *T. remus* as a biological control agent is advantageous due to its high dispersion and search behavior and the ability to parasitize the deep layers of the egg masses of *S. frugiperda* (Bueno et al., 2008; Figueiredo et al., 2002). The

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relationship between the consumption rate (parasitism in this case) and the host density is called the functional response (Fernández-Arhex; Corley, 2003). A natural enemy's potential is evaluated using the functional response for various situations, such as a lack or excess of prey, in a natural, protected, or directed environment (laboratory). Fernández-Arhex and Corley (2003) indicate that most parasitoids used in biological control have a type-II response, but that there is no relationship between the type of functional response and success in biological control. However, this information can infer the role of a natural enemy in a system and its relationship with the pest of interest. Therefore, it is of paramount importance to determine the behavioral aspects and functional response of the parasitoid *T. remus* on *S. frugiperda* under laboratory conditions. Thus, the objective of this study was to evaluate the functional response of *T. remus* on different densities of *S. frugiperda* eggs, in addition to describing its behavior during parasitism.

The specimens of the parasitoid *T. remus* and the host *S. frugiperda* were kept under controlled temperature conditions of  $25 \pm 2^\circ\text{C}$ , relative humidity of  $70 \pm 10\%$  and photophase of 12 hours. First, with the aid of a video coupled to a player and monitor, the foraging and oviposition behavior of the parasitoid on host eggs was recorded, and the analysis subsequently was carried out. For this, an egg mass of approximately 100 eggs was deposited in a Petri dish (90 mm in diameter), then the female *T. remus* was placed in the dish, at which time the foraging behavior analyses began. The interaction between *T. remus* and the egg densities of *S. frugiperda* was evaluated using flat-bottomed test tubes (8.5 cm high  $\times$  2.4 cm in diameter) with five, 10, 15, 20, 25, 30, and 40 eggs, and a female of the parasitoid. The parasitoids were kept in contact with the eggs for 24 hours, and the evaluation of the number of parasitized eggs was carried out 14 days later. The experimental design was completely randomized, with three replicates.

The functional response was estimated in two steps. The first step refers to the type of functional response, which was determined through logistic regression of the proportion of parasitized *S. frugiperda* eggs ( $N_a$ ) by *T. remus* as a function of the original egg densities per parasitoid, the initial number of eggs ( $N_o$ ), using the Proc CATMOD (SAS Institute), as follows:  $N_a/N_o = \exp(P_o + P_1N_o + P_2N_o^2 + P_3N_o^3) / 1 + \exp(P_o + P_1N_o + P_2N_o^2 + P_3N_o^3)$ .  $P_o$ ,  $P_1$ ,  $P_2$ , and  $P_3$  are the constant, linear, quadratic, and cubic coefficients, respectively. Initially, the cubic model was tested due to its ability to capture all possible variations of the functional response curves (Juliano, 2001).

The linear coefficient of the equation ( $P_1$ ) estimated from the proportion of preyed eggs by prey density was used to determine the type of functional response. The linear coefficient, when not significant, indicates a type-I functional response; when it is significant and negative, it indicates a type-II functional response; and when positive, a type-III functional response. There was no host replacement during the experiment, so the type of functional response was determined from the random equation proposed by Rogers (1972) (Eq. 1):

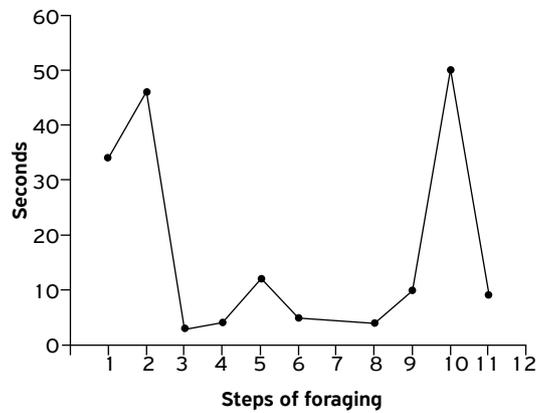
$$N_a = N_o \{1 - \exp [a(T_h N_a - T)]\} \quad (1)$$

Where  $N_a$ : Number of parasitized eggs;  $N_o$ : number of eggs supplied;  $a$ : attack rate;  $T_h$ : handling time;  $T$ : predator exposure time to prey (24 h).

The second step was to determine the parameters  $T_h$  and  $a$  of the functional response, estimated by nonlinear regression using the least squares method, according to the program written by Juliano (2001). The experimental design was completely randomized, with three replicates per host density. The influence of *S. frugiperda* egg density on the sex ratio of *T. remus* and the influence of parasitism on the number of hatched *S. frugiperda* larvae were compared by Tukey's test ( $p < 0.05$ ).

When the female is introduced into the arena, she performs some steps during foraging (Fig. 1). First, the female flaps her wings to move in the direction of the egg mass, and 34 seconds are used to locate the host. Soon after, the female walks on the mass of eggs, drumming her antennae for 46 seconds, followed by oviposition for 3 seconds. Subsequently, the female spent 4 seconds to locate the next egg, and, after 12 seconds drumming over the egg, spent about 5 seconds to oviposit, followed by another 4 seconds to locate the next egg. On the third oviposition, the female spent about 10 seconds drumming and 50 seconds oviposition. At the end of the third oviposition, the female took 5, 2, and 2 seconds to clean the antennae, legs, and ovipositor, respectively.

In addition, we observed that the insect cleaned its appendages every three ovipositions. Thus, a total cycle of 2.95 minutes was established considering the time of egg search, drumming, three ovipositions, and cleaning. If we consider the total time spent per egg, we obtain the total of 44.7 seconds per egg, a result that is very close to those obtained in previous studies (Schwartz; Gerling, 1974; Cave et al., 1987; Carneiro et al., 2010).



**Figure 1.** Foraging behavior of *Telenomus remus* on *Spodoptera frugiperda* eggs. Each step was measured in seconds according to the steps observed.

Source: Elaborated by the authors.

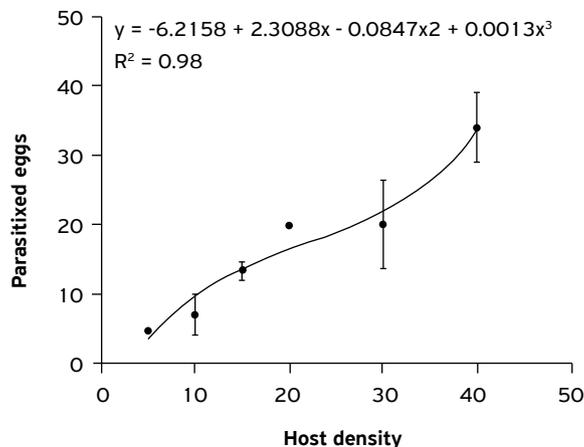
According to the result shown by the estimated parameters, the value of the quadratic coefficient was negative ( $p < 0.0005$ ). Thus, it can be assumed that the functional response of *T. remus* to the eggs of *S. frugiperda* was type III (Table 1). The functional response of the parasitoid is shown in Fig. 2. The functional response type III of *T. remus* on *S. frugiperda* means that there is an increase in the proportion of parasitism until the inflection of the sigmoid curve, followed by a reduction in the proportion of parasitism. The variable used in our study was egg density, but other factors can also influence the type of functional response. There is a divergence in the results obtained by Carneiro et al. (2010), in which they observed a functional response of type II, but we attribute this divergence to the different egg densities used in the two studies.

Different hosts may affect populations of *Trissolcus basalis* Thompson differently regarding the number of females that emerged, but without altering their functional response (Allahyari et al., 2004). In addition, four species of *Trissolcus* parasitizing pentatomid eggs showed a type-III functional response, having an interesting potential for implementation in biological control programs (Laumann et al., 2008). Even so, other authors are more cautious, preferring not to consider the functional response alone to determine the potential of species for biological control programs (Lester; Harmsen, 2002).

**Table 1.** Results of the logistic regression analysis of the proportion of *Spodoptera frugiperda* eggs parasitized by *Telenomus remus* females.

Parameters	Estimative	EP	$\chi^2$	P-value
Intercept	-1.3774	1.3040	1.12	0.2908
Linear	0.6515	0.2139	9.28	0.0023
Quadratic	-0.0350	0.00996	12.37	0.0004
Cubic	0.000528	0.000138	14.62	0.0001

Source: Elaborated by the authors.



**Figure 2.** Average number of *Spodoptera frugiperda* eggs parasitized by *Telenomus remus* females at different densities for 24 hours.

Source: Elaborated by the authors.

The results of attack rate ( $a'$ ), handling time ( $T_h$ ), and number of eggs parasitized in 24 hours by *T. remus* were 0.00664 and 0.5649·h<sup>-1</sup>, respectively (Table 2). No emergence of *T. remus* was observed at the density of 20 eggs of *S. frugiperda*. The sex ratio of the parasitoid was influenced by the host densities, with a ratio value = 0.5 observed in densities of 10 and 30 eggs, whose values were not different from the density of 15 eggs ( $F = 4.74$ ;  $p < 0.05$ ).

Regarding the number of hatched larvae, no difference was observed between the egg densities used ( $F = 0.917$ ;  $p = 0.502$ ) (Table 3). In this study, the functional response of *T. remus* parasitizing eggs for 24 hours was evaluated, but it is known that the time of exposure of the host to the parasitoid can influence the result (Xiang et al., 2020), so for a more complete knowledge, other parasitism periods should be evaluated. Another important detail is that, in previous studies, Morales et al. (2001) found that *T. remus* stabilizes its parasitism at the density of 150 host eggs, although their results are questioned by Carneiro et al. (2010).

**Table 2.** Attack rate ( $a'$ ) and handling time ( $T_h$ ) of *Telenomus remus* hosted by *Spodoptera frugiperda* eggs at different densities for 24 hours.

Parameters	Estimative ( $\pm$ EP)	Confidence interval (95%)
$a'$	0.00664 $\pm$ 0.00373	-0.00128–0.0146
$T_h$	0.5649 $\pm$ 0.5649	0.1972–0.9326

Source: Elaborated by the authors.

**Table 3.** Sex ratio of *Telenomus remus* and number of hatched larvae of *Spodoptera frugiperda* at different densities for 24 hours.

Density	Sex ratio <sup>1</sup>	Hatched larvae <sup>ns</sup>
5	0.8 $\pm$ 0.14 a	0.0 $\pm$ 0.00
10	0.5 $\pm$ 0.25 ab	0.0 $\pm$ 0.00
15	0.2 $\pm$ 0.24 ab	2.0 $\pm$ 1.15
20	0.0 $\pm$ 0.00 b	0.0 $\pm$ 0.00
30	0.5 $\pm$ 0.04 ab	1.7 $\pm$ 0.96
40	0.9 $\pm$ 0.03 a	5.7 $\pm$ 3.27

<sup>1</sup>Values followed by the same lowercase letter in the column are not different by Tukey's test at 5% probability; <sup>ns</sup>no statistical difference. Source: Elaborated by the authors.

Basic studies of interactions and behaviors between different trophic levels, such as functional response, help us to know the dynamics of biological control agents and their possible impact on the environment. Such studies can be useful in decision-making in integrated pest management programs. With the support of the data obtained in this study, field work, in which there is the interaction of more variables, should be developed to help decision-making in the use of biological control.

## AUTHORS' CONTRIBUTIONS

**Conceptualization:** Pinto, C.P.G.; **Resources:** Pinto, C.P.G.; **Methodology:** Pinto, C.P.G.; Romanello, N.; **Investigation:** Pinto, C.P.G.; Romanello, N.; **Data curation:** Pinto, C.P.G.; Romanello, N.; **Formal analysis:** Pinto, C.P.G.; Romanello, N.; **Visualization:** Pinto, C.P.G.; **Supervision:** Pinto, C.P.G.; **Writing – original draft:** Pinto, C.P.G.; Romanello, N.; **Writing – review and editing:** Pinto, C.P.G.; Romanello, N.; **Final approval:** Pinto, C.P.G.

## AVAILABILITY OF DATA AND MATERIAL

The datasets generated and/or analyzed during the current study are available from the corresponding author on reasonable request.

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## CONFLICTS OF INTEREST

All authors declare that they have no conflict of interest.

## ETHICAL APPROVAL

Not applicable.

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