Predation and reproductive output of the ladybird beetle *Stethorus tridens* preying on tomato red spider mite *Tetranychus evansi*

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Abstract Predatory behaviour and reproductive output of the ladybird beetle *Stethorus tridens* Gordon as function of the tomato red spider mite (TRSM), *Tetranychus evansi* Baker & Pritchard, densities was investigated in the laboratory. Adult female of *S. tridens* were isolated in cylindrical plastic arenas, containing a leaf disc of *Solanum americanum* Mill. with 5, 20, 40, 60, 80 or 100 *T. evansi* nymphs. The number of prey consumed and eggs laid were evaluated daily for ten consecutive days, starting at the oviposition. Oviposition of *S. tridens* was positively correlated with prey consumption and lower threshold prey consumption for *S. tridens* laying eggs was 16.3 mites per day. The instantaneous rate of attack (ca. discovery area) and the handling time were 0.0062 h^{-1} and 0.83 h, and 0.00254 h^{-1} and 0.78 h, respectively, for predators at the 1st- and 10th-oviposition day. The predator exhibited a type II functional response at 1st- and 10th-oviposition day with a maximum consumption per predator of 33 *T. evansi* nymphs per day at the highest prey density. The ladybird beetle *S. tridens* is often collected associated with red spider mite colonies on solanaceous wild plants and the results suggest the potential of this ladybird beetle to control *T. evansi* in tomatoes crops.

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Introduction

The tomato red spider mite (TRSM), *Tetranychus evansi* Baker & Pritchard, was originally described from Mauritius Island (Baker and Pritchard 1960). It was reported for the first time in Brazil, in the State of Bahia, as *Tetranychus marianae* McGregor (Silva 1954). Later it was reported from California (USA) causing significant yield losses (Oatman et al. 1967). The TRSM is considered a pest of various cultivated Solanaceae plants, frequently reaching high population levels (Jeppson et al. 1975). In the 1970s and 1980s, when tomato production was one of the most important activities in the irrigated areas around

Petrolina, State of Pernambuco, Brazil, TRSM was considered one of the most important pests of processing tomatoes in that region (Ramalho and Flechtmann 1979).

In the last 25 years, TRSM has become an important tomato pest in southern and eastern Africa, significantly reducing yield (Knapp et al. 2003). More recently, it was also found in Portugal (Ferreira and Carmona 1995), Spain (Ferragut and Escudero 1999) and France (Migeon 2005).

Several predatory mites have been reported in association with T. evansi but none has been identified as effective against the pest in laboratory studies. For instance, Moraes and Lima (1983) studied the suitability of T. evansi as prey for the Phytoseiidae mite Euseius concordis (Chant). Moraes and McMurtry (1985) evaluated the potential of eight Phytoseiidae species in the genera Amblyseius, Phytoseiulus, Phytoseius and Typhlodromus as predators of T. evansi. Rosa et al. (2005) studied the reproductive capacity of predatory mites of the families Ascidae, Phytoseiidae and Tydeidae on the same pest. Survival, predation capacity, and oviposition rate always were very low in those studies. However, Britto et al. (2005) studying the population dynamics of T. evansi on Solanum americanum Mill. (Solanaceae) in Recife, Pernambuco, Brazil, reported predation of TRSM by the ladybird beetle Stethorus tridens Gordon.

In several occasions during 2004–2006 the senior author observed the natural occurrence of S. tridens on solanaceous plants preying upon T. evansi. Acarophagous Coccinellidae can be effective in regulating populations of tetranychid mites (McMurtry and Johnson 1966; Olkowski et al. 1990). Stethorus species have been used commercially for the control of tetranychid mites in greenhouses: Stethorus punctum LeConte against Tetranychus urticae Koch (Houck 1991) and Stethorus bifidus (Kapur) against Tetranychus lintearius (Dufour) (Peterson et al. 2000). Therefore, S. tridens might be a potential control agent of T. evansi.

Prospective biological agents can be evaluated through functional and numerical responses. Functional response measures the predation rate as a function of prey density allowing estimation of the attack rate and handling time of the biocontrol agent. The most important aspects of modeling the functional response are the shape and parameters of the response curve. This is based on two basic parameters: the handling time of the prey (T_h), consisting of the time required for finding, killing and ingesting the prey, and the attack rate (a) that represents the search effectiveness (Hassell 1978; Casas et al. 1993). On the other hand, the numerical response represents the effect of changes in prey density on the reproduction rate of the predator (Holling 1959). The objective of this study was to investigate the functional and reproductive responses of S. tridens when preying on T. evansi.

Materials and methods

Environmental conditions, prey and predator

The experiments were carried out in the laboratory of Agricultural Acarology of "Universidade Federal Rural de Pernambuco" (UFRPE), Recife, at 25°C, 70 ± 10% R.H. and 12:12 h (L:D) photoperiod.

Adults and nymphs of the red spider mite T. evansi, were collected from S. americanum plants naturally occurring in the Experiment Area of the Agronomy Department of the UFRPE (Recife, PE). In the laboratory, T. evansi colony was maintained on S. americanum plants. Suitable host plants to sustain the colony were cultivated in open-sided greenhouse using pots (20 cm diameter × 20 cm height) filled with a mixture of soil and humus (4:1).

Adults of S. tridens were collected from plants of S. americanum infested by T. evansi on the Experiment Area of the Agronomy Department of the UFRPE (Recife, PE). They were placed in cages (50 cm in diameter × 90 cm high) covered with organdy fabric containing two or three plants of S. americanum infested with T. evansi were replaced every 15 days.

Pupae of S. tridens were collected from the rearing cages and maintained isolated until adult emergence in a transparent cylindrical plastic container (2.5 cm diameter × 2.5 cm high) with a circular opening in the lid fixed with organdy fabric. Newly molted adults were paired for 10 days. At the prevailing temperature (25°C) this period corresponds to the mating and the preoviposition periods (Fiaboe et al. 2005). During preoviposition period, S. tridens were fed a mixture of developing stages of TRSM available on heavily infested leaves of S. americanum. The leaf petiole was inserted into a small glass

tube containing water and sealed with paraffin to maintain the turgidity of the leaves. After the 10 days preoviposition period, each female was isolated in arena that consisted of transparent cylindrical plastic containers of 2.5 cm (ca. bottom diameter). The bottom of the arena was covered with filter paper discs of the same diameter soaked in distilled water and topped by a leaf disc of *S. americanum* of 2.5 cm diameter each.

Six densities of *T. evansi* nymphs (mix population of proto- and deuto-nymphs) were tested (5, 20, 40, 60, 80 and 100 nymphs per arena). Tests with the lower densities were replicated 14 times while those with the last two higher densities were replicated 10 times.

The number of prey consumed was evaluated every 24 h, when consumed prey and those individuals molted to adult stage were replaced. The new nymphs used to replace the predator consumption and the *T. evansi* adults for each established prey density in the study were originated from the colony maintained in the laboratory. The new nymphs were collected from heavily infested *S. americanum* leaves and they were individually transferred to the arena in according to the number required to match the prey density.

Statistical analysis

The functional response was estimated in two steps. The first step consisted of determining the general shape of the functional response curve based on logistic regression between the mites consumed per density using the CADMOD procedure of SAS (SAS Institute 1999–2001) The cubic model was initially tested due to its capacity to detect most possible

functional response graph variations (Juliano 1993). When the higher level term of the regression model was not significant, the model was reduced to the lower level term until significance was obtained. The sign of the linear coefficient of the equation obtained was used to determine the type of functional response: nonsignificant linear coefficient indicates functional response type I, significantly negative coefficient indicates functional response type II and significantly positive coefficient indicates functional response type III. The second step was to determine the T_h (handling time) and a' (attack rate) parameters of the functional response. These parameters were estimated with non-linear least square regression (PROC NLIN, SAS Institute 1999–2001), as described by Juliano (1993).

Oviposition response to prey densities was estimated by linear regression of the number of eggs laid per female on prey density using the PROC REG of SAS (SAS Institute 1999–2001).

Results and discussion

Step one of analysis revealed a type II functional response for adult female of *S. tridens* on the 1st and 10th oviposition days (Table 1 and Fig. 1). The model, however, required initial estimation searching rate and handling time. For functional response type II models the estimated searching rate is a linear function of original prey density ($a' = bN$) and the initial handling time is the ratio between the number of prey consumed and the duration of the experiment (24 h) for the highest prey density ($T_h = 24$ h$/N_a$), which accounts for the highest probability of encountering prey by the predator (Juliano 1993). Type II

Table 1 Estimated parameters of the logistic regression between the proportion of *T. evansi* nymphs consumed by ovipositing female of *S. tridens* at initial prey densities between 5 and 100 per arena on the 1st and 10th oviposition day

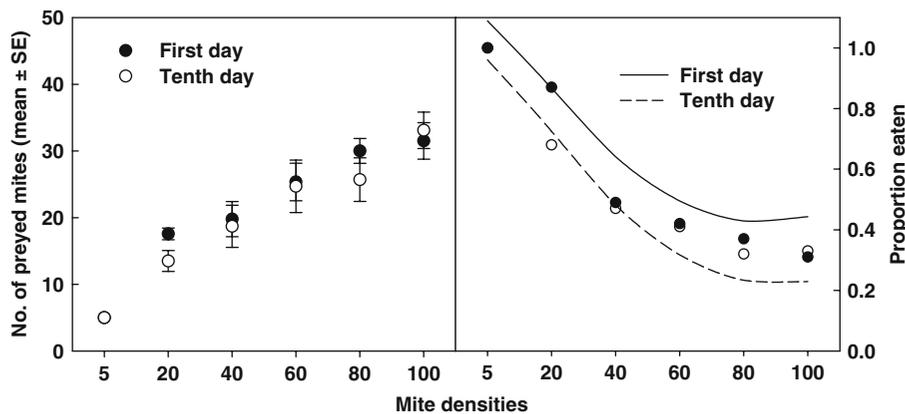| Oviposition day (age within reproductive period)	Parameter	Value (±SE)	df	χ^2	P
First (0–24 h)	Intercept	5.93 ± 0.58	1	104.40	<0.0001
	Linear	−0.27 ± 0.03	1	71.43	<0.0001
	Square	0.003 ± 0.0005	1	49.11	<0.0001
	Cubic	−2.0E-5 ± 2.8E-6	1	38.92	<0.0001
Tenth (216–240 h)	Intercept	2.48 ± 0.41	1	36.62	<0.0001
	Linear	−0.103 ± 0.024	1	17.67	<0.0001
	Square	0.0011 ± 0.0004	1	7.22	<0.0001
	Cubic	−4.7E-6 ± 2.4E-6	1	3.92	0.0478

Fig. 1 Left: mean number of *Tetranychus evansi* consumed by ovipositing females of *Stethorus tridens* at different prey densities on the 1st and 10th oviposition day. Right: representation of the type II functional response by the proportion of mite eaten as function of prey availability on the 1st and 10th oviposition day

functional response was similar reported for *S. punctum* (Hull et al. 1977), *Stethorus vagans* (Blackburn) (Ullah 2000) and *Stethorus japonicus* Kamiya (Gotoh et al. 2004), all preying on *T. urticae*.

The proportions of *T. evansi* consumed by *S. tridens* were similar at the 1st and 10th oviposition day according to 95% confidence intervals mean overlapping (Fig. 1). The response was characterized by a reduction on the proportion of prey consumed with increasing prey density, reaching a minimum consumption of ca. 0.3 of the prey at the highest density (100 *T. evansi* nymphs). Decreasing on proportion of prey consume with increasing prey density is common for arthropods predator according to Holling (1961) as a result of satiation level and handling time required for each prey item consumed.

For each density, predation varied little between the 1st and 10th oviposition day (Fig. 1). At density of five *T. evansi*, all prey were consumed and only 50% of the beetles survived to the end of the experiment, what suggests that prey density was below the level necessary for survival. At prey density of 20, the proportion of prey consumed was 0.87 and 0.68 on the 1st and 10th oviposition day, respectively (Fig. 1). According to Houck (1991), the feeding behaviour of *Stethorus* varies with prey density and the results found agree with this statement. At higher prey densities, the predators spend less time searching the prey resulting in higher prey consumption than in lower densities but predators require time to attack the prey. High prey density in this study could have interfered with the predation

process in two different ways: (a) positively, by inducing abandon of prey not completely consumed or (b) negatively, due to the interference caused by the increased numbers of prey running into the arena and around the predators.

No significant difference was found for handling time but the searching rate was 2.4 higher on the 1st day than on the 10th day (Table 2). This indicates that the predator was more effective in prey searching on the tenth day suggesting that captures improves as a function of experience. This agrees with the results of Santa-Cecilia et al. (2001). The authors working with *Cycloneda sanguinea* (L.) found that search potential was improved with increasing age within the reproductive period.

Oviposition of *S. tridens* females was observed when 20 prey items were available but not at 5 prey density. This suggests that densities of red spider mite below 20 nymphs characterize a situation of prey scarcity for *S. tridens*. Prey densities higher than 20,

Table 2 Values (Mean ± 95% confidence intervals) of searching rate and handling time for ovipositing females of *Stethorus tridens* feeding on *Tetranychus evansi* nymphs on the 1st and 10th oviposition days

Oviposition day	Searching rate (a)	Handling time (T_h)
First (0–24 h)	0.0062 ± 0.00254 a*	0.83 ± 0.04 a
Tenth (216–240 h)	0.00254 ± 0.0009 b	0.78 ± 0.06 a

* Means ± SE followed by the same letter in the column are not significantly different, 95% confidence intervals
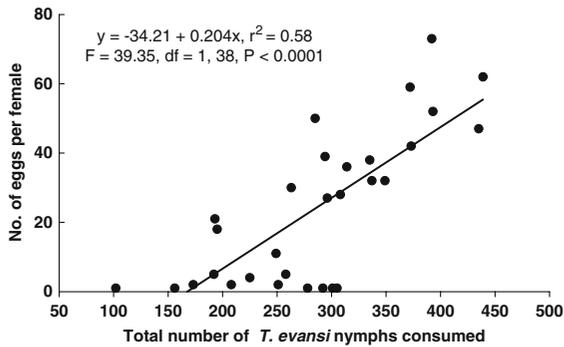
Fig. 2 Oviposition of *Stethorus tridens* as function of the *Tetranychus evansi* deuto-nymphs consumed

S. tridens oviposition varied from 1 to 73 eggs per female at the same prey density (data not show). Based on the oviposition response model (Fig. 2), when $y = 0$, *S. tridens* requires at least 16.3 prey daily to reproduce. Absence of reproduction under prey scarcity condition is common among predators when the energy goes to survival (McNamara and Buchanan 2005).

A significant correlation was observed between the number of eggs laid by the predator and the number of prey consumed at prey densities above 20 ($F_{1,28} = 39.35$; $P < 0.0001$) (Fig. 2). The average fecundity over 10 days was 11.5, 20.8, 30.4, 25.2 and 30.7 eggs per female at prey densities of 20, 40, 60, 80 and 100, respectively. Due to the high variation no significant differences were found between densities of 20 to 100 *T. evansi* nymphs ($F_{1,28} = 1.79$; $P = 0.1915$).

The results show that *S. tridens* females increase predation rate as function of *T. evansi* nymph densities and exhibit successful reproduction feeding on this pest. This fact, together with handling time and attack rate indicates that *S. tridens* is a promising predator of *T. evansi*. However, the contribution of *S. tridens* to control *T. evansi* in tomato fields requires further studies on the number of predators needed and the timing of release of the predator in the field conditions.

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