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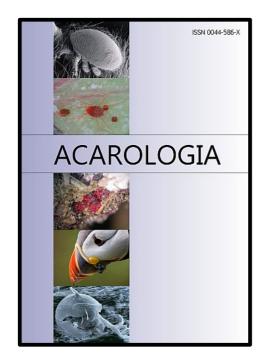
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## DOES THE ABILITY OF *BLATTISOCIUS* SPECIES TO PREY ON MITES AND INSECTS VARY ACCORDING TO THE RELATIVE LENGTH OF THE CHELICERAL DIGITS?

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ABSTRACT — Species of *Blattisocius* Keegan (Acari: Blattisociidae) are commonly found in storage facilities, seemingly feeding on mites or insects. Two species of this genus, *Blattisocius keegani* Fox and *Blattisocius everti* Britto, Lopes and Moraes, were recently found coexisting in a sample of a commercial dog food in southern Brazil. Although very similar, the species have rather different cheliceral structures, the first with the fixed digit distinctly shorter than the movable digit and the second with both digits of similar lengths. It was hypothesized that they coexisted because of their different feeding habits, the first assumed to perform better on insect prey and the second, on mite prey. A comparative study was conducted in the laboratory offering a mixture of all postembryonic stages of a mite and eggs of three insect species. *Blattisocius everti* had a better performance on the mite *Thyreophagus* sp. than *B. keegani*, but both species had about the same predation rate on *Thyreophagus* sp., and very low ovipositon rates on insects. The results corroborate the hypothesis of a better performance of *Blattisocius* with cheliceral digits of similar lengths (*B. everti*) on mite prey, but did not indicate that the possession of a short fixed digit is sufficient for the predator to feed on insect eggs. *Blattisocius everti* was shown to be able to develop and reproduce when offered *Thyreophagus* sp. as prey, with calculated  $R_0$  and  $r_m$  of 16.95 and 0.13, respectively, at  $25.0 \pm 0.5$  °C,  $80 \pm 5$  % of relative humidity and in the dark. The biotic potential of *B. everti* was comparable to what has been reported in previous work for other *Blattisocius* species offered acarid mites as prey.

KEYWORDS — biological control; biology; morphology; predatory mites; stored food

#### Introduction

Mites and insects are commonly found in storage facilities, seemingly feeding on stored products or on other organisms. In Brazil, the most harmful insects in storage are *Rhyzopertha dominica* (Fabricius) (Coleoptera: Bostrychidae), *Sitophilus* 

oryzae (Linnaeus) and Sitophilus zeamais Motschulsky (Coleoptera: Curculionidae) (Lorini, 2002). Also harmful are the insects Ephestia kuehniella Zeller (Lepidoptera: Pyralidae), Zabrotes subfasciatus (Bohemann) and Lasioderma serricorne (Fabricius) (Coleoptera: Bruchidae and Anobiidae, respectively) (Gallo et al., 2002), as well as sev-

eral mite species, especially *Tyrophagus putrescentiae* (Schrank) (Acaridae) (Flechtmann and Zem, 2002; Moraes and Flechtmann, 2008).

Recently, a new acarid mite, *Thyreophagus* n.sp., was found in bird feed in São Paulo state, southern Brazil (Camargo *et al.*, 2012). The importance of this species is unknown, but it is considered potentially important, given that a species of the same genus, *Thyreophagus entomophagus* (Laboulbène), has been mentioned as the causal agent of anaphylaxis in human beings (Blanco *et al.*, 1997).

Fifteen mite species have been classified in *Blattisocius* Keegan Blattisociidae). Species of this genus are often found in storage facilities (Hughes, 1976; Haines, 1978; Gerson *et al.*, 2003; Britto *et al.*, 2012). A synthesis of the studies evaluating the possible use of *Blattisocius* species for the control of different insect and mite species was presented by Thomas *et al.* (2011). Most of the studies refer to *Blattisocius dentriticus* Berlese, *Blattisocius keegani* Fox and *Blattisocius tarsalis* Berlese.

Specimens of *Blattisocius* were collected in July 2010 from a commercial dog food in Charqueada, São Paulo state, in association with *T. putrescentiae*. Morphological examination indicated that two species were represented (Britto *et al.*, 2012), *B. keegani* and a new species then described as *Blattisocius everti* Britto, Lopes and Moraes. Despite their considerable morphological similarity, their co-existence led Britto *et al.* (2012) to suggest that their different cheliceral structures could reduce competition between them; the former species has fixed cheliceral digit much shorter than movable digit, while the latter has digits of similar lengths.

The objectives of this study were to evaluate that hypothesis and to calculate life table parameters of *B. everti* when offered as food the prey on which it had the best performance in the initial phase of this work.

#### MATERIALS AND METHODS

A comparison of the chelicerae of the *Blattisocius* species considered in this study was done based on an examination done under phase contrast and interference contrast microscopes. For such, surplus

numbers of adult females taken from the respective colonies were mounted in Hoyer's medium on microscope slides. The slides were first checked to select the best specimens for the examination, whose chelicerae were positioned sidewise. Measurements were done using a graded eye-piece; illustrations were done using a drawing tube attached to the phase contrast microscope, while photographs were taken under the interference contrast microscope.

Mites were taken from stock colonies maintained at the Acarology Laboratory of Escola Superior de Agricultura Luiz de Queiroz, Universidade de São Paulo, Piracicaba, São Paulo state, Brazil for about two years. During that period, they were continuously fed with a mixture of all developmental stages of *Thyreophagus* sp. offered on a commercial dog food.

Experimental units consisted of plastic dishes (2.6 cm in diameter x 1.0 cm high) whose bottom was lined with a layer of a solidified paste consisting of a mixture of activated charcoal and gypsum (1v:9v) (Abbatiello, 1965). The layer was maintained humid by daily additions of distilled water. The units were kept in a rearing chamber at 25.0  $\pm$  0.5 °C, 80  $\pm$  5 % relative humidity and in the dark.

An initial test was conducted to determine the ability of B. keegani and B. everti to attack, oviposit and survive when offered separately a mite and three insect species as food sources. Treatments consisted of the exposure of the following amount of each organism to one adult female of the predator within each experimental unit: 25 nymphs of Thyreophagus sp., 25 eggs of E. kuehniella, 20 eggs of L. serricorne or 20 bean seeds containing an undetermined (but surplus) number of eggs of Z. subfasciatus. Those amounts were determined by preliminary evaluations of the daily attack rates of each predator. The test was initiated with 30 specimens of each predator, taken at random from the respective stock colonies. To prevent them from escaping, each unit was sealed with a piece of self-adhesive PVC film (Magipack®). The number of attacked prey (not evaluated for Z. subfaciatus), the number of eggs laid and survivorship were evaluated once a day for eleven consecutive days. Attacked prey

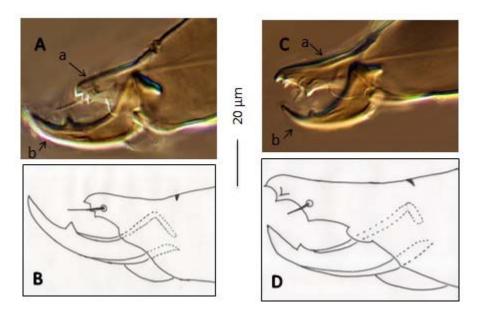


FIGURE 1: Chelicerae of *Blattisocius keegani* A – photo; B – line drawing and *Blattisocius everti* C – photo; D – line drawing; a – fixed digit; b – movable digit; bar = 20 µm.

were replaced and eggs laid were removed from the units daily. Eggs laid on the first day were not considered in the analysis, to exclude the effect of previous feeding of the predators.

A fertility life table was then constructed for *B. everti*, using as food source the species onto which the predator had best performed in the first test. Fifty adult females of *B. everti* were transferred from the stock colony to a new rearing unit containing a surplus number of a mixture of all developmental stages of *Thyreophagus* sp. as food. The unit was examined every two hours to collect the eggs laid by the predators, transferring one egg to each of 45 new experimental units.

The new units were examined every 12 hours to determine the duration of each immature stage. Immediately after reaching adulthood, a male randomly taken from the stock colony was transferred to each unit containing a female. Dead males were replaced by new males taken from the stock colony. Units containing adults were examined once a day to determine oviposition and survivorship. At this occasion, eggs laid were grouped in a single rearing unit, where predators were kept until reaching the adult stage, to determine sex ratio.

Rates of predation, oviposition, survivorship and mean durations of developmental stages were compared by the non-parametric Kruskal-Wallis tests, using R statistic program version 2.10.0 (2010). Life table parameters were calculated using the Jacknife method run in SAS program (Maia *et al.*, 2000).

#### RESULTS

Figure 1 shows the structure of the chelicerae of the *Blattisocius* species considered in the study. Based on five specimens of each species, the movable and the fixed cheliceral digits are respectively 30-32 and 16-18  $\mu$ m long in *B. keegani* and 27-29 and 26-28  $\mu$ m long in *B. everti*. In both species, the movable digit bears three teeth and the fixed digit bears one tooth, in addition to the apical tooth.

#### Predator performance on potential food sources

In relation to predation, *B. keegani* and *B. everti* did not differ significantly between themselves on *Thyreophagus* sp., *E. kuehniella* or *L. serricorne* (Table 1). Oviposition rates of *B. everti* were significantly higher on those food sources, but the difference be-

TABLE 1: Predation (number of specimens attacked  $\pm$  standard deviation), oviposition (number of eggs laid  $\pm$  standard deviation) and survivorship rates of *Blattisocius keegani* and *Blattisocius everti* in a period of 10 days when offered different organisms as prey at 25.0  $\pm$  0.5 °C, 80  $\pm$  5 % of relative humidity and in the dark.

Species offered as prey	Blattisocius keegani	Blattisocius everti		
Predation				
Thyreophagus sp.	$145.9 \pm 74.6 \text{ A}^*$	$169.1 \pm 42.4 \text{ A}$		
Ephestia kuehniella	$7.0 \pm 4.4 \text{ A}$	$5.6 \pm 4.1 \text{ A}$		
Lasioderma serricorne	$8.0 \pm 6.0 \text{ A}$	$9.4 \pm 5.7 \text{ A}$		
Oviposition				
Thyreophagus sp.	$4.0 \pm 3.2 \text{ c A}$	14.5 ± 9.1 c B		
Ephestia kuehniella	$1.0 \pm 1.3 \text{ b A}$	$2.9 \pm 3.4 \text{ b B}$		
Lasioderma serricorne	$0.6 \pm 1.3 \text{ ab A}$	1.7 ± 1.7 b B		
Zabrotes subfasciatus	$0.4 \pm 0.8 \text{ a C}$	$0.6 \pm 1.0 \text{ a C}$		
Survivorship				
Thyreophagus sp.	$8.7 \pm 2.1 \text{ a A}$	9.9 ± 1.7 b B		
Zabrotes subfasciatus	$7.2 \pm 3.4 \text{ a A}$	$7.8 \pm 3.4 \text{ a A}$		
Anagasta kuehniella	$8.6 \pm 1.5 \text{ a A}$	7.3 ± 2.7 a A		
Lasioderma serricorne	$7.2 \pm 2.6 \text{ a A}$	$7.6 \pm 3.6 \text{ a A}$		

<sup>\*</sup> For each parameter, different lowercase letters indicate significant differences between prey offered for the same predator, while different uppercase letters indicate significant differences between the two predators for the same prey (p<0.05; Kruskal-Wallis).

Table 2: Duration of the immature stages of development (days  $\pm$  standard deviation) of *Blattisocius everti* when offered a mixture of all developmental stages of *Thyreophagus* sp. as prey, at 25.0  $\pm$  0,5 °C, 80  $\pm$  5 % relative humidity and in the dark (n= initial numbers).

Stages	Female (n= 22)	Male (n= 20)	Total (n= 42)
Egg	$3.6 \pm 0.6 \text{ a}^*$	$3.5 \pm 0.2 a$	$3.5 \pm 0.4$ a
Larva	$1.0 \pm 0.0 \text{ c}$	$1.0 \pm 0.2$ c	$1.0 \pm 0.2 \text{ c}$
Protonymph	$2.3 \pm 0.7 \text{ ab}$	$2.3 \pm 0.7 \text{ b}$	$2.3 \pm 0.7 \text{ b}$
Deutonymph	$1.7 \pm 0.5 \text{ cb}$	$1.9 \pm 0.6 \text{ cb}$	$1.8 \pm 0.5 \text{ b}$
Egg – adult	$8.7 \pm 0.7$	$8.7 \pm 0.3$	$8.7 \pm 0.5$

<sup>\*</sup>Within each column, averages followed by different letters are statistically different (p<0.05; Kruskal-Wallis).

tween predators was not significant when *Z. subfasciatus* was offered as food.

For both predators, oviposition rates were significantly higher on *Thyreophagus* sp. than on any of the insects (Table 1). The difference was larger for *B. everti*, which reached an average of about 1.5

eggs per female per day when offered *Thyreophagus* sp. as food. On insects, oviposition rates reached at most about 0.3 eggs per female per day (*B. everti* on *E. kuehniella*).

Both predators had high rates of survivorship on all prey (at least average of 7.2 days) (Table 1). The only significant difference between predators referred to the higher survivorship of *B. everti* than of *B. keegani* on *Thyreophagus* sp. However, a complementary evaluation done after the conclusion of this study showed that average survivorship of starving *B. everti* kept in isolation in humid experimental units (n= 30) was  $7.0 \pm 3.1$ days.

#### Life table of B. everti

Significant differences between sexes in relation to duration of each developmental stage were not observed (Table 2). The ontogenetic development (egg - adult) of both sexes was completed in about 8.7 days. This duration was longer than reported by Nielsen (2001) for *B. tarsalis* at 25 °C when *E. kuehniella* eggs were offered as prey (7.0 days at 75 % and 6.0 days at 60 % relative humidity). However, it was shorter than reported by Thomas *et al.* (2011) for *B. keegani* at 25°C and 50-60 % of relative humidity, when *Amyelois transitella* Walker (Lepidoptera: Pyralidae) eggs were offered as prey (9.2 days).

Survivorship rates were high in this study; egg survivorship was 93.4 % and survivorship of other stages was 100 %. Sex ratio was 84 % of females. Pre-oviposition, oviposition and post-oviposition periods were  $2.5 \pm 1.1$ ,  $14.8 \pm 6.2$  and  $6.9 \pm 5.7$  days, respectively. These durations account for a female longevity of  $22.9 \pm 8.9$  days.

Mortality of adult females was first observed on the eleventh day from the adult emergence; mortality rate was approximately constant afterward, with the last females dying about 40 days of adult emergence. Maximum oviposition rate was reached on the ninth day of adult emergence; oviposition rate then reduced at about a constant rate, finishing 27 days from female emergence, paralleling the pattern of female mortality rate. This yielded a daily oviposition rate of  $0.7 \pm 1.5$  eggs per female and fecundity of  $24.5 \pm 13.8$  eggs per female. Calculated life table parameters were net reproduction rate ( $R_o$ ) 16.95, mean generation time (T) 21.2 days, intrinsic rate of population increase ( $r_m$ ) 0.13 and finite rate of population increase ( $\lambda$ ) 1.14.

#### **DISCUSSION**

The difference in relation to relative length of fixed and movable digits of *B. keegani* and *B. everti* observed in this study was already expected from information available in the literature (Haines, 1978; Britto *et al.*, 2012).

#### Predator performance on potential food sources

Notwithstanding the high survivorship of *B. everti* in the absence of food observed in the complementary test, experimental results supported the hypothesized best biological performance of a *Blattisocius* species with cheliceral digits of similar lengths (*B. everti*) as predators of acarid mites than of a *Blattisocius* species with fixed digit much shorter than movable digit (*B. keegani*). The similar predation rates of both predators on *Thyreophagus* sp. and the concurrent lower oviposition rate of *B. keegani* suggest that the short fixed digit does not preclude the predator from attacking (and killing) the mite prey, but that it makes it more difficult for the predator to consume that prey.

The results were not conclusive in relation to the performance of the predators on insects as food sources, given the low predation and oviposition rates on the insects, despite the significantly higher oviposition rate of B. everti on E. kuehniella and L. serricorne. Edentate or reduced chelicerae, or chelicerae with attenuate fixed digit have been mentioned as typical of obligatory parasitic Mesostigmata (Houck et al., 1991; Halliday et al., 1998). In addition to B. keegani, Blattisocius apis Basha and Yousef, B. patagiorum Treat and B. tarsalis, Mesostigmata of several other groups are known to have fixed cheliceral digit distinctly shorter than movable digit. Examples of species of that mite order with short fixed digit include Blaberolaelaps species (Laelapidae), so far reported only on cockroaches (Blattodea), while examples of species of the same order with fixed digit absent altogether and movable digit well developed include all known species of Otopheidomenidae (on certain families of moths, hemipterans, katydids and termites), and Varroa species (only known from honeybees) (Krantz and Walter, 2009).

It was expected that by having fixed cheliceral digit much shorter than movable digit, B. keegani could perform better than B. everti on the insects. The poor performance of the former on all insects offered as food reflects the fact that adaptation of the chelicera is just one of the requisites for insect feeding. This predator may require as prey either an insect species not offered in the study or an unavailable phase of the insects considered in the study (recently laid eggs or other developmental stages). Blattisocius keegani has been reported to feed on eggs of several lepidopteran (Stein, 1960; Trivedi et al., 1994; Thomas et al., 2011) and coleopteran (Baker, 1967; Beavers, 1972; Hughes, 1976) species, as well as on astigmatid mites (Baker, 1967; Hughes, 1976; Rezk, 2000; Taha et al., 2007). The results of this study are compatible with the fact that despite the co-existence of those predators on dog food infested with astigmatid mites, B. everti was always found in much higher numbers than B. keegani in the stock colony.

A much larger number of studies has shown the consumption of insects common on storage facilities by *Blattisocius tarsalis* (Berlese), another species with fixed cheliceral digit much shorter than movable digit. However, despite the common occurrence of astigmatid mites in those environments, little has been published about the relation between the latter and *B. tarsalis*, always with poor performance of the predator (Riudavets *et al.*, 2002; Thind and Ford, 2006).

#### Life table of B. everti

Efforts have been dedicated worldwide to finding new predatory mites for the control of distinct pest mites, but few studies have been conducted to evaluate the effect of *Blattisocius* species with cheliceral digits of similar lengths on mites of storage facilities. Yet, very promising result was reported by Rivard (1962) when *T. putrescentiae* was offered as prey to *Blattisocius dentriticus* (Berlese); values of r<sub>m</sub> of up to 0.621 were reported by the author.

The results of the life table study showed the ability of *B. everti* to develop and confirmed its ability to oviposit when offered an acarid mite as prey, suggesting that it should deserve further attention

in the search for prospective predators for possible use as a control agent of pest mites in storage facilities. The results also indicated the potential of *Thyreophagus* sp. as a food source for mass production of this and perhaps of other predatory mesostigmatid mites. The ability to mass produce a biological control agent using acarid mites as food sources is of major importance (Freire and Moraes, 2007), given that the predator cost of production is one of the liming factors in the adoption of biological control.

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