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Development and Reproduction of *Podisus nigrispinus* (Hemiptera: Pentatomidae) Fed with *Thyrinteina arnobia* (Lepidoptera: Geometridae) Reared on Guava Leaves

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ABSTRACT

The aim of this study was to evaluate the development and reproduction of P. nigrispinus *in laboratory when fed with* T. arnobia *reared on guava leaves. This predator showed nymphal stage of 21.11 days, survival of 60% and periods of pre-oviposition, number of eggs/mass and eggs/female and egg viability of 6.10 days, 26.24 eggs, 314.90 eggs and 82.65%, respectively. These results demonstrated that* T. arnobia *fed with guava leaves was an adequate supply of food to* P.nigrispinus.

Key words: biological control; defoliator; stinkbug

INTRODUCTION

Predators of the genus *Podisus* are natural enemies used for biological control. They occur in the entire American Continent (Thomas, 1992) and have been found in agriculture and forest areas in Brazil (Zanuncio et al., 1993). *Podisus nigrispinus* (Dallas, 1851) (Heteroptera: Pentatomidae), one of the most common species of this genus in the Neotropical areas, was reported in Brazil at Espírito Santo, Maranhão, Minas Gerais, Mato Grosso do Sul, Pará and São Paulo States. This species has been found in the following crops: cotton, coffee, eucalyptus, pinus, corn, soybean and wheat (Guedes et al., 2000; Pereira et al., 2001).

The brown eucalyptus caterpillar *Thyrinteina arnobia* (Stoll, 1782) (Lepidoptera: Geometridae) is one of the most important defoliator of *Eucalyptus* spp., *Psidium guajava*, *Campomanesia* spp. and *Eugenia* spp. (Oliveira et al., 2005). Outbreaks of this species in forest plantations have been controlled, in some situations, with insecticides. However, insecticides may kill natural enemies (Guedes et al., 1992) and contaminate the environment. Therefore, there is a need to develop alternative control methods for this pest (Zanuncio et al., 1991). Due to the facility

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of rearing predatory stinkbug in laboratory, these insects have been produced and released in biological control programs of defoliating caterpillars (Zanuncio et al., 2002).

However, this approach may decrease their efficiency in the field because the stimuli they receive in mass rearing are different from those in natural prey. Therefore, biological aspects of these predators can be adversely affected (Sznajder and Harvey, 2003). The knowledge about the performance of *P. nigrispinus* reared on natural prey is important not only to obtain the information about this predator on another host, but also to obtain data on its establishment in areas with the culture.

Holtz et al. (2007) evaluated the performance of *P. nigrispinus* fed with *T. arnobia* caterpillars and demonstrated that this predator was not adapted to that prey when reared on eucaliptus leaves. The adaptation of herbivore in differents hosts has been the subject of ecological studies (Agrawal, 2000). However, low attention has been given to the investigation about how the predators can adapt to the prey when them come from differents kind of host. The main of this study was to evaluate the development and reproduction of *P. nigrispinus* in laboratory, when fed with *T. arnobia* reared on guava leaves.

MATERIALS AND METHODS

Rearing the prey *Thyrinteina arnobia*

Caterpillars of *T. arnobia* were obtained from a mass rearing at Entomology Laboratory of Federal University of Espirito Santo. The pupae were conditioned in PVC tubes with 200 mm diameter and 25 cm height. The adults of *T. arnobia* were transferred just after emergence to a 38 x 41 x 45 cm wood box with paper leaf in its internal wall as oviposition site. A glass door was put on one side of this box to facilitate the handling of the insects. These adults received a 10% honey solution in anesthetic tube type and their egg masses were daily collected and maintained in an acclimatized chamber at 25 ± 3ºC, relative humidity of 70 ± 10% and photo phase of 12 h. The eggs of *T. arnobia* were maintained in Petri dishes (9.0 x 1.2 cm) until emergence of the caterpillars that were transferred to plastic tubes (15 x 12 cm) with the leaves of guava (*Psidium guajava*) until the pupa stage.

Maintenance and multiplication of the predator *Podisus nigrispinus*

P. nigrispinus was reared in an acclimatized room at 25 ± 3ºC, relative humidity of 70 ± 10% and photophase of 14 h. Ten adults of *P. nigrispinus* were maintained in plastic cup of 500 ml with absorbent paper as oviposition site. Water was supplied in an anesthetic tube type fixed at the cover of these cups and with its extremity locked with cotton. *P. nigrispinus* was fed with *Tenebrio molitor* (Linnaeus, 1758) (Coleoptera: Tenebrionidae) larvae.

Laboratory experiment

Stage I

A total of 80 replications were used in which each one was constituted by an individualized second instar *P. nigrispinus* nymph in a Petri dish (9.0 x 1.5 cm). A cotton pad (3 x 3 cm) was put on the inside top of each dish and daily moistened with distilled water to maintain the humidity and water supply for these nymphs. The inferior part of the dishes was covered with paper filter to absorb the excess humidity. *P. nigrispinus* nymphs received fourth instar larvae of *T. arnobia*, which were replaced when necessary.

Stage II

Twenty pairs of recently emerged *P. nigrispinus* adults were obtained from the stage I. Each of these pairs was conditioned in a plastic cup (9.5 cm height and 10 cm diameter). An anesthetic tube type was put the inside cover of these cups with its extremity locked with a cotton pad to supply water to the insects. The internal part of these cups was wrapped with paper as oviposition site. These adults received caterpillars of *T. arnobia* which were replaced when necessary.

Statistical analysis

The weight and weight gain was observed for the each instar and up to the third day of the adult stage (males and females). The duration and viability of each instar of *P. nigrispinus* were also evaluated. The pre-oviposition period, the numbers of egg masses, eggs per egg mass, eggs per female and nymphs per egg mass besides the viability of eggs per egg mass per female of *P. nigrispinus* were evaluated. The analyses of variance at 5% probability level were performed for all the characteristics investigated.

RESULTS AND DISCUSSION

The weight of the nymphs of *P. nigrispinus*, independent of gender (male and female), were similar in the second, third and fourth instar (Table 1). However, the females from the fifth instar were heavier than males. Gain of weight during the second, third and fifth instars for nymphs which originated the females or males of *P. nigrispinus* were similar (Fig. 1A). However, gain of weight was higher for the females in the fourth instar compared to the males.

The males and females of this predator lost weight on the first day of the adult stage, which continued for the males on their second day of life. On the other hand, *P. nigrispinus* females had approximately 8% gains of weight during this period (Fig. 1B). Heavier weight of *P. nigrispinus* nymphs at the end of the nymph period for those that originated females and in the adult stage was similar to that found for *Podisus distinctus* (Stal, 1860) (Heteroptera: Pentatomidae) fed with *T. molitor* larvae (Zanuncio et al., 1998; Matos Neto et al., 2004) and to *P. nigrispinus* fed with *Zophobas confusa* Gebien, 1906 (Coleoptera: Tenebrionidae) larvae (Zanuncio et al., 1996). This higher weight of the females than males was due to biomass accumulation by the females, which was necessary for their reproduction from the beginning of the fifth instar. Weight gain by the females of predatory Pentatomidae has also been related to the development of their ovary and egg formation (Oliveira et al., 1999; Oliveira et al., 2002).

Table 1 - Nymphs of weight (mean ± standard error) in the second, third, fourth and fifth stages and adults until the third day (mg) of *Podisus nigrispinus* with *Thyrinteina arnobia* (25 ± 3 º C, RH 70 ± 10% and photophase of 12 hours).

Instar	Females Weight (mg)*	Range of average	Males Weight (mg)	Range of average
II	0.73 ± 0.22 a	(0.6 – 0.9)	0.74 ± 0.02 a	(0.6 – 0.9)
III	3.14 ± 0.17 a	(2.4 – 4.4)	3.21 ± 0.30 a	(2.3 – 6.8)
IV	10.20 ± 0.97 a	(6.2 – 16.2)	9.91 ± 1.00 a	(6.2 – 18.6)
V	29.95 ± 2.51 a	(15.9 – 46.5)	22.42 ± 0.89 b	(17.9 – 30.1)
1º adult day	65.39 ± 1.74 a	(51.7 – 75.4)	45.61 ± 1.64 b	(35.4 – 68.5)
2º adult day	64.23 ± 4.27 a	(33.1 – 85.0)	41.20 ± 2.08 b	(26.4 – 50.5)
3º adult day	69.21 ± 4.95 a	(41.9 – 111.7)	40.30 ± 2.36 b	(27.7 – 57.0)

* Means followed by the same letter in the line are not different at 5% of probability.

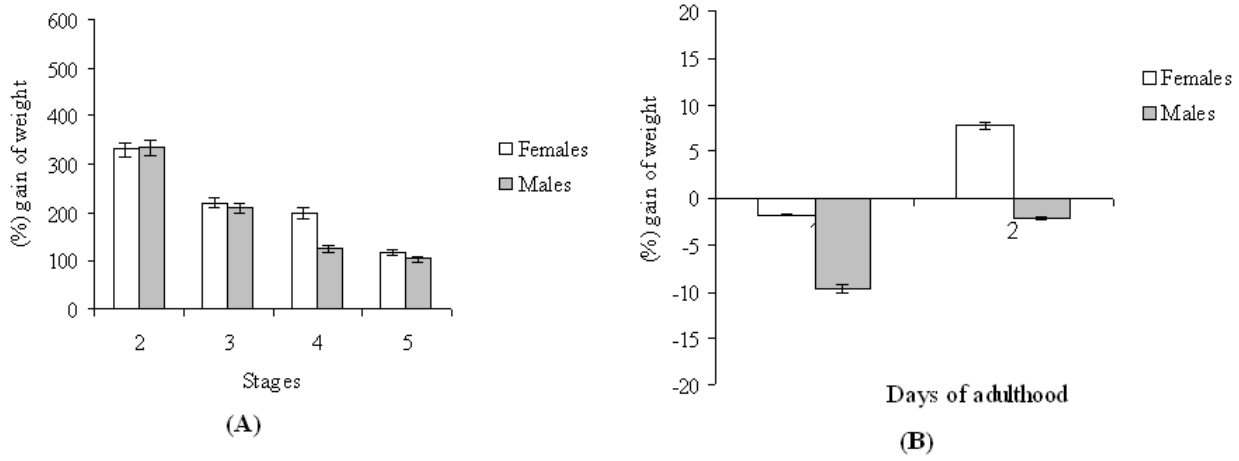

Figure 1 - Gain of weight (%) of females and males of *Podisus nigrispinus* the second, third, fourth and fifth stages (A) and the first and second days of adulthood (B) fed with larvae *Thyrinteina arnobia*. 25 ± 3 º C, RH 70 ± 10% and photophase of 12 hours.

Duration from second to fifth instar (5.18 to 6.89 days) of *P. nigrispinus* was similar to that of 4.1 to 6.7 days for this predator fed with *Musca domestica (Linnaeus, 1758*) (Diptera: Muscidae) larvae and 3.4 to 6.0 days with *Bombyx mori* (*Linnaeus*, 1758) (Lepidoptera: Bombycidae) larvae (Zanuncio et al., 1990). These values were also similar for this predator with different prey because it presented duration of each instar from 3.3 to 6.9 days with *Rachiplusia nu* (Guenée, 1852) (Lepidoptera: Noctuidae) (Saini, 1994) larvae and 3.4 to 6.2 days with *Alabama argillacea* (Huebner, 1818) (Lepidoptera: Noctuidae) caterpillars (Oliveira et al., 2002). The duration of 21.11 days for the nymph stage of *P. nigrispinus* (Table 2) was similar to that of this predator fed *Spodoptera frugiperda* (J.E. Smith, 1797) (21.6 days) and *T. molitor* pupae (20.4 days) (Oliveira et al., 2004). These results also showed that *T. arnobia* larva was an adequate prey to meet the development and reproduction requirements of *P. nigrispinus*.

The nymph viability of each instar of *P. nigrispinus* (Table 2) was similar to that found for this predator with caterpillars of *B. mori* (85 a 96%) from the second until fifth nymphal stage (Fernandes et al., 1996). The total viability (60%) was also similar when this predator were fed with *S. frugiperda* (64%) and with *T. molitor* (68%) (Oliveira et al., 2004).

The longevity of *P. nigrispinus* was 35.54 days for the females and 43.08 days for the males (Table 3). The shorter longevity of *P. nigrispinus* females than the males when fed with *T. arnobia* caterpillars agreed with that reported for this predator fed with *S. frugiperda* larvae (39.6 days) (Oliveira et al., 2004). It could be explained by the high energy use of the females for egg formation and oviposition. This occurs due to different energy allocation between the physiological processes with increase of one energy demand for egg production and decrease for maintenance (i.e. longevity) (Sibly and Calow, 1986).

Table 2 - Duration of the nymphal stages and (days) (mean ± standard error) of *Podisus nigrispinus* fed *Thyrinteina arnobia*. 25 ± 3 ºC, RH 70 ± 10% and photophase of 12 hours.

Instar	Duration (days)	Range of average	Viability (%)
II	5.18 ± 0.21	(3 – 9)	86
III	4.61 ± 0.11	(4 – 6)	86
IV	4.43 ± 0.16	(3 – 7)	95
V	6.89 ± 0.22	(6 – 11)	86
Total	21.11 ± 0.52	(18 – 27)	60

Table 3 - Mean ± standard error of biological aspects of *Podisus nigrispinus* adults fed with *Thyrinteina arnobia* in the laboratory (25 ± 3 ºC, RH 70 ± 10% and photophase of 12 hours).

Parameters	*Podisus nigrispinus*	*Range of average*
Pre-oviposition (days)	6.10 ± 0.91	(2 – 11)
Number of eggs/females	314.90 ± 59.31	(121 – 679)
Egg viability (%)	82.65 ± 1.33	(22 – 100)
Longevity (females)	35.54 ± 3.02	(19 – 61)
Longevity (males)	43.08 ± 4.72	(17 – 65)

The pre-oviposition period per female of *P. nigrispinus* was 6.10 days (Table 3), which was similar to 6.3 days found for this predator in *T. arnobia* reared in eucalyptus leaves (Holtz et al., 2006) and with *A. argillacea* caterpillars (Oliveira et al., 2002).The number of eggs per female of *P. nigrispinus* fed with *T. arnobia* (314.90), was similar to that found for this predator when fed with *A. argillacea* caterpillars and cotton crop (348,10) (Oliveira et al., 2002) and with *T. molitor* (296,66; 325,00) (Espindula et al., 1996; Oliveira et al., 2004) and higher than that when fed with *Diatraea saccharalis* (Fabricius, 1794) (Lepidoptera: Crambidae) (97,12) (Vacari et al., 2007) and with *T. arnobia* (57.0) reared with eucalyptus (Holtz et al., 2006; 2007). The highest number of eggs found for *P. nigrispinus* when fed with *A. argillacea* and cotton crop on *T. molitor*

larvae and with caterpillars of *T. arnobia* reared in guava leaves, suggested that these preys may have better nutritional requirements for *P. nigrispinus*.

Eggs viability of this predator was 82.65% (Table 3), which was higher than that reported by Holtz et al. (2006) for *P. nigrispinus* fed with *T. arnobia* (56.4 %), when reared with eucalyptus leaves. In *T. molitor* (79.2 %) and *S. frugiperda* (85.2 %) (Oliveira et al., 2004), results were similar showing that *P. nigrispinus* fed with *T. arnobia* from guava leaves had adequate nutritional quality.

Results showed that *T. arnobia* reared on guava leaves could be an appropriate prey to *P. nigrispinus*. Holtz et al. (2007) reported that this caterpillar reared on eucalyptus leaves was not a good prey, because these leaves had a specific essence which produced secondary substances, which might affect stinkbug development. However, Holtz et al. (2003) evaluated the effect of these substances on *T. arnobia* and found that these compounds did not affect the development of this caterpillar on eucalyptus, but others substances that come from guava could affect their development. Santos et al. (2000) and Oliveira et al. (2005) found that *T. arnobia* had better development on guava leaves, which could be considered as the best host for it, while eucalyptus leaves had poor digestibility and nutritional quality which reduced the development of this caterpillar. For this reason, when *T. arnobia* was reared with eucalyptus leaves, probably the larvae had a lower quality and therefore affected the development of the predator.

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