

EFFECT OF HONEY FEEDING BY *THYRINTEINA ARNOBIA* MALES AND FEMALES ON THEIR REPRODUCTION AND LONGEVITYDENISE E. EUZÉBIO¹, JOSÉ C. ZANUNCIO¹, ROSENILSON PINTO², CARLOS F. WILCKEN³, FRANCISCO S. RAMALHO⁴
AND ERALDO LIMA¹¹Departamento de Entomologia, Universidade Federal de Viçosa, 36570-000, Viçosa, Minas Gerais, Brazil²Faculdade de Engenharia-Universidade Estadual de Minas Gerais, 35930-314, João Monlevade, Minas Gerais, Brazil³Departamento de Produção Vegetal, Faculdade de Ciências Agronômicas, Universidade Estadual Paulista, 18603-970, Botucatu, São Paulo, Brazil⁴Unidade de Controle Biológico/EMBRAPA Algodão, Av. Osvaldo Cruz, 1143, 58107-720, Campina Grande, Paraíba, Brazil

Corresponding author; E-mail: zanuncio@ufv.br

ABSTRACT

Nutrients obtained during the larval period can be sufficient for reproduction, but adult feeding may be necessary to reproductive maturity and ripening of eggs. Adult food requirements of *Thyriniteina arnobia* (Stoll) (Lepidoptera: Geometridae), the main defoliator of eucalyptus in Brazil, may present problems in the laboratory. The fecundity and longevity of *T. arnobia* adults fed on 15% honey solution were evaluated. This solution did not improve the reproductive capacity and longevity of *T. arnobia* females but it favors those of males, which could increase mating probability. This is important because *T. arnobia* males emerge sooner than females, and feeding them could increase their longevity and chances of mating various females.

Key Words: food supply; longevity; nutrition; reproduction

RESUMO

A alimentação no período larval de Lepidoptera pode garantir nutrientes para crescimento e/ou reprodução, mas estes insetos podem necessitar de alimento na fase adulta para maturação do aparelho reprodutor e dos óvulos. *Thyriniteina arnobia* (Stoll) (Lepidoptera: Geometridae), o principal desfolhador de plantas de eucalipto no Brasil, apresenta dificuldades de criação em laboratório. A fecundidade e longevidade de adultos de *T. arnobia* alimentados com solução de mel a 15% foram avaliadas. Essa solução não melhorou a capacidade reprodutiva e a longevidade de fêmeas de *T. arnobia*, mas aumentou a longevidade de machos, o que pode permitir o acasalamento com diferentes fêmeas. Isto é importante porque machos de *T. arnobia* emergem mais cedo e a alimentação pode aumentar a longevidade dos mesmos e permitir o acasalamento com várias fêmeas.

Palavras Chaves: Alimentação suplementar; longevidade; nutrição; reprodução

Geometridae larvae feeding on plants may become pests in cultivated forests (Swank et al. 1981). *Thyriniteina arnobia* (Stoll, 1782) (Lepidoptera: Geometridae) defoliates native Myrtaceae and it has been reported in eucalypt plantations with population outbreaks as the most important defoliator of this culture in Brazil (Bernardino et al. 2007; Zanuncio et al. 2003). The integrated management of this pest includes use of insecticides. (Oliveira et al. 2011), predators (Holtz et al. 2006; Zanuncio et al. 2006) and parasitoids (Menezes et al. 2012; Pereira et al. 2008a, 2008b).

Nutrition of adults of holometabolous insects is complex (Romeis & Wäckers 2002; Shirai 2006; Geister et al. 2008) because they depend, primarily, on nutrients accumulated during the larval stage (O'Brien et al 2002; Jervis & Boggs 2005), and food supplements for adults (Bauerfeind & Fischer 2005, 2009; Kehl & Fischer 2012; Milano et al. 2010).

Adult geometrid moths usually have a functional proboscis, and it is likely that most tropical species rely on flower nectar as their food source, although evidence is scarce (Axmacher et al. 2009).

Nutrition during the adult stage can affect the insect life cycle including longevity, preoviposition period and fecundity (Wackers et al. 2007). Sugar is important for reproduction (Watanabe 1992; O'Brien et al. 2004; Fischer & Fiedler 2001) and longevity (Carroll & Quiring 1992) of adult Lepidoptera. Most species of this order feed during adult stage on carbohydrate-rich diet (Romeis & Wackers 2000) what generally increases longevity and fecundity (Leather 1995).

Floral nectar is a source of food containing different types of sugars such as sucrose and fructose and glucose, and other nutrients such as amino acids (Romeis & Wackers 2000). Lepidoptera males and females can present different foraging strategies. Thus males feed on flowers with high sugar levels, while females may feed on flowers with high proportions of glucose and amino acids (Rusterholz & Erhardt 2000). These nutrients, which are important for the production of eggs, are also found in honey.

Thyriniteina arnobia adults are fed honey in the laboratory (Oliveira et al. 2005; Batista-Pereira et al. 2004) but their real need of this food is not clear. The objective was to study the effect of honey feeding by adults on reproduction and longevity of *T. arnobia* to improve its mass rearing.

MATERIALS AND METHODS

Adults *T. arnobia* were obtained from a rearing facility of the Laboratory of Biological Control (LBCI) of the Federal University of Viçosa (UFV) in Viçosa, Minas Gerais State, Brazil. Larvae of this species were reared for one generation on *Psidium guajava* L.(Myrtales: Myrtaceae) plants in the field until the pupal stage and then transferred to the laboratory where they were maintained at $25 \pm 2^\circ\text{C}$ and $70 \pm 8\%$ RH until they reached adult stage. Newly emerged *T. arnobia* adults were separated into pairs in 500 mL plastic vials, covered with a perforated lid for mating and oviposition at $22.39 \pm 1.38^\circ\text{C}$ and $82.35 \pm 6.56\%$ RH. A 15% honey solution was supplied on cotton balls and changed daily. The pots with *T.*

arnobia pairs were held in an open area near the LBCI in shelves until their death.

Treatments consisted of 32 pairs of *T. arnobia* fed with 15% honey solution, and 30 unfed pairs. Each pair was a replication.

Egg masses of *T. arnobia* were collected and maintained at $25.0 \pm 2.0^\circ\text{C}$, $70 \pm 8\%$ RH and 12 h photophase until they had hatched. The preoviposition, oviposition and post-oviposition periods, and the numbers of egg masses and eggs per female, longevitys of males and females and egg viabilitys of *T. arnobia* were evaluated daily. Egg masses of females with more than one egg mass were used to evaluate egg viability; and the viabilitys of first and last egg masses were evaluated. Data were subjected to the Wilcoxon test at the 5% probability level.

RESULTS

The mean preoviposition period of *T. arnobia* was similar between treatments with 2.06 days for females fed on honey solution and 1.9 days for non fed ones ($P < 0.05$) (Table 1).

The mean numbers of eggs and egg masses per *T. arnobia* female (Table 1) were, respectively, 1,173.2 eggs and 3.0 egg masses (fed) and 1,140.0 eggs and 3.4 egg masses (unfed), and with viabilitys and incubation periods of 85.8% (fed) and 78.5% (unfed) and 9.1 days (fed) and 9.2 days (unfed), respectively. These differences between the 2 treatments were not significant. In both treatments the first egg mass to be deposited by a female had about 3-fold more eggs than the second egg mass, and all subsequent egg masses had even fewer eggs that the second egg mass (Fig. 1). Egg viabilitys were 82.43% for the first egg mass and 74.57% for the last one in the honey-fed treatment, and 78.58% for the first egg mass and 66.26% for the last egg mass in the unfed treatment; and the Wilcoxon test values of both treatmentst were similar values by ($P < 0,05$).

However the longevity of the honey-fed *T. arnobia* males (9.6 days) was longer than of unfed males (7.3 days) ($P < 0.05$) (Table 1). Honey-fed

TABLE. 1. REPRODUCTIVE CAPACITIES AND LONGEVITIES OF *THYRINTEINA ARNOBIA* (LEPIDOPTERA: GEOMETRIDAE) EITHER FED 15% HONEY SOLUTION OR NOT FED. THEY WERE HELD AT $22.39 \pm 1.48^\circ\text{C}$ AND $82.35 \pm 6.56\%$ RH.

	Fed (T1) (n = 32)	Unfed (T2) (n = 30)
Preoviposition (days) ^{ns}	2.06	1.90
Total number of eggs/female ^{ns}	1173.20	1140.03
Egg masses per female ^{ns}	3.03	3.41
Egg viability (%) ^{ns}	85.79	78.52
Egg incubation (days) ^{ns}	9.15	9.22
Female longevity (days) ^{ns}	9.35	10.07
Male longevity (days)*	9.58	7.37

* Significant with the Wilcoxon test ($P < 0.05$). ^{ns} Non-significant.

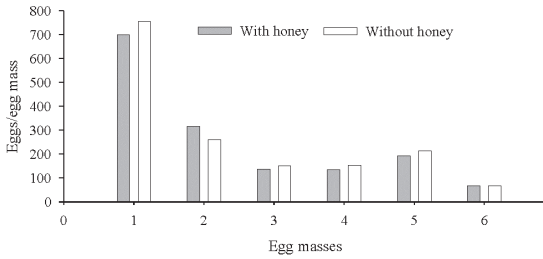


Fig. 1. Number of eggs per egg mass of *Thyriniteina arnobia* (Lepidoptera: Geometridae) fed or unfed on 15% honey solution at 22.39 ± 1.48 °C and $82.35 \pm 6.56\%$ RH.

males showed 100% survival until the fifth day, compared to only 45% of unfed males. Few unfed males survived until the twelfth day, but 20% of honey-fed males survived for that period (Fig. 2A).

The longevity of honey-fed *T. arnobia* females was 9.3 days compared to 10.07 days for the unfed ones; thus these longevitys were similar values between treatments (Table 1). The survival curves of honey-fed and unfed females were similar and both decreased continuously with time (Fig. 2B).

DISCUSSION

The honey diet of the adults had definite effect on reproduction, but not on the pre-oviposition periods of fed vs. unfed females. This indicates that *T. arnobia* females do not need feeding to initiate oviposition. This is similar to that reported for *Pseudaletia sequax* (Franclemont, 1951) (Lepidoptera: Noctuidae) whose pre-oviposition did not differ when receiving water or different carbohydrates (Marchioro & Foerster 2012). This

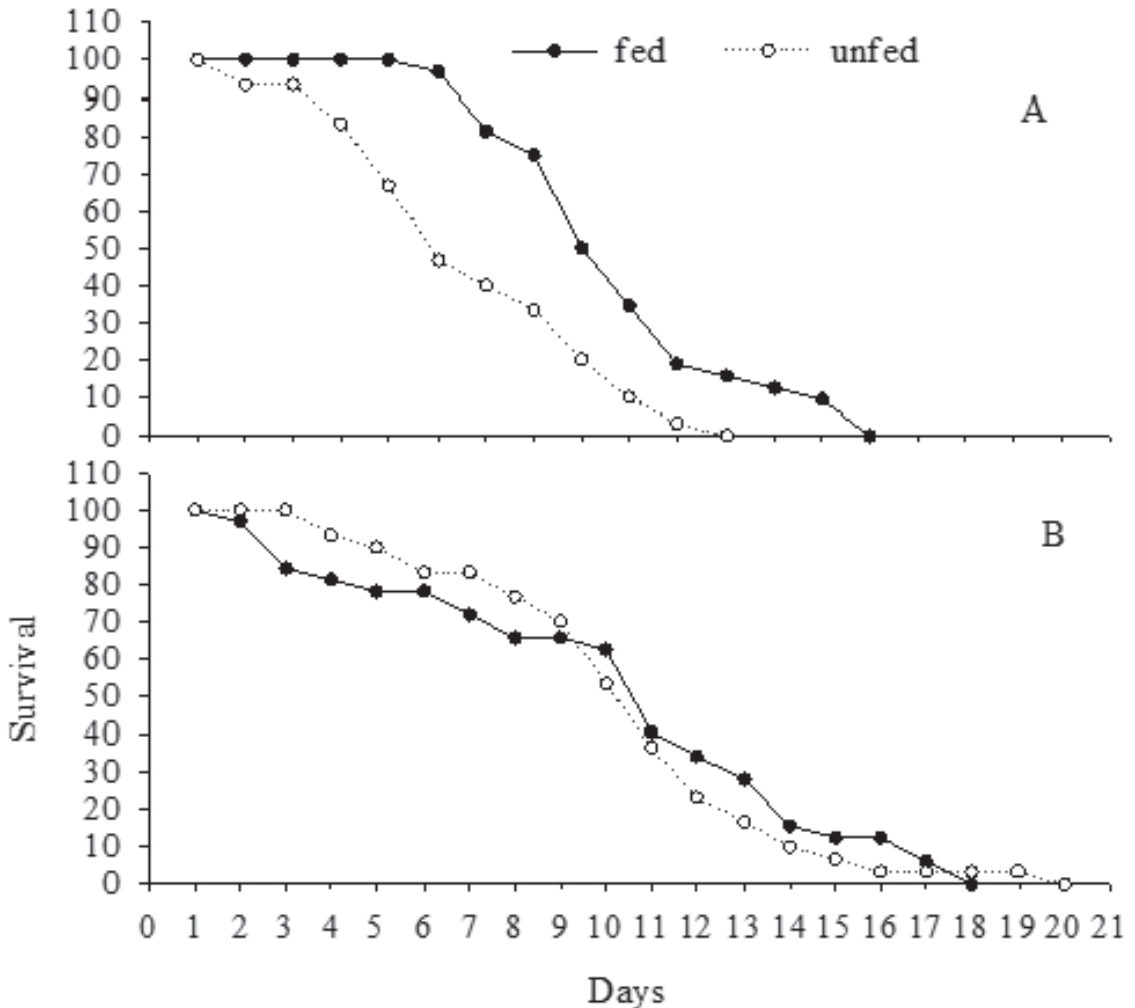


Fig. 2. Daily survival (%) of males (A) and females (B) *Thyriniteina arnobia* (Lepidoptera: Geometridae) fed or unfed on 15% honey solution at 22.39 ± 1.48 °C and $82.35 \pm 6.56\%$ RH.

is not uncommon, and many lepidoptera species need no food during pre-oviposition because the females use the reserves they retained from the larval stage to produce eggs (Wheeler et al. 2000).

In both treatments the *T. arnobia* females when 2 days old laid most of their eggs in the first egg mass; and because the few number of eggs/eggmass in egg masses #3 to #6, the longevity of the females did not greatly affect the number of eggs that they lay. *Thyriniteina arnobia* does not need to feed in the adult stage. Likewise *Diatraea saccharalis* (Fabricius 1794) (Lepidoptera: Crambidae) adult females have no need, and their fecundities are similar are increased by being fed honey, sucrose, glucose in comparison to no food (Parra et al. 1999). Some lepidoptera adults can obtain carbohydrates and proteins from nectar, which are also important for egg production (Telang et al. 2001).

Feeding during the adult stage increased longevity of *T. arnobia* males. The same result had been found for *Spoladea recurvalis* (Fabricius, 1775) (Lepidoptera: Pyralidae) males fed a honey solution, which increased their speed and duration of flight compared to those receiving only water (Shirai 2006). This indicates the importance of feeding males that emerge sooner than the females. This occurs in the case of *T. arnobia* because the males have one instar less than the females. Thus, feeding may increase flight capacity of males, their capacity to find females and their mating probability. Moreover, male feeding is important because males can transfer nutrients to females in the spermatophore during mating (Watanabe & Hirota 1999).

Therefore, the similarity of longevity of the *T. arnobia* females between the 2 treatments may be attributed to the nutrients accumulated during the larva stage. This shows that appropriate food is critically more important for female than male *T. arnobia* larvae. In addition, in other lepidopterans, nutrients accumulated during the larval stage are known to supply adults with energy and nutrients to lay eggs (Awmack & Leather 2002). Besides, the importance of food to the youngest instars of *T. arnobia* is evident from their preferences between eucalyptus species depending on the instar and the age of the leaves (Lemos et al. 1999). Larval development in *T. arnobia* depends on the diet, which subsequently affects the reproduction and calling behavior of the female (Batista-Pereira et al. 2004). Adults of other lepidopteran species may need richer carbohydrate diets for reproduction and survival (Bauerfeind & Fischer 2005). Besides in some species, supplemental sugar feeding may increase of lifetime fecundity by 10-fold (Wackers et al. 2007).

CONCLUSIONS

Supplemental feeding of *T. arnobia* males on a honey solution is important to increase their longevity and to increase their capacity to finding females over a longer period of time. Moreover, *T. arnobia* adult females did not need supplemental feeding as adults, because they have a sufficient reserve of nutrients from the larva stage to enable them to reproduce. In addition, since *T. arnobia* females lay most of their eggs in the first egg mass, the prologation of their longevity does not result in many more progeny.

ACKNOWLEDGMENTS

To "Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq), Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES)" and "Fundação de Amparo à Pesquisa do Estado de Minas Gerais (FAPEMIG)".

REFERENCES CITED

- AWMACK, C. S., AND LEATHER, S. R. 2002. Host plant quality and fecundity in herbivorous insects. *Annu. Rev. Entomol.* 17: 817-844.
- AXMACHER, J. C., BREHM, G., HEMP, A., TUNTE, H., LYARUU, H. V. M., HOHENSTEIN, K. M., AND FIEDLER, K. 2009. Determinants of diversity in afrotropical herbivorous insects (Lepidoptera: Geometridae): plant diversity, vegetation structure or abiotic factors? *J. Biogeogr.* 36: 337-349.
- BATISTA-PEREIRA, L. G., WILCKEN, C. F., NETO, S. D. P., AND MARQUES, E. M. 2004. Comportamento de chamamento de *Thyriniteina arnobia* (Stoll) (Lepidoptera: Geometridae) em *Psidium guajava*, *Eucalyptus grandis* e em dieta artificial. *Neotrop. Entomol.* 33: 21-28.
- BAUERFEIND, S. S., AND FISCHER, K. 2005. Effects of adult-derived carbohydrates and amino acids micro-nutrients on female reproduction in a fruit-feeding butterfly. *J. Insect Physiol.* 51: 545-554.
- BAUERFEIND, S. S., AND FISCHER, K. 2009. Effects of larval starvation and adult diet-derived amino acids on reproduction in a fruit-feeding butterfly. *Entomol. Exp. Appl.* 130: 229-237.
- BERNARDINO, A. S., ZANUNCIO, T. Z., ZANUNCIO, J. C., LIMA, E. R., AND SERRÃO, J. E. 2007. Note on gynandromorphism in the eucalyptus defoliator *Thyriniteina arnobia* (Stoll, 1782) (Lepidoptera: Geometridae). *An. Acad. Brasileira Cienc.* 79: 235-237.
- CARROLL, A. L., AND QUIRING, D. T. 1992. Sucrose ingestion by *Zeiraphera canadenses* Mut. and Free. (Lepidoptera: Tortricidae) increases longevity and lifetime fecundity but not oviposition rate. *Canadian Entomol.* 124: 335-340.
- FISCHER, K., AND FIEDLER, K. 2001. Effects of adult feeding and temperature regime on fecundity and longevity in the butterfly *Lycaena hippothoe* (Lycaenidae). *J. Lepid. Soc.* 54: 91-95.
- GEISTER, T. L., LORENZ, M. W., HOFFMANN, K. H., AND FISCHER, K. 2008. Adult nutrition and butterfly fitness: effects of diet quality on reproductive output,

- egg composition, and egg hatching success. *Front. Zool.* 5: 1-13.
- HOLTZ, A. M., ZANUNCIO, J. C., MARINHO, J. S., AND PEREIRA, C. J. 2006. Características biológicas de adultos de *Podisus nigrispinus* e *Supputius cincticeps* (Hemiptera: Pentatomidae) alimentados com *Thyriniteina arnobia* (Lepidoptera: Geometridae). *Idesia* 24: 41-48.
- JERVIS, M. A., AND BOGGS, C. L. 2005. Linking nectar amino acids to fitness in female butterflies. *Trends Ecol. Evol.* 20: 585-587.
- KEHL, T., AND FISCHER, K. 2012. Larval starvation reduces responsiveness to feeding stimuli and does not affect feeding preferences in a butterfly. *J. Insect Physiol.* 58: 1028-1035.
- LEATHER, S. R., AND HARDIE, J. 1995. Factors affecting fecundity, fertility, oviposition, and larviposition in insects pp. 143-174 *In* S. R. Leather [ed.], *Insect Reproduction*. Boca Raton, CRC Press, 255 pp.
- LEMOES, R. N. S., CROCOMO, W. B., FORTI, L. C., AND WILCKEN, C. F. 1999. Seletividade alimentar e influência da idade da folha de *Eucalyptus* spp. para *Thyriniteina arnobia* (Lepidoptera: Geometridae). *Pesq. Agropec. Brasileira* 34: 7-10.
- MARCHIORO, C. A., AND FOERSTER, L. A. 2012. Importance of carbohydrate sources to the reproductive output of the wheat armyworm *Pseudaletia sequax*. *Agric. Forest Entomol.* 14: 29-35.
- MENEZES, C. W. G., SOARES, M. A., SANTOS, J. B., ASSIS JR., S. L. A., FONSECA, A. J., AND ZANUNCIO, J. C. 20012. Reproductive and toxicological impacts of herbicides used in eucalyptus culture in Brazil on the parasitoid *Palmistichus elaeisis* (Hymenoptera: Eulophidae). *Weed Res.* 52: 520-525.
- MILANO, P., BERTI FILHO, E., PARRA, J. R. P., ODA, M. L., AND CÔNSOLI, F. L. 2010. Efeito da alimentação da fase adulta na reprodução e longevidade de espécies de Noctuidae, Crambidae, Tortricidae e Elachistidae. *Neotrop. Entomol.* 39: 172-180.
- O'BRIEN, D. M., FOGEL, M. L., AND BOGGS, C. L. 2002. Renewable and nonrenewable resources: Amino acid turnover and allocation to reproduction in Lepidoptera. *Proc. Natl. Acad. Sci.* 99: 4413-4418.
- O'BRIEN, D. M., BOGGS, C. L., AND FOGEL, M. L. 2004. Making eggs from nectar: the role of history and dietary carbon turnover in butterfly reproductive resource allocation. *Oikos* 105: 279-291.
- OLIVEIRA, H. N., ZANUNCIO, J. C., PEDRUZZI, E. P., AND ESPINDULA, M. C. 2005. Rearing of *Thyriniteina arnobia* (Lepidoptera: Geometridae) on guava and eucalyptus in the laboratory. *Brazilian Arch. Biol. Technol.* 48: 801-806.
- OLIVEIRA, H. N., ESPINDULA, M. C., DUARTE, M. M., PEREIRA, F. F., AND ZANUNCIO, J. C. 2011. Development and reproduction of *Podisus nigrispinus* (Hemiptera: Pentatomidae) fed with *Thyriniteina arnobia* (Lepidoptera: Geometridae) reared on guava leaves. *Brazilian Arch. Biol. Technol.* 54: 429-434.
- PARRA, J. R. P., MILANO, P., CONSOLI, F. L., ZERIO, N. G., AND HADDAD, M. L. 1999. Efeito da nutrição de adultos e da umidade na fecundidade de *Diatraea saccharalis* (Fabr.) (Lepidoptera: Crambidae). *An. Soc. Entomol. Brasil* 28: 49-57.
- PEREIRA, F. F., ZANUNCIO, J. C., TAVARES, M. T., PASTORI, P. L., JACQUES, G. C., AND VILELA, E. F. 2008a. New record of *Trichospilus diatraeae* (Hymenoptera: Eulophidae) as a parasitoid of the eucalypt defoliator *Thyriniteina arnobia* (Lepidoptera: Geometridae) in Brazil. *Phytoparasitica* 36: 304-306.
- PEREIRA, F. F., ZANUNCIO, T. V., ZANUNCIO, J. C., PRATISSOLI, D., AND TAVARES, M. T. 2008b. Species of Lepidoptera defoliators of Eucalyptus as new host for the parasitoid *Palmistichus elaeisis* (Hymenoptera: Eulophidae). *Brazilian Arch. Biol. Technol.* 51: 259-262.
- ROMEIS, J., AND WACKERS, F. L. 2000. Feeding responses by female *Pieris brassicae* butterflies to carbohydrates and amino acids. *Physiol. Entomol.* 25: 247-253.
- ROMEIS, J., AND WACKERS, F. L. 2002. Nutritional suitability of individual carbohydrates and amino acids for adult *Pieris brassicae*. *Physiol. Entomol.* 27: 148-156.
- RUSTERHOLZ, H. P., AND ERHARDT, A. 2000. Can nectar properties explain sex-specific flower preferences in the Adonis Blue butterfly *Lysandra bellargus*? *Ecol. Entomol.* 25: 81-90.
- SHIRAI, Y. 2006. Flight activity, reproduction, and adult nutrition of the beet webworm, *Spoladea recurvalis* (Lepidoptera: Pyralidae). *Appl. Entomol. Zool.* 41: 405-414.
- SWANK, W. T., WAIDE, J. B., CROSSLEY, D. A., AND TODD, R. L. 1981. Insect defoliation enhances nitrate export from forest ecosystems. *Oecologia* 51: 297-299.
- TELANG, A., BOOTON, V., CHAPMAN, R. F., AND WHEELER, D. E. 2001. How female caterpillars accumulate their nutrient reserves. *J. Insect Physiol.* 47: 1055-1064.
- WACKERS, F. L., ROMEIS, J., AND RIJN, P. 2007. Nectar and pollen feeding by insect herbivores and implications for multitrophic interactions. *Annu. Rev. Entomol.* 52: 301-23.
- WATANABE, M. 1992. Egg maturation in laboratory-reared female of the swallowtail butterfly, *Papilio xuthus* L. (Lepidoptera: Papilionidae), feeding on different concentration solutions of sugar. *Zool. Sci.* 9: 133-141.
- WATANABE, M., AND HIROTA, M. 1999. Effects of sucrose intake on spermatophore mass produced by male swallowtail butterfly *Papilio xuthus*. *Zool. Sci.* 16: 55-61.
- WHEELER, D. E., TUCHINSKAYA, I., BUCK, N. A., AND TABASHNIK, B. E. 2000. Hexameric storage proteins during metamorphosis and egg production in the diamondback moth, *Plutella xylostella* (Lepidoptera). *J. Insect Physiol.* 46: 951-958.
- ZANUNCIO, J. C., ZANUNCIO, T. V., FREITAS, F. A., AND PRATISSOLI, D. 2003. Population density of Lepidoptera in a plantation of *Eucalyptus urophylla* in the state of Minas Gerais. *Brazilian Animal Biol.* 53: 17-26.
- ZANUNCIO, J. C., LEMOS, W. P., LACERDA, M. C., ZANUNCIO, T. V., SERRÃO, J. E., AND BAUCE, E. 2006. Age-dependent fecundity and fertility life tables of the predator *Brontocoris tabidus* (Heteroptera: Pentatomidae) under field conditions. *J. Econ. Entomol.* 99: 401-407.