

Emergence of *Palmistichus elaeisis* Delvare & LaSalle, 1993 (Hymenoptera: Eulophidae) from Pupae of *Heraclides anchisiades capys* (Hübner, [1809]) (Lepidoptera: Papilionidae) in the Laboratory*

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Heraclides anchisiades capys (Hübner, [1809]) (Lepidoptera: Papilionidae) is an abundant insect pest on crops of *Citrus* spp. (Rutaceae) in Brazil and pupae of this defoliator could be used for the mass rearing of parasitoids. The aim of this study was to evaluate the parasitism of *Palmistichus elaeisis* Delvare & LaSalle, 1993 (Hymenoptera: Eulophidae) on *H. anchisiades capys* pupae in the laboratory. Twenty 1-day-old *H. anchisiades capys* pupae were individualized in test tubes (14 cm length × 2.2 cm diameter) with a drop of honey as food and with ten mated *P. elaeisis* females for ten days. The duration of the life cycle (egg to adult), emergence rate, total individuals emerged per pupa, sex ratio, size of the body and longevity of *P. elaeisis* males and females emerged from *H. anchisiades capys* pupae were evaluated. The duration of the life cycle of *P. elaeisis* was 21.15 ± 0.15 days. Forty percent of *H. anchisiades capys* pupae showed emergence of parasitoids, with an average of 323 ± 38 individuals from each one and a sex ratio of 0.95 ± 0.02 . The total number of parasitoids emerged was 2,584 individuals. The size of the body and the head capsule (mm) of female and male *P. elaeisis* progeny were 1.92 ± 0.04 ; 0.52 ± 0.02 and 1.28 ± 0.04 ; 0.40 ± 0.02 , respectively. The average longevity was 23.72 ± 1.15 days for females and 30.1 ± 2.42 days for males of *P. elaeisis*. *Heraclides anchisiades capys*, abundant on crops of *Citrus* spp. in Brazil, has potential to be used as a host for mass rearing of *P. elaeisis* in the laboratory for biological control programs.

Key words: biological control, *Citrus*, *Heraclides anchisiades capys*, *Palmistichus elaeisis*, parasitism, parasitoids, production, pupa.

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Biological parameters and methods to control *Heraclides anchisiades capys* (Hübner, [1809]) (Lepidoptera: Papilionidae) (Figs 1A-1D) were studied on *Citrus* spp. (Rutaceae) crops in Brazil (OLIVEIRA & URBAN 1977; LOPES *et al.* 1979; BASTOS 1980; MOTA & BASTOS 1980). Morphology of immature and adults and behavior, defense

mechanisms and aspects of parasitism were also evaluated for this insect (LEITE *et al.* 2010a, 2010b, 2010c, 2011). Pupae of *H. anchisiades capys* have thin and thick rough skin with irregular ridges, rolling forecasts and three staining patterns: dark brown or brown-gray with patches of defined contour or greenish (Figs 1E-1H). Aver-

*This work was carried out at Federal University of Viçosa (UFV) in Viçosa, Minas Gerais State, Brazil.

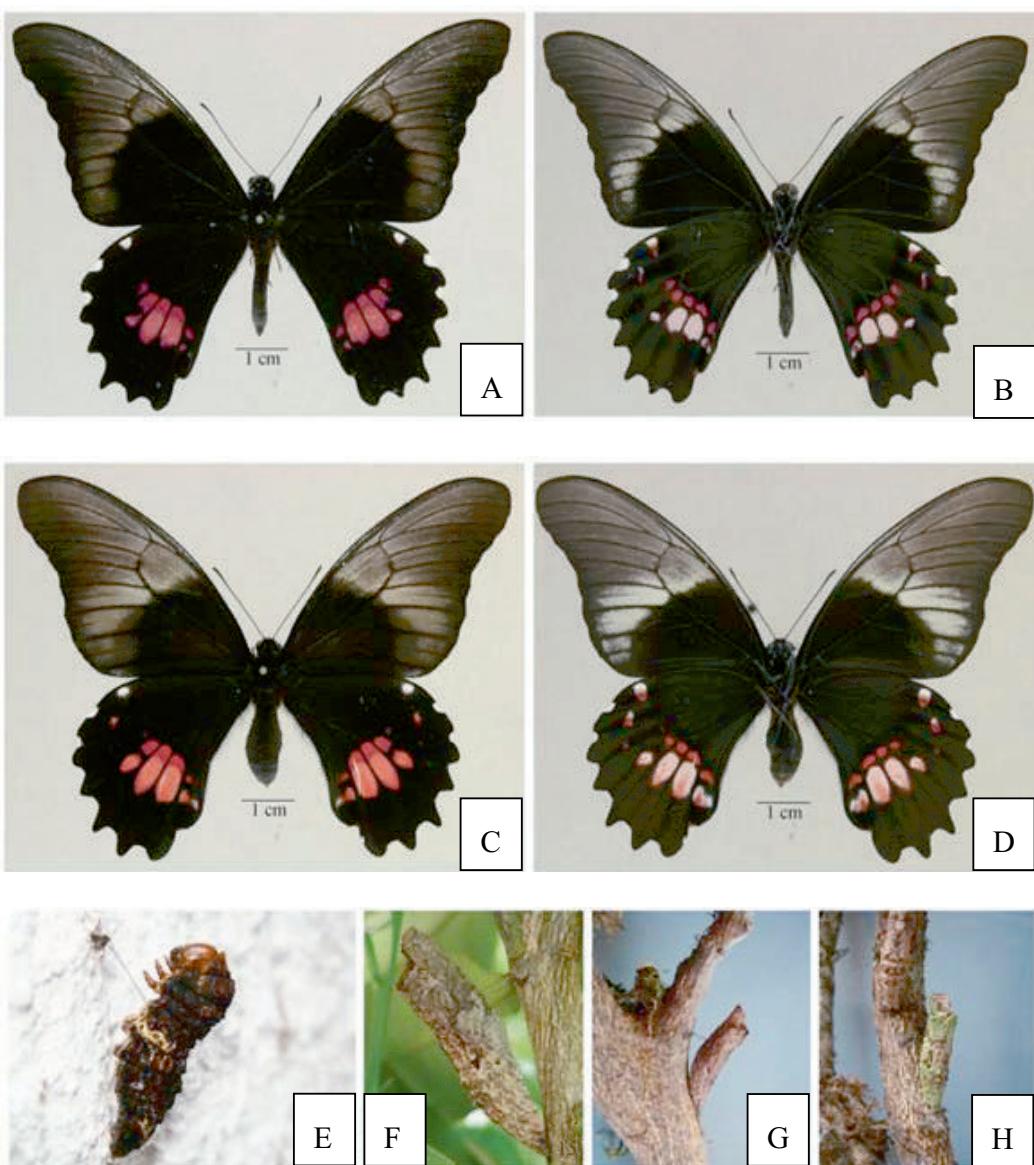


Fig. 1. *Heraclides anchisiades capys* (Hübner, [1809]) (Lepidoptera: Papilionidae). A and B – male: A, dorsal view and B, ventral view; C and D – female: C, dorsal view and D, ventral view; E – pre-pupa; F, G and H – pupa; F, pupa pattern brown-gray; G, pupa pattern dark brown and H, pupa pattern greenish (LEITE *et al.* 2010a).

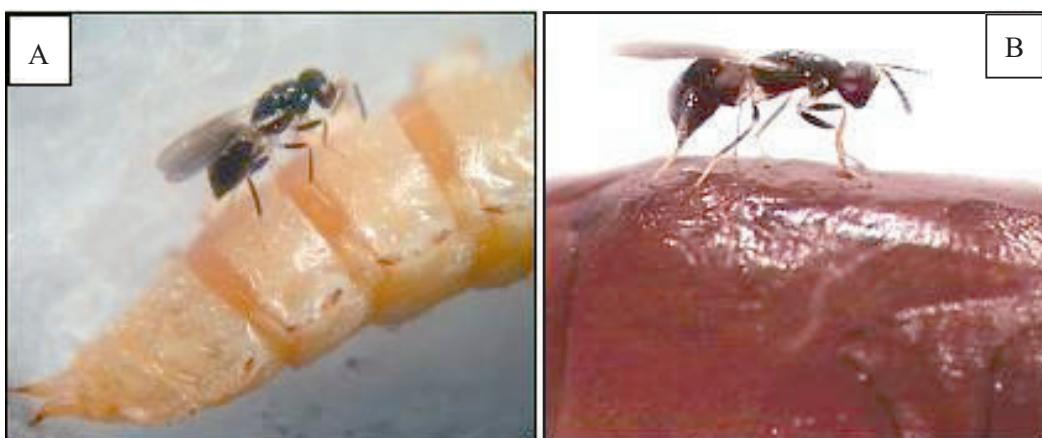


Fig. 2. Females of *Palmistichus elaeisis* Delvare & LaSalle, 1993 (Hymenoptera: Eulophidae) parasitizing *Tenebrio molitor* L., 1758 (Coleoptera: Tenebrionidae) – A and *Anticarsia gemmatalis* Hübner, 1818 (Lepidoptera: Noctuidae) – B, pupae, respectively (TAVARES *et al.* 2011b).

age length of pupae of both sexes of this insect is 33.7 mm with a minimum duration of this stage of 24 days (LEITE *et al.* 2010a).

Larvae and pupae of *H. anchisiades capys* have defense mechanisms against natural enemies. The osmeterium gland, with a defense function, is protracted under threat and its odor from first-instar caterpillars is barely noticeable, but it is intense from second-instar. Pupation of this insect may occur on *Citrus* spp. plants and also on neighboring plants or any substrates, such as walls (LEITE *et al.* 2010a).

First-instar *H. anchisiades capys* caterpillars group in the center of *Citrus* spp. leaves until the fourth-instar. From this instar, caterpillars gather on the main trunk of plants during the day. Pupae of *H. anchisiades capys* have a similar color as the substrate camouflaged with branches, leaves, mosses and lichens, possibly as a defense against natural enemies. Pupae with dark brown tegument resemble twigs or broken branches and the brown-gray ones can be confounded with lichens on the substrate whereas the greenish ones with mosses on branches, trunks and leaves (LEITE *et al.* 2010a).

Heraclides spp. were reported from USA to Bolivia, Paraguay and Argentina (JOHNSON & ROZYCKI 1986). In Brazil, *H. anchisiades capys* occurs in most regions where *Citrus* spp. are cultivated feeding on young and old leaves of this plant. Caterpillars of this insect eat the whole leaf from its margins in semicircular movements leaving the midrib intact (LEITE *et al.* 2010a).

Pupae of *H. anchisiades capys* could be used to mass rear natural enemies, including the generalist and gregarious endoparasitoid *Palmistichus elaeisis* Delvare & LaSalle, 1993 (Hymenoptera: Eulophidae) of urban areas and agroforest systems. This insect parasitizes *Citioica anthonilis* (Herrich-Schaffaefer, [1854]) (Lepidoptera: Saturniidae), *Methona themisto* (Hübner, 1818) (Lepidoptera: Nymphalidae) and *Thagona tibialis* (Walker, 1855) (Lepidoptera: Lymantriidae) pupae in the field and laboratory (TAVARES *et al.* 2011b, 2013a, 2013b).

The aim of this study was to evaluate the suitability of *H. anchisiades capys* pupae for breeding the parasitoid *P. elaeisis* in the laboratory.

Material and Methods

Twenty-five fourth-instar *H. anchisiades capys* caterpillars were collected on one *Citrus* sp. plant on the campus of the Federal University of Viçosa (UFV) in Viçosa, Minas Gerais State, Brazil ($20^{\circ}44' S$, $42^{\circ}50' W$, 650 m above sea level) in 2011. These insects were brought to the Laboratory of Biological Control of Insects (LCBI) of the

UFV and placed in rearing cages (32 cm long \times 30 cm wide \times 30 cm high) with branches of this plant until pupation in a room at $25 \pm 1^{\circ}C$, 12 h photoperiod and relative humidity of $75 \pm 5\%$. Branches of this plant were changed daily and the cage was cleaned twice a day to remove feces (LEITE *et al.* 2010a).

Twenty recent *H. anchisiades capys* pupae (average duration from fourth-instar to pupa, 20 days) were individualized in test tubes (14 cm long \times 2.2 cm diameter) with ten mated *P. elaeisis* females with a drop of honey on the side of the tube. *Palmistichus elaeisis* was obtained from a first-generation emerged from *T. tibialis* pupae collected on one *Terminalia catappa* L. (Combretaceae) tree at the campus of UFV (TAVARES *et al.* 2011b). Parasitoids remained for ten days in these tubes, according to methodology proposed for *P. elaeisis* parasitizing *Anticarsia gemmatalis* Hübner, 1818 (Lepidoptera: Noctuidae) and *Tenebrio molitor* L., 1758 (Coleoptera: Tenebrionidae) pupae (Figs 2A and 2B) in the laboratory (ZANUNCIO *et al.* 2008). Pupae were left in these tubes until emergence of parasitoids or adults of *H. anchisiades capys*.

Palmistichus elaeisis adults were sent to Dr. Christer Hansson of the Department of Biology of the University of Lund in Sölvvegatan, Lund, Sweden, where it was identified. Five *H. anchisiades capys* pupae were left in rearing cages until emergence of the lepidopteran, which were sent to the Department of Zoology of the Federal University of Paraná (UFPR) in Curitiba, Paraná State, Brazil, where the species was identified by Dr. Olaf Hermann Hendrik Mielke.

The duration of the life cycle (egg to adult), emergence rate, total individuals emerged per pupa, sex ratio, size of the body and longevity of *P. elaeisis* males and females emerged from *H. anchisiades capys* pupae were evaluated.

Results and Discussion

This is the first report of *P. elaeisis* parasitizing *H. anchisiades capys* pupae in the laboratory. Pupae of this pest were also parasitized by *Pedinopelt gravenstii* (Guérin-Ménéville, 1826) (Hymenoptera: Ichneumonidae) in the field with the emergence of one individual per pupa through a circular ventro-lateral hole in the area of the forewings (COSTA LIMA 1935; SILVA 1935; LEITE *et al.* 2010a).

The duration of the life cycle (egg to adult) of *P. elaeisis* was 21.15 ± 0.15 days. Adults of this parasitoid emerged from forty percent of *H. anchisiades capys* pupae, with an average of 323 ± 38 individuals emerged from each one and a sex ratio of 0.95 ± 0.02 . The total number of para-

sitoids emerged was 2,584 individuals. The size of the body and the head capsule (mm) of *P. elaeisis* female and male progeny were 1.92 ± 0.04 ; 0.52 ± 0.02 and 1.28 ± 0.04 ; 0.40 ± 0.02 , respectively. The average longevity was 23.72 ± 1.15 days for females and 30.1 ± 2.42 days for males of *P. elaeisis*. The number of individuals emerged per pupa was higher than those reported by other authors. Four *P. elaeisis* females per *T. molitor* pupa had 90.76% emergence and 70.07 ± 2.50 individuals per pupa (ZANUNCIO *et al.* 2008); six *P. elaeisis* females per *A. gemmatalis* pupa had 100% emergence and 110.20 ± 19.37 individuals per pupa (PEREIRA *et al.* 2010a), while 10 *T. pupivorus* females per *A. gemmatalis* pupa had 35% emergence and 241.9 ± 12.4 individuals per pupa (TAVARES *et al.* 2011a). On the other hand, each *Bombyx mori* L., 1758 (Lepidoptera: Bombycidae) pupa produced 550 *P. elaeisis* individuals, which was attributed to its greater size (PEREIRA *et al.* 2009).

Production of *P. elaeisis* from *H. anchisiades capys* pupae may allow for obtaining adults of this parasitoid for release in crops of *Citrus* spp., *Eucalyptus* spp. (Myrtaceae), *Triticum* spp. (Poaceae), *Gossypium* spp. (Malvaceae) and *Morus* spp. (Moraceae). This natural enemy parasitized *Thyrinteina arnobia arnobia* (Stoll, 1782) and *Thyrinteina leucoceraea* Rindge, 1961 (Lepidoptera: Geometridae) (PEREIRA *et al.* 2011), *A. gemmatalis* (PEREIRA *et al.* 2010b), *Pseudaleitia sequax* Franclemont, 1951, *Alabama argillacea* Hübner, 1818 (Lepidoptera: Noctuidae), *Dirphia moderata* Bouvier, 1919 (Lepidoptera: Saturniidae) and *Halysidota personata* Watson, 1980 (Lepidoptera: Arctiidae) (PEREIRA *et al.* 2008) pupae.

Heraclides anchisiades capys pupae can successfully rear *P. elaeisis* in the laboratory for release and the control of this and other pests in the field.

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