A Survey of *Itoplectis melanocephala* (Grav.) - A New Ichneumonid Endoparasite on Wax Moth *Galleria mellonella* L.

Mehmet Faruk GÜRBÜZ¹ Janko KOLAROV² Ayşegül BUNCUKÇU¹

¹Süleyman Demirel University, Faculty of Arts and Sciences, Department of Biology, 32260, Isparta, TURKEY, e-mail: mfg@fef.sdu.edu.tr

²University of Plovdiv, Faculty of Pedagogy, 24 Tsar Assen Str., 4000 Plovdiv, BULGARIA, e-mail: j.kolarov@pu.acad.bg

ABSTRACT

Itoplectis melanocephala (Gravenhorst, 1829) (Hymenoptera, Ichneumonidae, Pimplinae), a solitary idiobiont endoparasite, is redescribed. Investigations on the development of the parasite on the newly discovered host wax moth *Galleria mellonella* (Linnaeus, 1758) (Lepidoptera: Pyralidae) were made. Under the laboratory conditions longevity of *Itoplectis melanocephala* females is between 24 st -40 th days, while in males it is 20 st -30 th days. The egg productivity and the hatching of eggs between maximum 6 st -20 th days. Durations of male wasp egg to pupal periods last for 7 st -15 th days, while it is found between 13 st -18 th days for females. On average of adult (male:female) sex ratio is 2:1.

Key words: Itoplectis melanocephala (Grav.), parasite, Ichneumonidae, redescription, biology, Galleria mellonella (L.).

INTRODUCTION

Biological control has become an increasingly important pest management strategy due to an increased concern over the use of pesticides (Hu & Vinson, 1998). The use of parasitoids to control pests could be accelerated and would offer new opportunities for biological control if a wide variety of exotic and native species could be mass produced and released (Greany *et al.* 1984). Thus, *in vitro* culture of parasitoids is considered as one possible way to solve the problem (Greany, 1986; Greany *et al.*, 1984); Grenier *et al.*, 1994).

Ichneumonidae includes parasites mainly of insects and spiders; therefore they are potential biological control agents (Ueno, 1999). Artificial rearing may increase practicability of augmentation for widespread pest control. Biological control has the commonly had both pure and applied definitions and connotations (Huffaker & Dahlsten, 1999). *Itoplectis* spp. (Hymenoptera, Ichneumonidae, Pimplinae) are solitary endoparasitoids which attack mostly on Lepidopterous pupae (Townes, 1969). *Itoplectis* species play and appreciable role in regulating the numbers of agricultural and forest pests (Kasparyan, 1973).

The present work is on *Itoplectis melanocephala* as a biological control agents of *Galleria mellonella*. In vitro rearing of *Itoplectis melanocephala* on *Galleria mellonella* was carried out initially by us, which is given in detail in this work. The aim of this study was to investigate the biology of *Itoplectis melanocephala* on *Galleria mellonella* in laboratory conditions as a natural enemy.

In the present paper, a redescription of the species is proposed. The biological development of *Itoplectis melanocephala* on the wax moth host *Galleria mellonella* was exposed to laboratory conditions. Therefore, as a parasite of insect pests it may be a potential biological control agent.

MATERIALS AND METHODS

Itoplectis melanocephala was collected in TR Adana-Yumurtalık between 13-19. III.2007. Yumurtalık, which is situated in the Eastern Mediterranean, Turkey (37°47'N 38°39'E), covers an area of 0 up to 3 m. elevation above sea level. Yumurtalık Lagoons, which have different ecological habitat character, abundant vegetation and existence of the animal, are open laboratories for scientific research. Yumurtalık has been the Natural Reserve of Eastern Mediterranean in Turkey since 1994. *Astragalus subuliferus* and *Bupleurum polyactis* are two endemic species for Yumurtalık lagoon. Also *Pinus halepensis*, which covers about 54 hectares in Yumurtalık, is a single natural sample. In this place, there are *Myrtus communis, Erica manipuliflora, Pistacia lentiscus* and lots of lemur plants.

The material was obtained from grass-type plants by using a standard insect sweeping-net. The sample was brought to the laboratory and a colony was formed. It was reared on *G. mellonella* under laboratory conditions of $30\pm2^{\circ}$ C and 40% r.h.. The colony was maintained by cocoons containing larvae and prepupae of *Galleria mellonella*. *Itoplectis melanocephala* was cultured at a constant temperature of 20° C and a photoperiod of 12-h light; and 12-h dark. The experiments were repeated three times under the laboratory conditions of $30\pm2^{\circ}$ C, 55-60% r.h. and 12-h photoperiod. The adults of *Itoplectis melanocephala* were placed in a 25•25•25-cm cage containing cotton pieces with 50% honey solution.

The three pupaes were given daily to a copulated female of *Itoplectis melanocephala*. After one day, the pupaes were opened and it was counted how many eggs were left from parasitoid with a 'Nikon C-PS' stereomicroscope. This experiment gave us the egg production of the copulated female during its life. Then the eggs were put in % 0,1 saline (NaCI)-water solution to determination the ratio of hatching for one day. These experiments showed us the female of *Itoplectis melanocephala*'s egg production and hatching. On the other hand sex ratio was determined through experiment. To count the sex ratio, three pupaes were given again to the parasitoid. The next day, the pupaes were taken into a beaker which was closed with a patch. We waited about 13-20 days to observe whether they were male or female. These processes continued until the parasitoid died. Therefore the investigation of the biology of *Itoplectis melanocephala* was also completed. All data were subjected to analysis of variance (ANOVA), and results were considered statistically significant when P<0,05.

RESULTS AND DISCUSSION

Redescription

Itoplectis melanocephalus (Gravenhorst, 1829)

Many ichneumonids had been described only shortly, by using mainly the coloration of the body. In this paper we proposed more detailed redescription according to move up to date requirements.

Female: Front wing 9,0 - 9,4 mm., body 10,5 - 11,5 mm., ovipositor sheath 2,2 mm. long. Head strongly narrowed behind (Fig. 1). Flagellum with 24 segments, first segment 4,5 as long as wide. Occipital carina moderately strong, connected oral carina at distance from mandible base twice longer of malar space. Eye orbit strongly excavated of the antenna base (Fig. 2). Face moderately deep punctured, 1,4 as wide as high, rather flat centrally and ventral, somewhat raised medially dorsal, with an elongate tubercle laterally running parallel with the eye orbit. Malar space mat, 0,25 as long as the mandible base. Frons shine, transversally striated just before front ocellus. The distance between eye and lateral ocellus is almost as long as ocellus diameter.

Mesosoma short, only 1,3 as long as high. Epomia is present, notaulus is shallow. Epicnemal carina is not connecting front lateral margin of mesothorax, ending above middle of mesopleurum. Meso and metapleurum are with fine and dense punctures. Front wing with second recurrent vein (2m-Cu) vertical to parallel vein (Cu-1a). Nervulus is interstitial, nervellus is strongly reclival, intercepted far above the middle. Propodeum is coarse punctured, with strong pleural, apical half of basal and basal half of median longitudinal carinae. Middle part of apical carina is somewhat developed too, so combination of basal area and area superomedia is fully defined (Fig. 3). Propodeal spiracle is distinctly closer to pleural carina. Legs are stout, hind femur is only 3,0 as long as wide. Apical hind tarsal segment is enlarged, more than 4,5 as long as fourth segment. Front tarsal claws are simple, without any basal teeth.

Metasomal tergites are centrally with coarse punctures tending to run together and form irregular longitudinal striation. First tergite has strong lateral and dorsal carinae, II – V tergites with strong lateral swellings (Fig. 4). Ovipositor sheath 0,83 as long as hind tibia.

Body black. Flagellar segments except basal and apical margin, palpi, legs except base of front coxa and sometimes last tarsal segment of hind legs apically, postscutellum, metapleurum, base of pterostigma, propodeum and metasoma orange, tegula dark brown.

Male: Front wing 6,7 mm., body 8,7 mm. long. Notaulus more distinct. Orange colouration more intensive, also occupying scutellum, scapus from below, hind lower corner of mesopleurum and hind part of mesosternum. Metasoma apically (after seventh tergite) black.

Material examined: TR, Adana, Yumurtalık, Halep Çamlığı, 0-3m., 13-19.III.2007, 1 female; reared material in laboratory from *Galleria mellonella* L. parasitising by the same female, 11 males and 17 females; Egypt, Tourach, 1 female; Iran, Isfahan, 1590 m, 24.X.2001, 1 female; Mazandaran, Amol, VIII.2005, 1 male and 6 females; Guilan, Amlash, X.2004, 1 female; Minoo-Dasht, Golestan, VII.2003, 1 female; Ilam, 19.VIII.2001, 1 female.



Figs. 1- 4. *Itoplectis melanocephala.* 1. Head from above, 2. Head in front, 3. Propodeum from above, 4. Metasoma in lateral view.

A key for determination of the Palearctic species of *Itoplectis* is given by Kasparyan (1973). *Itoplectis melanocephala* is easily distinguished from the other Palearctic species by the following characters: Front tarsal claws simple, without basal teeth; last tarsal segment 4-5 times as long as forth; metasoma entirely orange coloured. It is more close to *Itoplectis naranyae* Ashmead, distributed in Eastern Palearctic region ? and Mexico. The mean distinguishing characters among both species was discussed by Perkins (1957).

Host: Aletia impura Hb. (Noctuidae), Chilo phragmitellus Hb. (Crambidae), Depressaria daucella Denn. and Schiff., Depressaria pastinacella Dup. (Depressariidae), Leucania obsoleta Hb. (Noctuidae), Monda rogenhoferi (Psychidae), Ostrinia nubilalis Hb. (Pyraustidae).

Host plant: Peucedanum palustre, Phragmites, Fitton et al. (1988).

Distribution of Turkey: Edirne, Hadımağa (Yurtcan & Beyarslan, 2005) and Adana, Yumurtalık.

General distribution: Europe including British Islands, Turkey, Egypt, Kenya, Iran, Kazakhstan, Tajikistan, Uzbekistan, Chitinsk region of Russia, Mongolia and China.

According to classification proposed by Taglianti *et al.* (1999) the species belongs to the groups of species in Central Asian - European - Mediterranean chorotypes.

Biology

Itoplectis melanocephala is an idiobiont solitary pupae endoparasite on *Galleria mellonella* which is very suitable as a laboratory host.

Longevity in *Itoplectis melanocephala* females is between 24 st -40 th days (mean 32 ± 11,31), while it is 20 st -30 th days in males (mean 25 ± 7,07). Longer life span of females than that of males is common in parasites (Ueno & Tanaka, 1994). Durations of male wasp egg to pupal periods last for 7 st -15 th days (mean 11 ± 5,66), while it was found to be between 13 st -18 th days (mean 15,5 ± 3,54) for females. This is probably because many parasites lay seminal eggs on large hosts while they do not lay seminal eggs on small hosts. Copulation takes place between 70-160 seconds (mean 115 ± 63,64). Hatched males and females copulate in the first 3-4 minutes. However, only one egg develops for each host. The egg productivity of *Itoplectis melanocephala*, the hatching of eggs, pupae, and sex ratios are presented in Table 1. It was found that there is no statistical difference in the fertility of egg of copulated females between 1st - 15th days and the days between 16 st - 20th are the most fertile period with 9,25 ± 4,59 average. Between 21st - 25th days it (in the average) decreases nearly to half, and between 26th - 30th days it is the least fertile period. The maximum hatching rate of copulated females are found between 6st -10th days and between 16st - 20th days.

Soliter *Itoplectis melanocephala* reached maximal sex ratio of female and male 1:2 between 6st -15th days. This ratio decreased while life-span was extended. It was found that there is no statistical difference in the fertility of the egg of virgin females between 6st -20th days. The most fertile period is between 11st -15th days. Between 21st - 35th days it (in the average) decreases.

Itoplectis melanocephala's host feeding behaviour is observed. Oviposition is not observed when parasitoid used the host as a prey. *Itoplectis melanocephala* is a synoviogenic parasitod. Maternal age has a significant impact on levels of offspring production and sex ratio. The maximum fecundity of offspring production and sex ratio is the first tree weeks (Table 1). *Agrothereutes hospes* Tschek (Ichneumonidae, Cryptinae) hosts that are fed upon are killed as a result of host feeding, preventing the female from using the same host individual for both oviposition and for host feeding (Gurbuz *et al.*, 2006). Maternal age is of great importance on offspring fertility and sex ratio. It's during between 2-4 weeks when fertility makes a peak, and because of this, it can be used as a biological control agent.

This study has revealed that *Itoplectis melanocephala*, which is to be an important biological agent accepted *Galleria mellonella* as a laboratory host. For this reason, *Itoplectis melanocephala* can be a candidate as a biological agent against harmful lepidopterous species.

ACKNOWLEDGEMENTS

This work was supported by the TBAG- U/168(106T189) and we would like to express our thanks to TUBITAK.

Table 1. Itoplectis melanocephala's virgins and adult copulation females average of five days eggs productivity, hatching of eggs and the sex ratio.

	Copulatio	Copulation females	Sex ratio of copulation females	lation females	Virç	Virgins
Adult Age (days)	Eggs production (mean*±SH².)	Hatching of eggs (mean*±SH².)	The ratio of female (mean*±SH².)	The ratio of male (mean*±SH².) ^y	Eggs production (mean*±SH².) ^y	Hatching of eggs (mean*±SH².)
1-5	6,60±4,47 bc	5,80±2,37 bc	0,47±0,67 b	0,33±0,49 ab	6,0±4,60 b	5,20±1,65 b
6-10	8,20±4,53 bc	7,20±3,84 c	0,33±0,49 ab	0,67±0,62 b	12,54±7,97 c	9,67±7,53 c
11-15	7,27±3,92 bc	6,27±2,74 bc	0,27±,0,60 ab	0,53±0,92 ab	13,93±6,58 c	11,20±6,61 c
16-20	9,27±5,79 c	7,27±3,91 c	0,07±0,26 a	0,33±0,62 ab	13,47±5,63 c	10,00±5,49 c
21-25	5,07±5,83 b	4,0±3,58 b	0,07±0,26 a	0,27±0,59 ab	9,80±3,47 bc	7,80±2,68 bc
26-30	1,10±2,09 a	0,6±0,79 a	0±0 a	0,07±0,26 a	1,8±2,68 a	1,60±1,41 a
31-35					2,30±3,25 a	0,70±0,99 a

* The average of three repeats

z. Standard error.

y. The values that are shown with the same letter in the same column are equal to other (P<0,05)

REFERENCES

- Fitton, M. G., Shaw, M. R., Gauld, I. D., 1988, Pimplinae Ichneumon-flies. Hymenoptera, Ichneumonidae (Pimplinae). *In:* Handbook for the Identification of British Insects, 7: 1-110.
- Greany, P. D., 1986, *In vitro* culture of hymenopterous larval endoparasitoids. *Journal of Insect Physiology*, 32: 409–419.
- Greany, P., Vinson, S. B., Lewis, W. J., 1984, Insect parasitoid: finding new opportunities for biological control. *BioScience*, 34: 690–695.
- Grenier, S., Greany, P. D., Cohen, A. C., 1994, Potential for mass release of insect parasitoids and predators through development of artificial culture techniques. In: Rosen, D., Bennett, F.D., Capinera, J.L. (Eds.). Pest Management in the Subtropics. Biological Control - a Florida Perspective. Intercept Ltd., Andover, England, 181–205.
- Gurbuz, M. F., Kolarov, J., Aksoylar, M. Y., Akdura, N., 2006, A Survey of the *Agrothereutes hospes*, an ectoparasite on wax moth *Galleria mellonella. Journal of Pest Science*, 79: 31-34.
- Hu, J. S., Vinson, S. B., 1998, The in vitro development from egg to prepupa of *Campoletis sonorensis* (Hymenoptera: Ichneumonidae) in an artificial medium: importance of physical factors. *Journal of Insect Physiology*, 44: 455–461.
- Huffaker, C. B. and Dahlsten, D.L., 1999, *Scope and Significance of Biological Control. In:* Bellows, T. S., Fisher, W. T. (Eds.). Handbook of Biological Control. Academic, USA, 1.
- Kasparyan, D. R., 1973, A review of the Palearctic Ichneumonids of the tribe Pimplini (Hymenoptera, Ichneumonidae). The genera *Itoplectis* Foerst. and *Apechthis* Foerst. *Entomologicheskoye Obozreniye*, 52(3): 665-681.
- Perkins, J. F., 1957, Notes on some Eurasian "Itoplectis" with description of new species (Hym., Ichneumonidae). *Mitteilungen der Schweizerischen Entomologischen Gesellschaft*, 30(4): 323-326.
- Taglianti, A. V., Audisio, P. A., Biondi, M., Bologna, M. A., Carpaneto, G. M., de Biase, A., Fattorini, S., Piattella, E., Sindaco, R., Venchi, A., Zapparoli, M., 1999, A proposal for a chorotype classification of the Near East fauna, in the framework of the Western Palaearctic region. *Biogeographia*, 20: 31–59.
- Townes, H., 1969, *The genera of Ichneumonidae*, Part 1. Memoirs of the American Entomological Institute., 11-300.
- Ueno, T., 1999, Adult size and reproduction in the ectoparasite *Agrothereutes lonceolatus* Walker (Hym., Ichneumonidae). *Journal of Applied Entomology*, 123: 357-361.
- Ueno, T., Tanaka, T., 1994, Comparative biology of six polyphagous solitary pupal endoparasite (Hymenoptera: Ichneumonidae) Differential host suitability and sex allocation. *Annals of the Entomological Society of America*, 87(5): 592-598.
- Yurtcan, M., Beyarslan, A., 2005, Polysphinctini and Pimplini (Hymenoptera: Ichneumonidae: Pimplinae) from the Thrace region of Turkey. *Fragmenta Faunistica*, 48(1): 63-72.

Received: September 18, 2008 Accepted: June 10, 2009