

## FIRST RECORD OF *HEXAMERMIS* CF. *ALBICANS* (SIEBOLD, 1848) (NEMATODA: MERMITHIDAE) INFECTING LEPIDOPTERAN LARVAE FROM IRAN

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**Summary.** During the course of entomological investigations on natural enemies of forest pests in the Arasbaran area, northwest of Iran, a mermithid nematode, *Hexameris* cf. *albicans* was found parasitizing the abdominal cavity of larvae of two lepidopterans, *Euproctis chrysorrhoea* and *Lymantria dispar* (Lymantriidae). Infection occurs in the early instars of the host larvae. Parasitism continues until the late instar of the larval host, when the nematode emerges and remains in a post-parasitic free-living stage until it moults into an adult. The mean body length of the parasite ranges from about 14 to 22 cm. One to four mermithids were found per host. About 34% of *E. chrysorrhoea* and 57% of *L. dispar* larvae were found to be parasitized by the nematodes. However, the frequency of the parasite varied in different areas. The infection was more frequent in dense as opposed to more sparsely forested regions. This is the first record of *Hexameris* cf. *albicans* (Siebold, 1854) from Iran.

**Keywords:** Entomogenous nematode, *Euproctis chrysorrhoea*, *Lymantria dispar*.

Entomogenous nematodes are widespread in all insect orders, including Lepidoptera (Drea *et al.*, 1977; Luckmann and Poinar, 2003). Mermithidae is a family of nematodes that are parasitic in many kinds of insects (Nielsen, 2004; Platzer, 1981), spiders (Vandergast and Roderick, 2003), leeches, crustaceans, nematodes, and other invertebrates throughout the world. The most common hosts of mermithids are insects and at least fifteen different orders have been reported to serve as hosts (Kaiser, 1991). Parasitism by a mermithid is fatal to the host. Mermithid larvae are usually found in the body cavities of all stages of susceptible insect species. The parasites nourish themselves parenterally from the insect's haemolymph, storing resources for the free living stage, which does not feed. It is not unusual to find mermithids of up to 20-50 cm body length inside insects. Epizootics of insects, caused by mermithid parasitism, occur and population levels of blackflies, mosquitoes, chironomids, grasshoppers, walking sticks, ants, certain lepidopterans, and other insects are held down by these self-perpetuating biological control agents (Kaiser, 1991). According to Poinar (1975), nearly 50% of all papers on the nematodes parasitic on insects refer to Coleoptera as hosts. Several species of mermithid nematodes, especially *Hexameris* spp., parasitize Lepidoptera (Wouts, 1984). *Hexameris* cf. *albicans* (Siebold, 1854) species include very different sized parasites and they have been found parasitic in many different insects as well as in some slugs and snails (Kaiser, 1977).

To assess the role of mermithids on lepidopteran pests in Iran, a sampling was undertaken in the Aras-

baran forest, Kaleibar Township, East Azarbaijan. Therefore, at the beginning of 2007 and 2008, fifth instar larvae of *Euproctis chrysorrhoea* (L.) and *Lymantria dispar* (L.) were collected by hand from various localities of five oak-dominated forests. The larvae were dissected under a stereomicroscope and checked for parasitism by mermithids and the number and location of infections were determined. In late July of both years additional host specimens of last larval instar were collected and placed, together with oak leaves, in a polystyrene box. Emerging post-parasitic mermithids were collected from the boxes daily, transferred to dishes with moist soil and maintained at 15 °C in the darkness for 3-4 weeks to promote moulting of the nematode and to allow identification at sexual maturity. Also, some mature mermithids were collected by digging into the soil in the vicinity of contaminated trees. Specimens were fixed in hot 4% formaldehyde and, for microscopic observations, were transferred gradually into pure glycerine before mounting on microscope slides.

*Hexameris* cf. *albicans* was recovered from larvae of *E. chrysorrhoea* and *L. dispar* collected from various localities. The infection rate varied in different areas. Infection was much higher in dense regions of the forest compared with sparse regions. Lepidopteran larvae from five areas [Ainaloo (n = 180), Makidi (n = 165), Kerenghan (n = 135), Naposhteh (n = 180) and Andabil (n = 190)] were parasitized with 39.5%, 33%, 30%, 49% and 48%, respectively. All infected lepidopteran larvae died after emergence of the nematodes. Among the two lepidopteran species observed, the highest infection rate (57%) was observed in fifth and sixth instar larvae of *L. dispar* (Table I). The rate of mermithid parasitism in *E. chrysorrhoea* and *L. dispar* larvae (dissected larvae and larvae from which nematodes had emerged)

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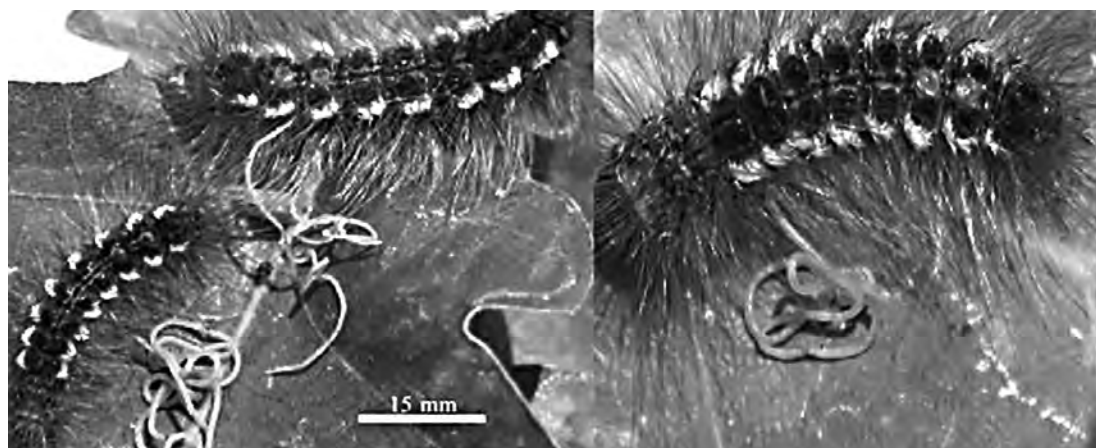


Fig. 1. Mermithid nematode (*Hexameris* cf. *albicans*) emerging from specimens of *Euproctis chrysoorrhoea* larvae collected from Arasbaran forests, Iran.

varied between 31-38.3% and 48.2-57%, respectively (Table I). One to four mermithid nematodes were found per host larva. Differences in parasite load in fourth and fifth instar larvae were also observed: 69% of the fifth instar larvae and 10% of the fourth instar larvae were parasitized by more than one nematode.

The parasites were generally found in the abdominal and posterior part of the haemocoel of the caterpillars. No encapsulation of parasites was observed in *E. chrysoorrhoea* or *L. dispar* larvae.

The body length of the mermithids increased significantly through the stages of the host's development. The largest mermithid (body length 220 mm, body width 0.27 mm) was found in a last instar caterpillar of *L. dispar*.

Mermithids from lepidopterans have frequently been recorded from various parts of the world (Poinar, 1975; Wouts, 1984; Kaiser, 1991). Usually they are found as parasites in caterpillar larvae. High infection rates in a field population were found by Kaburaki and Imamura (1932), with 76% of the rice borer, *Chilo simplex*, infected in Japan. Artyukhovsky (1953) reported that up to 60% of the gypsy moth caterpillars were parasitized by *H. albicans* south of Moscow. In 1953, Polozhentsev and Artyukhovsky (1953) reported infection of *H. albicans* in *L. dispar*. Poinar (1975) finally listed more than 60 species of Lepidoptera as hosts for *H. albicans*. Nick-

le and Grijpma (1974) found up to 25% of the shoot borers, *Hypsipyla grandella* (Zeller), infected by *H. albicans* at the end of the wet season in Costa Rica. They listed 70 other species of Lepidoptera that are parasitized by mermithids. Heavy parasitism (87.5%) of larvae of *E. chrysoorrhoea* by *H. albicans* was reported in Trabzon forest in Turkey in 1997 (Damirbag and Yaman, 1999). *Hexameris albicans* was reported from *E. chrysoorrhoea* by von Linstow in 1898 (Poinar, 1975). It was recovered from larvae of *L. dispar* and *Stilpnotia salicis* (L.) collected from various localities in Austria and Germany (Drea *et al.*, 1977). Also, infection of the lymantrid pests with different species of *Hexameris* in northern Iran and neighbouring countries of the former Soviet Union was reported by Sultanov (1977).

In our study, results similar to those of neighboring countries were obtained. Therefore, more investigations may lead to the identification of further hosts and/or species of mermithids. More work will be needed to evaluate the potential of mermithids as bio-control agents. Unfortunately, most entomologists preserve insect specimens in 75% ethanol and thus mermithids are likewise preserved in their immature stages, at which species identification is not possible (Poinar, 1975).

*Hexameris* nematodes often emerge from the last insect larval stage as a so called post-parasitic larva. The violence of this emergence usually leads to the death of

Table I. Prevalence of mermithid parasites in lepidopteran larvae collected from Arasbaran forest.

Insect species	Larval instar	Examined larvae (n)	Dissected larvae (containing mermithid)	Larvae parasitized (pierced) by emerging nematodes	Parasitism (%)	Total parasitism (%)
<i>Euproctis chrysoorrhoea</i>	4	400	124	-	31	34.6
	5	360	-	138	38.3	
<i>Lymantria dispar</i>	4	450	220	-	48.8	52.9
	5, 6	400	-	231	57	

the host. Thus, unlike some other insect parasites, infection rates correspond to mortality rates, similar to parasitoids. The mermithid was recovered from larval instars collected from all field sampled and in both high and low host density populations. The majority of the nematodes emerged from *L. dispar* larvae collected as fifth and sixth larval instars.

*Hexameris albicans* has been reported as a parasite of many different hosts throughout the world (Kaiser, 1977, 1991; Poinar, 1975) including slugs, beetles and lepidopterans. Although the morphology of these nematode populations is very similar, their overall size differs and we hypothesize that a species complex may be present. Genetic analysis of different populations parasitizing lepidopterans from Iran and beetles from Austria are in progress and should help to clarify this issue. *Hexameris albicans* was also reported from *E. chrysorrhoea* by von Linstow in 1898 (in Poinar, 1975).

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