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An investigation on endohelminth parasite fauna of *Cobitis simplicispinna* Hanko, 1924 from Asia Minor

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Abstract

In this study, endohelminth parasite species were investigated in 49 *Cobitis simplicispinna* from Pinarbaşı spring water, Asia Minor between September 2009 and October 2011. Two endohelminth parasite species were determined in intestinal lumen of the host fishes. 116 number *Crowcrocoecum skrjabini* (%58, 4.0±3.7) and 198 Acanthocephala samples (%86, 4.6±3.5) were found. Among these species, *Crowcrocoecum skrjabini* is a new records for endohelminth parasite fauna of *Cobitis simplicispinna* from Asia Minor. Thus, a new locality has been added to geographical distributions of the parasite species. It was contributed to revealing the endohelminth parasitic fauna of *Cobitis simplicispinna* from the study area.

Keywords: Acanthocephala, *Cobitis simplicispinna*, *Crowcrocoecum skrjabini*

1. Introduction

Fish species in the Cobitidae family are common populations in freshwater systems in Eurasia [1-7]. Species in the Cobitidae family prefer stagnant or slow flowing and eutrophic waters as habitats [8]. These types of habitats are suitable environments for parasitic organisms to survive or complete their development. There were various studies on parasitic infections belonging to the *Cobitis* genus in Cobitidae family: *Allocreadium transversale* species was identified in the intestine of *Cobitis taenia* [9]. Bauer [10] compiled the protozoan and metazoan parasite species of *Cobitis* genus. In *Cobitis elongatoides*, *Ligula colymbi* was detected at high infection density by Halačka *et al.* [11]. Six trematode species (*Diplostomum* sp., *Echinostomatidae* sp., *Tylodelphys clavata*, *Posthodiplostomum cuticola*, *Metorchis xanthosomus*, *Allocreadium transversale*) and one nematode species (*Rhabdochona adolescence*) were recorded in individuals from *Cobitis elongatoides* and *Cobitis taenia*. Scholz *et al.* [12] found the *Neogryporhynchus cheilancristrotus* species in *Cobitis taenia*. Zrnčić *et al.* [13] found *Gyrodactylus cobitis* from monogenic trematodes and *Ligula colymbi* from sestoada in three *Cobitis* species (*C.elongata*, *C.elongatoides*, *C.bilineata*). In addition, *Gyrodactylus cobitis* species was recorded on gills and fins of *Cobitis simplicispinna* [14].

The aim of this study was to determine the endohelminth parasite fauna of *Cobitis simplicispinna* from Pinarbaşı Çıldırım water source, Emirdağ-Afyonkarahisar, Asia Minor. As a result of the study, it will contribute to the determination of endohelminth fauna of *Cobitis simplicispinna*. With the data to be obtained, a new locality will be added to the distribution of the parasitic species identified from the host fish samples.

2. Materials and Methods

Samples of *Cobitis simplicispinna* were caught from Pinarbaşı Çıldırım water source, Emirdağ, Afyonkarahisar, Asia Minor between September 2009 and October 2011 using fish landing nets. Fish samples were transported to the Research Laboratory in containers containing water. They were examined within 24 to 48 hours and maintained in aerated aquarium tanks during this time.

During the examination process, first of all, the total lengths of the fish samples to be examined in terms of parasitology were noted. Species determination of the fish samples was carried out according to Geldiay and Balık [8]. For endohelminth parasite examination eyes, body cavity, gonads, liver, spleen, mesentery and intestinal lumen of each fish sample were examined with the help of stereo dissecting microscope.

If parasites found in fish samples, the location and number of parasites were recorded separately for each fish (Fig 1). Parasites in *Cobitis simplicispinna* were fixed with dilute formol (1/4000) and then transferred to Mayer's hematoxylin dye medium and then to alcohol series (35, 50, 70%). It was then made into a permanent preparation with the help of glycerin-gelatin. Bykhovskaya-Pavlovskaya^[15] was used to describe the parasites and species. Photographs of the parasites were taken under an Olympus BX60 light microscope with the aid of a Tescom micropublisher 3.3 RTV camera.



Fig 1: sample of *cobitis simplicispinna* caught from study area (original)

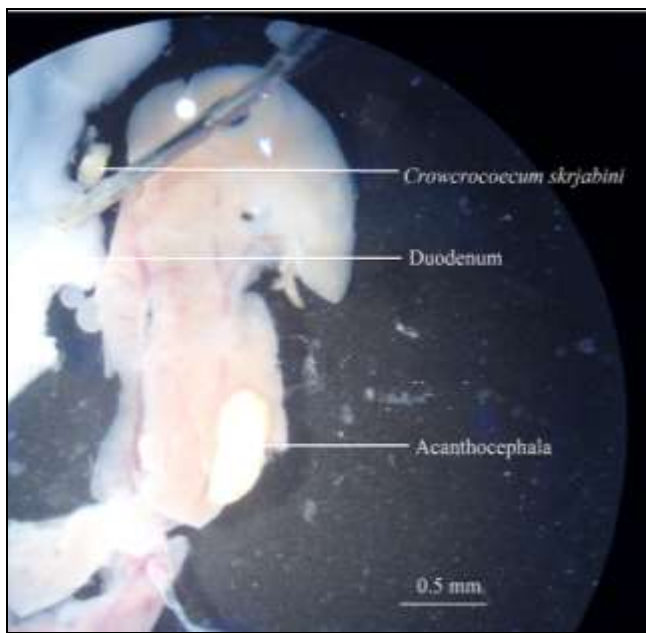


Fig 2: Endohelminth specimens in intestinal lumen of *C. simplicispinna*

3. Results

In this study, 49 *Cobitis simplicispinna* specimens were examined in terms of endohelminth parasite fauna with the help of dissecting microscope in eye, body cavity, digestive tube, gonads, liver, mesentery and intestinal lumen. As a result of the research, *Crowcrocoecum skrjabini* Ivanitskii, 1928 and *Acanthocephala* specimens were found in intestinal lumen of *Cobitis simplicispinna* (Fig 2). Of the species, *Crowcrocoecum skrjabini* was determined to be a new records for the endohelminth fauna of Asia Minor.

3.1. Systematic, anatomical features of the species *Crowcrocoecum skrjabini*

Phylum: Platyhelminthes

Classis: Trematoda Rudolphi, 1808

Order: Fasciolata Skryabin and Schul'ts, 1937

Family: Opecoelidae Ozaki, 1925

Genus: *Crowcrocoecum* Skryabin and Koval, 1956

Species: *Crowcrocoecum skrjabini* Ivanitskii, 1928

Synonyms: *Coitocaecum skrjabini* Ivanitskii, 1928

C. macrostomum Pigulevskii, 1931

C. ovatum Pigulevskii, 1931

The body length of *Crowcrocoecum skrjabini* Ivanitskii, 1928 was measured as 2235 μm (1750-3000 μm), body width of 814 μm (690-1000 μm), with a thick round body and flat cuticle. Oral sucker at the anterior terminal was defined as 247 μm (220-250 μm) long and 227 μm (217-260 μm) wide. The length of the pharynx was measured as 113 μm (98-137 μm) and the width as 134 μm (120-162 μm). It was observed that the two intestinal branches in the form of cylindrical ceceums merged with each other at the posterior subterminal and ended in a ring. It was determined that two testicles were lined up one after the other in the area between the intestinal ceceums at the posterior end of the body. The length of the anterior testis was 242 μm (237-250 μm) and the width was 225 μm (225 μm), and the length of the posterior testis was measured as 296 μm (290-300 μm). Sperm sac (seminal vesicul) and bursa (bursa genitalis) were seen anterior to the ventral sucker. The length of the pretesticular ovary was recorded as 213 μm (182-212 μm), and its width was recorded as 156 μm (125-175 μm). A short uterus was found anterior to the testis. Vitellus glands were noted to the posterior terminal of the body beyond the intestinal branches in the posterior subterminal. The eggs, located in sparsely rows in the uterus, were large and measured 81 μm (72-93 μm) in length and 58 μm (52-65 μm) in width (Fig 3).

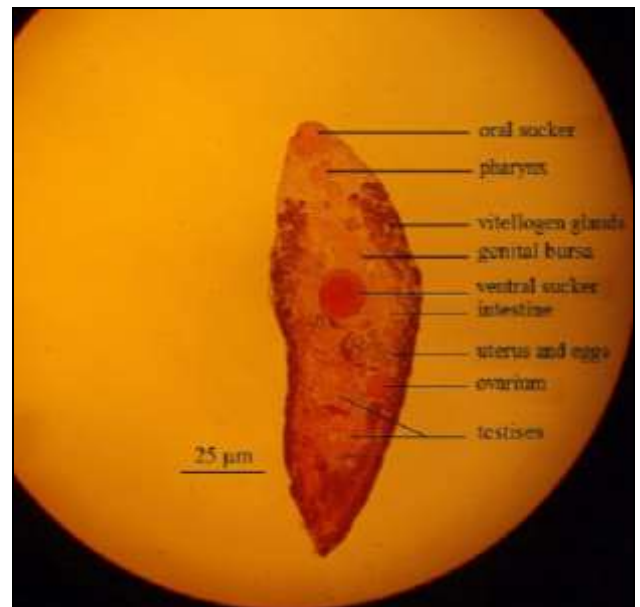


Fig 3: Anatomical structure in *Crowcrocoecum skrjabini* (original)

3.2. Systematic, anatomical features of the *Acanthocephala*

Phylum: Acanthocephala Koelreuter, 1771

The body is cylindrical and of medium size, with males measuring 14 mm (12–18) long and 2.1 mm (1.4–2.3) wide. The body is covered with a flat cuticle. The body is orange or yellowish. The bulb-shaped proboscis equipped with hooks that can go in and out from the anterior side of the body is 430 (410–537) μm in length and 317–337 (324) μm in width.

Proboscis is connected to the body with a very long cylindrical neck. On the other hand, more research is needed on the anatomical, morphological and genetic characteristics of mature individuals to determine the taxonomic positions of the specimens (Fig 4).

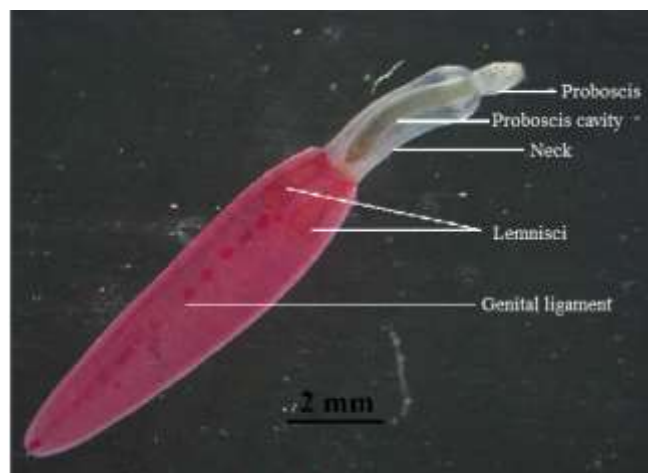


Fig 4: General structure in an Acanthocephala specimen (original)

3.3 Infection Data

In 49 *Cobitis simplicispinna* samples examined within the scope of the research, samples from *Crowcrocoecum skrjabini* from Trematoda and Acanthocephala taxon were found in intestinal lumen as endohelminth fauna. *Crowcrocoecum skrjabini* was found for the first time with in the study from Asia Minor. Therefore, the species *Crowcrocoecum skrjabini* is a new records for the Asia Minor. The infection cases of these endohelminths were given in Table 1.

Table 1: Infection values of endohelminth parasites found in *Cobitis simplicispinna*, Pinarbaşı cildirin water. N: number of infected fish, prevalence of infection (%), M: minimum-maximum number of parasites, mean density, and standard deviation

Parasite tan	Microhabitat	N & (%)	M& (X±S.D.)
<i>Crowcrocoecum slayabini</i>	Intestinal lumen	29 (58)	1-17 (4,0±3,7)
<i>Acanthocephala specimens</i>	Intestinal lumen	43 (86)	1-16 (4,6±3,5)

4. Discussion

In the study, 2 taxa were determined in *Cobitis simplicispinna* from Pinarbaşı Çıldırım water source. The first taxon, *Crowcrocoecum skarjabini*, belongs to the Platyhelminthes, as its body is flattened in the dorso-ventral direction and has bilateral symmetry. *Crowcrocoecum skarjabini* is in the class of Trematoda because of its non-segmented body, mouth sucker in the anterior terminal of their body and abdominal sucker in the median area. Individuals belonging to the second taxon, Acanthocephala, were characteristic with having hooked bothrium^[15].

Fish of the family Cobitidae prefer stagnant or slow flowing and eutrophic waters rich in invertebrate fauna^[8]. These types of habitats are suitable environments for parasites to survive or complete their development. Studies on parasitic fauna of fish species such as *Cobitis*, which are not economically important, are scarce. The main reason for this is the difficulties in catching fish species that are not economically important and their low density^[5]. Various studies were conducted on parasitic fauna diversity of fish in the cobitidae taxon, including *Cobitis simplicispinna*.^[10, 16-27]

Mature trematodes such as *Sphaerostomum bramea* and *S.globiporum* in fish of Cobitidae taxon live in the intestinal

lumen. According to Moravec^[23], these trematodes are not cobitid-specific parasites, but are also found in other fish species such as *Tinca*, *Cyprinus*, *Carassius*^[25]. On the other hand, *Allocreadium transversale* and *Crowcrocoecum skrjabini* are parasitic species specific to fish in the cobitidae family^[5]. The recording of the *Crowcrocoecum skrjabini* species in *Cobitis simplicispinna* individuals from the present research area supports this view.

Metacercariae stage trematodes found in cobitid fish are grouped under four families: Diplostomatidae, Clinostomatidae, Heterophyidae, Echinostomatidae. Among them, *Posthodiplostomum cuticola*, a metacercaria larva of Diplostomatidae, parasitizes in the skin, *Diplostomum spathaceum* in eye, *Tylodelphys cavlata* in muscle tissue and *T.cranicaria* in brain^[5] and *Tylodelphys* sp. in some cyprinid fishes^[28]. Mature individuals of this specimens are seen in fish-eating birds and rarely in mammals. The first three of the metacercaria are located in a wide range of hosts, including cobitids^[15]. However, no trematode species from the metacercaria stage were recorded in *Cobitis simplicispinna* individuals from the study area.

As a result, samples of 2 endohelminth parasite taxa were found in 49 *Cobitis simplicispinna*. Of these, *Crowcrocoecum skrjabini* is a new records for *Cobitis simplicispinna* parasite fauna from Asia Minor. The other parasitic group, Acanthocephala more research is needed on the anatomical, morphological and genetic characteristics of mature individuals to determine the taxonomic positions.

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