FIRST RECORD OF *ACANTHOCEPHALUS LUCII* (MÜLLER, 1776) LÜHE, 1911 (PALEACANTHOCEPHALA: ECHINORHYNCHIDAE) IN MINNOW (*PHOXINUS LUMAIREUL* LINNAEUS, 1758) FROM BELČIŠTA WETLAND, N. MACEDONIA

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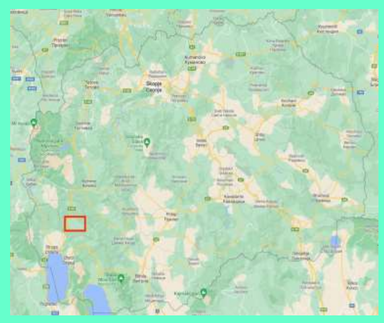

Abstract. The representatives of the phylum Acanthocephala are of great importance in fish pathology. *Acanthocephalus lucii* (Müller, 1776) Lühe, 1911 (Paleacanthocephala: Echinorhynchidae) is a common pathogenic acanthocephalan parasite in many fresh water predatory fish or those that feed on large benthos, in Euroasia. In our study,a total of 33 specimens of 3 cyprinid fish *(Squalius squalus, Phoxinus lumaireul* and *Pelasgus minutus)* from Belčišta wetland (southwest N. Macedonia) were subjected to a parasitological investigation, by seasons. Only fresh fish were subjected to routine identification, dissection, and observation methods. Parasite identification was performed morphologically, based on the character of proboscis with hooks and the reproductive system, using referent key for determination. One specimen of *Acanthocephalus lucii* was found in autumn, in intestines of minnow (*Phoxinus lumaireul* Linnaeus,1758). This parasite species would lead to impairing of fish health status.

The record of *Acanthocephalus lucii* in minnow in the present study is considered as the first in N. Macedonia. At the same time, minnow represents a new host for this parasite worldwide. This emphasizes the danger of the spread of parasites through stocking with newly introduced fish species.

Keywords: *Acanthocephalus lucii*, minnow, Belčišta wetland, first record

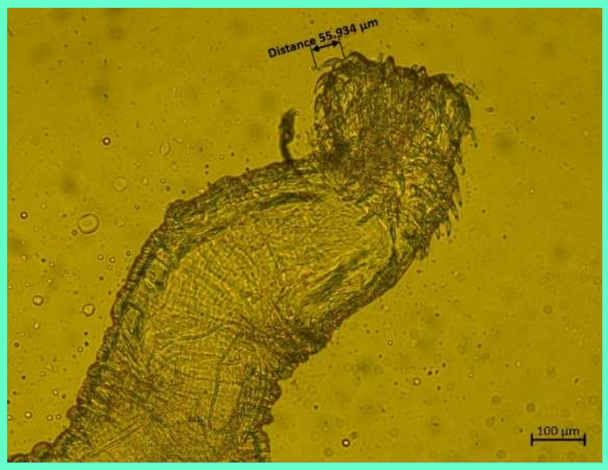

Fig. 3. *Acanthocephalus lucii* from minnow (*Phoxinus lumaireul)* from Belčišta wetland - proboscis of a female worm with hooks – original photo.

INTRODUCTION

Belčišta wetland (also called Sini Viroj) is located in the municipality of Debrca, below Ilinska Mountain, at an altitude of about 767 m. It is a remnant of the former Desaret Lake, which flooded the Debrca valley in the Pliocene. The wetlandis fed with 11 karstic springs, known as Sini Viroj, which originate from the springs north and northeast of the village of Novo Selo, in the direction of the village of Belčišta. The surface of the wetland covers around 400 hectares. There are also several wetland lakes in the wetland, 3 of which are larger: Belčiški Sin Vir, Novoselski Sin Vir 1 and Novoselski Sin Vir 2 and Sino Duvlo.With the retreat of the Desaret Lake along the Sateska River, numerous endemic species of plant and animal life continued to exist in the waters of the wetland. Belčišta wetland is the largest and one of the most important wetlands in N. Macedonia, with flooded forests and wet pastures, because it is well preserved. Specific is also due to different types of wetland habitats, especially flooded alder forests. There is a great diversity of flora and fauna. In this water ecosystem, according to research, a total of 55 different plant species and seven different fish species have been recorded. There are also 9 representatives of birds, 9 mammals, representatives of 9 amphibians and reptiles, as well as 14 representatives of invertebrates. Belčišta wetlandis part of the Emerald National Network of the Republic N. Macedonia, and is proposed to be nominated as a Natura 2000 location.

Acanthocephala (spiny-headed worms or thorn-headed worms) are necrotrophic worms that live as adults exclusively within the small intestines. They have a retractable proboscis armed with spines that is inserted into the mucosa. They have separate sexes and lack of circulatory, respiratory and digestive systems.

They have an indirect life cycle, which utilizes an arthropod intermediate host. Various decapods and other crustaceans serve as intermediate hosts for those with aquatic life cycles. The most common intermediate host for fish acanthocephala are amphipod crustaceans of the genus *Gammarus* or isopods of the genus *Asselus.* Within the arthropod, an acanthor hatches from the egg and penetrates the gut wall and enter the haemocoele. There it develops into an acanthella, in which the body wall and rudiments of the internal organs begin to take form. The final larval stage is the cystacanth, which possesses the proboscis of the adult form.

Acanthocephala stick deep into the intestinal wall, causing severe destruction of the intestinal epithelium, with consequent proliferation of connective tissue. Perforation of the intestinal wall can also occur, when the parasites can be found attached to the liver parenchyma. Species from the genus *Acanthocephalus* are even more dangerous, because they cause similar changes and also change the place of fixation. In species with a small proboscis, only the intestinal epithelium and lamina propria mucosae are damaged, with changes limited to the site of fixation. Signs of the disease include anemia, weight loss and retardation in the growth and development of the young. Changes caused by acanthocephala can be followed by secondary bacterial infections that cause more severe inflammatory processes, intoxications and mass deaths. In case of simultaneous infections, the disease also takes a more severe course, with frequent deaths (Stojanovski, 1997).

Acanthocephalus lucii can be a sensitive bioindicator for lead pollution (Jankovska et al., 2011). They found that lead accumulates in higher concentration in *A. lucii* than in different tissues (liver, gonads and muscle) of perch. The bioconcentration factors for lead indicated that parasites accumulate metals to a higher degree than fish tissues – lead concentrations in acanthocephalans were 9.32, 19.27 and 55.05 higher than in liver, gonads and muscles of host, respectively.

MATERIAL AND METHODS

Fish material from a total of 33 specimens of 3 cyprinid fish from Belčišta wetland (southwest N. Macedonia) were subjected to a parasitological investigation, by seasons this year. Only fresh fish were subjected to routine identification, dissection, and observation methods. Cleaned parasites were separated and put in certain fixatives, prepared for determination with determined techniques of staining and clearing (Vasiljkov, 1983; Gussev, 1983). For the collection of acanthocephalan species, intestines of fish were examined using the stereomicroscope „Zeiss Stemi 305" and microscope „Zeiss Primovert" and parasites were removed. For morphological examination, permanent slide of whole individual parasite was prepared by staining with acetocarmine, dehydrating with ascending grades of alcohol and mounting in Canada balsam. Identification was made throughout the morphology of proboscis with hooks and the reproductive system, using referent key for determination (Bauer, 1987).

RESULTS AND DISCUSSION

From Belčišta wetland three cyprinid fish were investigated parasitologically: Chub - *Squalius squalus* (Bonaparte, 1837) – 15 specimens, minnow – *Phoxinus lumaireul* (Schinz, 1840) - 10 specimens and Ohrid' grunče – *Pelasgus minutus* (Karaman, 1924) - 8 specimens. We found 1 specimen of *Acanthocephalus lucii – female* at 1 minnow this autumn (Table 1).

The parasite species is determined morphologically throughout observation on the shape and measurements of proboscis with hooks and the reproductive system.

Body of *Acanthocephalus lucii* is cylindrical. slightly spread on the front part, length is 15 mm. Proboscis is cylindrical and medially swollen, with a bold, flat epical end. Hooks are well-developed, uniformally slender throughout the length of the proboscis, but smallest posteriorly. The root of the hooks is spread, but without lateral extensions. The hooks are in 16 longitudinal rows, 8 in each row. The length of the proboscis is about 0.13 mm, and the length of front hooks is about 0.05 mm. Eggs are elongate-fusiform, smooth, with no corrugations or special topography. Eggs are 0.10 x 0.02 mm.

Acanthocephalus lucii (Müller, 1776) Lühe, 1911 is found in various European freshwater fishes where it is widely distributed. Bauer (1987) cited that more than 40 species of freshwater fish are host for this parasite. Petrochenko (1956) stated that *A. lucii* is found within the fishes of families: Cyprinidae, Percidae, Esocidae, Anguillidae, Cottidae and others.

According to Bauer (1987) and Benesh and Valtonen (2007), intermediate host for *A. lucii* is *Asselus aquaticus* that is already found in Belčišta wetland (Zoroski, 2022).

According to literatural data of parasitological investigations from the Balkan Peninsula and more widely (Ergens, 1960, 1970; Kazic, 1970; Chankovic et al., 1968; Kyskaroly & Tafro, 1988; Brglez, 1973; Kakacheva-Avramova, 1983; Nedeva-Lebenova, 1991; Cakic, 1992; Hristovski, 1983; Stojanovski, 1997, 2003), etc.), this parasite is found:

1. In Bulgaria at: *Barbus meridionalis petenyi*, *Leuciscus cephalus*, *Silurus glanis* and *Perca fluviatilis*.
2. In Serbia at *Cottus gobio* .
3. In Bosnia and Hercegovina at: *Salmo trutta*, *Salvelimus fontinalis*, *Esox lucius*, *Tinca tinca*, *Cyprinus carpio*, *Silurus glanis*, *Perca fluviatilis*, *Lucioperca lucioperca*, *Acerina cernua*, *Umbra krameri*, *Aspius aspius*, *Pelecus cultratus*, *Noemachilus barbatulus.*
4. In Slovenia at: *Esox lucius*, *Salmo trutta*, *Oncorhynchus mykiss* and *Lota lota*.

Parasite species of the genus *Acanthocephalus* is previously found in N. Macedonia – that is *A. anguillae*. Hristovski (1983) found this parasite at *Salmo letnica, S. ohridanus, S. macedonicus, S. peristericus, Oncorhynchus mykyss, Tinca tinca, Anguilla anguilla, Silurus glanis, Squalius squalus, Gambusia affinis, Cyprinus carpio, Perca fluviatilis* and *Barbus macedonicus* in different lakes and rivers from N. Macedonia. Stojanovski (1997) found this parasite at *Barbus rebeli*, *Scardinius knezevici* and *Anguilla anguilla* from Lake Ohrid.

The record of *Acanthocephalus lucii* in minnow in the present study is considered as the first in N. Macedonia. At the same time, minnow represents a new host for this parasite worldwide.

This parasite species would lead to impairing of fish health status, by causing severe destruction of the intestinal wall, and they can be followed by secondary bacterial infections. This emphasizes the danger of the spread of parasites through stocking with newly introduced fish species.

Data on non-native fish parasites are important for evaluating the health status and their general impact on native fish populations. The introduction of non-native species can have notable effects on the populations of native species, the ecosystem, but can also result in socio-economic consequences, caused by major damages in fisheries and aquaculture in a country.

During the investigations that follow, we expect to find more species of parasites in the fish in the Belčišta wetland, some of which will certainly be new to the parasite fauna of fish from N. Macedonia, and this will contribute to supplementing knowledge and obtaining a more complete picture of the pathology of fish in N. Macedonia, but also the measures that would be taken to prevent the diseases.

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Table 1. Infestation of cyprinid fish from Belčišta wetland with *Acanthocephalus lucii*.

Fish species	Parasite species	Season							
		Winter		Spring		Summer		Autumn	
		Examined	Infested	Examined	Infested	Examined	Infested	Examined	Infested
Squalius squalus	Acanthocephalus lucii	1	0	5	0	3	0	6	0
Phoxinus lumaireul		0	0	0	0	0	0	10	1
Pelasgus minutus		1	0	0	0	0	0	7	0
IN TOTAL		2	0	5	0	3	0	23	1