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ECOPARASITOLOGICAL STUDY OF SIX SPECIES OF FISH FROM THE BULGARIAN SECTION OF THE DANUBE RIVER

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Abstract

For the period 2019-2021, an ecoparasitological study of six fish species - grass carp (Ctenopharyngodon idella Valenciennes, 1844); silver carp (Hypophthalmichthys molitrix Valenciennes, 1844); pumpkinseed (Lepomis gibbosus Linnaeus, 1758); Prussian carp (Carassius gibelio Bloch, 1782); gudgeon (Gobio gobio Linnaeus, 1758); European bitterling (Rhodeus amarus Bloch, 1782) was conducted. The fish were caught from 3 biotopes (Koshava, Kudelin, Novo selo) from the upper section of the Danube River in Bulgaria. A total of two parasite species (Pomphorhynchus laevis Zoega in Müller, 1776) Porta, 1908 and Contracaecum sp. (larvae) were found in two of the investigated fish species. Four fish species were not infected. The ecological indices (mean intensity; mean abundance; prevalence) of parasites were calculated. Kudelin and Koshava biotopes are new habitats for the found helminth species of the infected fish species.

Key words*: freshwater fish, helminths, Koshava, Kudelin, Novo selo.*

INTRODUCTION

Studies on the parasite fauna of *Ct. idella* from the Danube River and the river basin in other countries and Bulgaria are few (Kakacheva-Avramova et al., 1978; Oros & Hanzelová, 2009; Hanzelová et al., 2011). Different authors (Urdeș & Hangan, 2013; Đikanović et al., 2018b; Gologan, 2020; Stroe et al., 2022) have researched parasites of *H. molitrix* from the Danube River and its basin. Studies on helminths of *L. gibbosus* from the Danube River are lacking, but there are data on the species from the river basin (Djikanovic et al., 2011; Djikanović et al., 2018a; Gologan, 2020; and others), and from five locations, part of the Ukraine Black Sea drainages (Kvach et al., 2023). Few authors have studied the parasite fauna of *C. gibelio* (syn. *Carassius auratus* (Bloch); *Carassius auratus gibelio* (Bloch, 1782)) from the Danube River in Bulgaria (Kakacheva-Avramova et al., 1978; Atanasov, 2012), in Romania (Stroe et al., 2022), as well as from the river basin in other countries (Gologan, 2020; Vuić et al., 2022) and in Bulgaria (Shukerova, 2005; 2010). The data on the parasite fauna of *G. gobio* (syn. *Gobio gobio gobio* (Linnaeus, 1758)) from the Danube River (Margaritov, 1966; Kakacheva-Avramova et al., 1978) and the river basin (Kakacheva-Avramova, 1969; Kakacheva-Avramova & Menkova, 1978) in Bulgaria date from the second half of the last century. Helminthological studies on *G. gobio* were also conducted from the river basin on the territory of other countries (Djikanovic et al., 2011; Ondračková et al., 2021; and others). Research on helminths of *Rh. amarus* is scarce (Dávidová et al., 2008).

The purpose of the present study is to provide new data on the helminths of six fish species from the freshwater ecosystem of the Danube River in Bulgaria; as well as new data on the ecological indices of found endohelminth species.

MATERIALS AND METHODS

For the period 2019-2021, a total of 21 specimens from 6 species of fish were subjected to parasitological research. The fish were caught from three sites along the Danube River in the vicinities of the villages Kudelin, Novo selo, and Koshava (noted as biotopes), Vidin Province, Northwestern Bulgaria. Kudelin, Novo selo, and Koshava biotopes are located at 844, 833, and 807 river km, respectively (Figures 1-2).

The fish were caught according to fishing permits for scientific research purposes, issued by the Executive Agency for Fisheries and Aquaculture (EAFA) to the Ministry of Agriculture, Bulgaria. The ecoparasitological examination of the caught fish specimens (Zashev & Margaritov, 1966) was preceded by the determination of the species affiliation (Karapetkova & Zhivkov, 2006; Fröse & Pauly, 2022) and by the recording of basic metric data. Basic ecological indices were calculated, such as mean intensity (MI); mean abundance (MA), and prevalence (P%) (Bush et al., 1997).

Figure 1. Location of Kudelin, Novo selo, and Koshava biotopes from the Danube River, Northwestern Bulgaria (https://www.google.bg/maps/place/Видин)

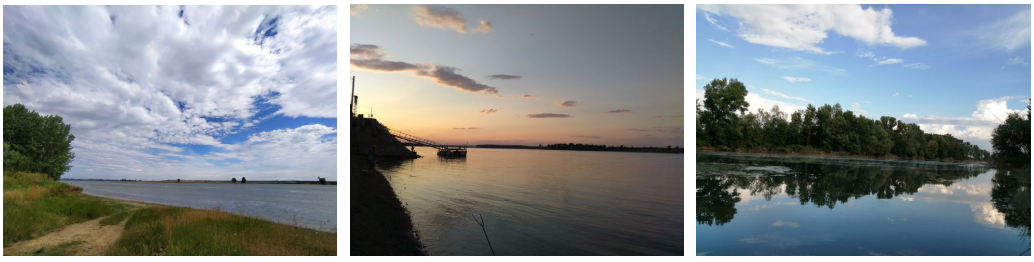

Figure 2. Views from Danube River, Kudelin, Novo selo, and Koshava biotopes; left to right (author's photos)

RESULTS AND DISCUSSIONS

The object of an ecoparasitological study were six fish species: grass carp, *Ctenopharyngodon idella* (Valenciennes, 1844); silver carp, *Hypophthalmichthys molitrix* (Valenciennes, 1844); pumpkinseed, *Lepomis gibbosus* (Linnaeus, 1758); Prussian carp, *Carassius gibelio* (Bloch, 1782); gudgeon, *Gobio gobio* (Linnaeus, 1758); European bitterling, *Rhodeus amarus* (Bloch, 1782) from the freshwater ecosystem of the Danube River, differing in their way of feeding. All studied fish species are benthopelagic. *Ct. idella* has a body length of up to 130 cm and a weight of 35 kg. The species uses for food aquatic vegetation, detritus, insects, and invertebrates. The body length of *H. molitrix* reaches up to 1 m, and the weight is up to 16 kg. Food mainly includes phytoplankton and detritus. *L. gibbosus* is a small fish, with the weight of up to 250 g and a body length of up to 20 cm. It uses for food roe of other fish, zooplankton, and benthic invertebrates. *C. gibelio* has a body length of up to 45 cm and a weight of up to 3 kg. The diet of the species consists of plankton, benthic invertebrates, and plant food. *G. gobio* is a small fish with a body length of up to 20 cm and a weight of up to 100 g. It mainly uses aquatic vegetation and benthic invertebrates for food. *Rh. amarus* is a tiny fish, whose body reaches a length of up to 8 cm and weighs up to

15 g. The diet of the species includes plant food (algae), as well as crustaceans, insect larvae, and others (Karapetkova & Zhivkov, 2006; Fröse & Pauly, 2022). Four of the investigated fish species are included in the IUCN Red List, of which three species (*L. gibbosus*; *G. gobio* and *Rh. amarus*) are in the "LC" category, and one species (*H. molitrix*) is in the "NT" category. Only *Rh. amarus* is included in the Bern Convention, the Habitats Directive, and the Biological Diversity Act (Convention on the conservation of European wildlife and natural habitats, 1982; Council Directive 92/43/EEC of 21 May 1992 on the conservation of natural habitats and of wild fauna and flora, 1992; Biological Diversity Act, 2002; Freyhof & Brooks, 2011).

Ecoparasitological studies

For the period 2019-2021, a total of eight specimens of two herbivorous fish species were examined (1 specimen of *Ct. idella* from Kudelin biotope and 7 specimens of *H. molitrix* from Kudelin biotope); two specimens of one predatory fish species (*L. gibbosus* from Koshava biotope) and eleven specimens of three omnivorous fish species (9 specimens of *C. gibelio* form Kudelin and Novo selo biotopes, 1 specimen of *G. gobio* from Koshava biotope and 1 specimen of *Rh. amarus* from Kudelin biotope). In the study, an infection with a total of 2 species of helminths - *Pomphorhynchus laevis* (Zoega in Müller, 1776) Porta, 1908 and *Contracaecum* sp. (larvae) was found (Table 1). Both helminth species were found in *C. gibelio*. One helminth species was found of *Ct. idella* - *P. laevis*. Four fish species (*H. molitrix*, *L. gibbosus*, *G. gobio* and *Rh. amarus*) were not infected. *P. laevis* is a common parasite for two of the studied fish species. Definitive hosts of *P. laevis* are freshwater fish species of the families Cyprinidae, Salmonidae, Percidae, Siluridae, etc. An intermediate host is *Gamarus pulex* (Linnaeus, 1758) (Petrochenko, 1956; Kakacheva-Avramova, 1983; Bauer (Ed.), 1987). Definitive hosts of *Contracaecum* sp. are waterfowl (*Ardea*, *Egretta*, *Podiceps*, *Phalacrocorax*), and intermediate hosts are copepods of the genera *Cyclops*, *Acanthocyclops*, *Macrocyclops*, *Mesocyclops*, *Eucyclops*, *Arctodiaptomus*, *Diaptomus* (Bauer (Ed.), 1987; Moravec, 2013).

Table 1. Taxonomic position, localization, season, hosts of *Pomphorhynchus laevis* and *Contracaecum* sp.

Helminth species	***Pomphorhynchus laevis* (Zoega in Müller, 1776) Porta, 1908**	***Contracaecum* sp.**
Taxonomic position	**CLASS ACANTHOCEPHALA (RUDOLPHI, 1808)** Family Pomphorhynchidae Yamagiti, 1939 Genus *Pomphorhynchus* Monticelli, 1905	**CLASS NEMATODA RUDOLPHI, 1808** Family Anisakidae Skrjabin et Karokhin, 1945 Genus *Contracaecum* Railliet & Henry, 1912
Localization	intestine	in capsules on the serous membrane of the organs in the abdominal cavity of the fish
Season	Summer *C. gibelio*; autumn *Ct. idella*	summer
Hosts	*Carassius gibelio*, *Ctenopharyngodon idella*	*Carassius gibelio*

Ecoparasitological study of Ctenopharyngodon idella

During the examination of one specimen of *Ct. idella* from Kudelin biotope, one species of class Acanthocephala - *P. laevis*, was found. *P. laevis* had equal mean intensity and mean abundance (MI = MA = 2.00) (Table 2).

Table 2. Species diversity and ecological indices in the helminth community of *Ctenopharyngodon idella* from the Danube River

Ctenopharyngodon idella **(N = 1/Kudelin)** **Parasite species**	**n**	**p**	**MI**	**MA**	**P%**	**R**
Pomphorhynchus laevis (Zoega in Müller, 1776) Porta, 1908	1	2	2.00	2.00	100.00	2

N - number of investigated fish; n - number of infected fish; p - number of fish parasites; MI - mean intensity; MA - mean abundance; P% - prevalence; R - range

Ecoparasitological study of Carassius gibelio

During the examination of 7 and 2 specimens of *C. gibelio* from Kudelin and Novo selo biotopes, respectively, infection was found in only one specimen from each biotope. One helminth species from class Acanthocephala - *P. laevis*, was found in the Prussian carp from Kudelin biotope, and one helminth species from class Nematoda - *Contracaecum* sp. was found in the Prussian carp from Novo selo biotope. Of the two detected helminth species, *Contracaecum* sp. had higher ecological indices (MI = 49.00, MA = 24.50 and P% = 50.00) (Table 3).

Table 3. Species diversity and ecological indices in the helminth community of *Carassius gibelio* from the Danube River

Carassius gibelio **(N = 7/Kudelin)** **Parasite species**	**n**	**p**	**MI**	**MA**	**P%**	**R**
Pomphorhynchus laevis (Zoega in Müller, 1776) Porta, 1908	1	3	3.00	0.50	16.67	3
Carassius gibelio **(N = 2/Novo selo)** **Parasite species**	**n**	**p**	**MI**	**MA**	**P%**	**R**
Contracaecum sp. (larvae)	1	49	49.00	24.50	50.00	49

N - number of investigated fish; n - number of infected fish; p - number of fish parasites; MI - mean intensity; MA - mean abundance; P% - prevalence; R - range

Oros & Hanzelová (2009); Hanzelová et al. (2011) examined for parasites *Ct. idella* from the Latorica River, part of the Danube River basin in Slovakia, and found the trematode *Sphaerostoma bramae* (Müller, 1776) Lühe, 1909 (syn. *Sphaerostomum bramae* Müller, 1776). *P. laevis* has been reported in grass carp from the Bulgarian section of the Danube River (Kakacheva-Avramova et al., 1978). Both parasites found in the present study in *C. gibelio* were reported either for the Danube River or the river basin in Bulgaria. *P. laevis* has been reported in *C. gibelio* from the Bulgarian section of the Danube River (Kakacheva-Avramova et al., 1978; Atanasov, 2012). While *Contracaecum* sp. is reported as *Contracaecum microcephalum* (Rudolphi, 1809) of Prussian carp from Srebarna Lake (Shukerova, 2005; 2010). Stroe et al. (2022) studied the parasite fauna of *C. gibelio* and *H. molitrix* from the Romanian section of the Danube River (Brăila station). The authors reported Trichodina sp., *Dactylogyrus vastator* (Nybelin, 1924) of Prussian carp, and *Diplostomum spathaceum* (Rudolphi, 1819) of silver carp. Vuić et al. (2022) reported *Contracaecum* larvae of Prussian carp from Lake Sakadaš, part of the Danube River basin in Croatia (Tables 4-5).

Table 4. Distribution of the found helminths (in the present study) of *Ctenopharyngodon idella* from the Danube River and its basin

Helminth species / Biotopes	Kudelin biotope	Danube River in other countries	Danube River Basin in other countries	Danube River in Bulgaria	Danube River Basin in Bulgaria
Pomphorhynchus laevis (Zoega in Müller, 1776) Porta, 1908	+	-	-	+	-

Table 5. Distribution of the found helminths (in the present study) of *Carassius gibelio* from the Danube River and its basin

Helminth species / Biotopes	Kudelin biotope	Novo selo biotope	Danube River in other countries	Danube River Basin in other countries	Danube River in Bulgaria	Danube River Basin in Bulgaria
Pomphorhynchus laevis (Zoega in Müller, 1776) Porta, 1908	+	-	-	-	+	-
Contracaecum sp.	-	+	-	+	-	+

According to the obtained results, the way of feeding is not determining by the infection. The mean intensity of the intermediate hosts in the studied biotopes has a predominant influence.

CONCLUSIONS

In the study of six fish species, it was found that only two species (*C. gibelio* and *Ct. idella*) were infected. The intermediate hosts of the found helminth species, rather than the diet of the hosts, are of predominant importance for the infection. When comparing the ecological indices of *P. laevis* from the two studied fish species, the highest prevalence was found for *P. laevis* of grass carp (Kudelin biotope) - P% = 100.00. In Prussian carp (Kudelin and Novo selo biotopes), higher mean intensity and mean abundance were established for *Contracaecum* sp. (MI = 49.00 and MA = 24.50). Kudelin and Novo selo biotopes are new habitats for the found helminth species of grass carp and Prussian carp.

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