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HELMINTHS AND HELMINTH COMMUNITIES OF ROUND-SCALED BARBELL (*BARBUS CYCLOLEPIS* HECKEL, 1837) AND ITS BIOINDICATOR ROLE

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Abstract

During 2018, 22 specimens of the round-scale barbell from the Tamrashka River (Bulgaria) were examined for helminths. B. cyclolepis is an endemic fish species of the Maritsa River Water Collection. Five parasite species (Allocreadium isoporum Ergens & Lom, 1970; Caryophyllaeides fennica (Schneider, 1902) Nybelin, 1922; Pomphorhynchus laevis (Müller, 1776); Rhabdochona hellichi (Šramek, 1901) Chitwood, 1933; Rhabdochona gnedini Skrjabin, 1948) belonging to four classes and four families were fixed. The Tamrashka River is a new habitat for All. isoporum, C. fennica, P. laevis, Rh. hellichi and Rh. gnedini of B. cyclolepis in Bulgaria. The dominant structure of the parasite communities was discussed based on the level of the component community. The bioindicator role of the studied parasite populations and communities are presented.

Key words*: Aegean Water Basin, B. cyclolepis, bioindication, helminths, helminth communities*.

INTRODUCTION

Barbus cyclolepis Heckle, 1837, is an endemic fish species of the Maritsa River Basin (Kolev, 2016). The Tamrashka River is one of the most significant right tributaries of the Maritsa River, Aegean Water Basin. The river springs west of Modar Peak (about 1800 m above sea level) in the Chernatitsa Ridge, Western Rhodopa Mountain, and Southern Bulgaria. After the village of Parvenets, the Tamrashka River enters the Upper Thracian Plain under the name Parvenetska River. It flows into the Maritza River at 164 meters above sea level western of the town of Plovdiv. The freshwater fish, including *Barbus cyclolepis*, and its helminths are used as biological elements for bioindication (MacKenzie et al., 1995; Lambert and El Gharbi, 1995; Kelepertzis et al., 2012; Kirin et al., 2013). At the same time, the studies on the helminths and helminth communities of *B. cyclolepis* are extremely limited. In Bulgaria to this time, have not studied on the bioindicative importance of Round–scaled barbell, its helminths and helminth communities. The study aims to present the results from the examination of helminth and helminth communities of endemic fish species *Barbus cyclolepis* Heckle, 1837 from the Tamrashka River, Bulgaria.

MATERIALS AND METHODS

During 2018, 22 specimens of *Barbus cyclolepis* Heckel, 1837 from the Tamrashka River, Bulgaria were examined for helminths. The fish were caught by angling according to permission from Ministry of Agriculture, Food and Forestry of the Republic Bulgaria. The scientific name of the fish was present, according to Bianco (1998); Froese and Pauly (Eds.) (2019). The fish were caught in a section of river between the villages of Hrabrino and Parvenets ($40^{0}03'01.60$N and $24^{0}3838.57$E), in Rodopa Mountain, about 17.26 km far away from the town of Plovdiv, Southern Bulgaria. The helminthological studies were carried out according to the Zashev and Margaritov (1966); Bauer (Ed.) (1987); Moravec (2013). The detected and isolated helminth specimens were fixed in 70% of ethyl alcohol. Species diversity of representatives from classes Trematoda and Cestoda were determined on permanent slides carried out by the methods of Georgiv et al. (1986) and Scholz and Hanzelová (1998) and

from classes Acanthocephala and Nematoda – on temporary slides carried out by the method of Moravec (2013). Helminth community structure was analysed by two levels: on the level of component community (prevalence (P%); mean intensity (MI) for the determined species) and on the level of infracommunity (total number of fish species; total and mean number of fish specimens; Brillouin's diversity index (HB) and Pielou's evenness index (E)).

In the component community, the found species were divided as core species (P% > 20), component species (P% > 10) and accidental species (P% < 10), according to the criteria of Magurran (1988); Bush et al. (1997) and Kennedy (1997). The obtained results were statistically processed using the software products Statistica 10 (StatSoft Inc., 2011) and MS Exel (Microsoft 2010).

RESULTS AND DISCUSSIONS

Fish communities

The Round–scaled barbell or Maritsa barber (*Barbus cyclolepis* Heckel, 1837; Cyprinidae) inhabits the bottom of reservoirs, the middle and upper part of the rivers with sandy–gravelly or rocky bottom. In the spring it goes down to the deeper parts of the rivers, and in the summer it goes up to the fast streams.

The Maritsa barbell measures up to 30 cm in length and weighs up to 1 kg. The colour of the fish depends mainly on the environment. Most often the colour of fish is silvery, streaked with fine dark spots.

The fins are yellowish, only in certain cases, turning pure orange. The Round-scaled barbell feeds mainly on demersal organisms.

The species prefers for food larvae and larvae of insects, all crustaceans, invertebrates, and also worms, caterpillars and insects. Sexually mature at 2-3 years. The fish species propagated mainly in the period April – May.

The species lives up to 10 years. It is most widespread fish species in the rivers Maritsa, Mesta, Struma and their tributaries, also in rivers in north-eastern Greece (Bianco, 1998; Karapetkova and Zhivkov, 2006; Kottelat and Freyhof, 2007; Kolev, 2013; 2016). *B. cyclolepis* is an IUCN as non–threatened species (= LC = Least Concern) (Froese & Pauly (Eds.), 2019). The species is included in Annexes 2 and 4 of the Biodiversity Law of the Republic of Bulgaria. *B. cyclolepis* is an endemic species for the Maritsa catchment area (Kolev, 2016).

Helminth community structure

The helminths and helminth communities of 22 specimens *B. cyclolepis* from the Tamrashka River (Aegean Water Basin, Bulgaria) were examined. Five species of helminths (*Allocreadium isoporum (*Ergens and Lom, 1970); *Caryophyllaeides fennica* (Schneider, 1902; Nybelin, 1922); *Pomphorhynchus laevis* (Müller, 1776); *Rhabdochona hellichi* (Šrameć, 1901; Chitwood, 1933; *Rhabdochona gnedini (*Skrjabin, 1948), belonging to 4 classes, four orders and four families have been found. Only from the class Nematoda, two species of helminths were fixed. The other three classes are represented by one species of helminths (Table 1).

Table 1. Biodiversity and ecological indices of helminths and helminth communities of *Barbuscyclolepis* from the Tamrashka River

Barbus cyclolepis (N[1] = 22) Helminth species	n[2]	p[3]	P%[4]	MI[5]
Trematoda Class Order Fasciolida Family Allocreadidae				
Allocreadium isoporum (Looss, 1894)	15	54 1-12	68.18	3.6
Cestoda Class Order Caryophyllaeidea Family Caryophyllaeidae				
Caryophyllaeides fennica (Schneider, 1902) Nybelin, 1922	2	2	9.09	1
Acanthocephala Class Order Echinorhynchidae Family Pomphorhynchidae				
Pomphorhynchus laevis (Müller, 1776)	9	27 1-4	40.9	3.0
Nematoda Class Order Spirurida Family Rhabdochonidae				
Rhabdochona hellichi (Šramek, 1901) Chitwood, 1933	2	3 1-2	9.09	1.5
Rhabdochona gnedini Skrjabin, 1948	1	4	4.54	4

[1]N = total number of examined fish specimens.
[2]n = total number of infected fish specimens.
[3]p = total number of helminth specimens.
[4]P% = prevalence.
[5]MI = mean intensity.

The adult specimens of *Allocreadium isoporum* (Looss, 1894) are developing in different species of Cyprinidae. The parasite's life cycle

involves the participation of two intermediate hosts. The first intermediate hosts are snails of *Sphaerium* genus, and the second are larvae's of insects of Ephemera genera, Linnaeus, 1758 (Ephemeridae), *Anabolia* Cuvier, 1827 (Limpetridae) and *Chaetopterix* (Limnephilidae) (Bauer, 1987; Kakacheva–Avramova, 1983). The species was reported from *B. cyclolepis* in Bulgaria (from Syuyutlijka rivers, Asenitsa – Kakacheva–Avramova, 1965; from Vacha River – Margaritov, 1965; from Tundzha River – Kakacheva–Avramova, 1972; from Luda Yana River – Kirin, 2002a). According to this research, the Tamrashka River is a new habitat for the *All. isoporum* trematode species as parasite species of *B. cyclolepis*.

Caryophyllaeides fennica (Schneider, 1902) Nybelin, 1922 is an intestinal parasite of *Squalius cephalus* (Linnaeus, 1758), *Leuciscus idus* (Linnaeus, 1758), *Barbus barbus* (Linnaeus, 1758), *B. meridionalis petenyi* Heckel, 1852, *Rutilus rutilus* (Linnaeus, 1758), *Abramis brama* (Linnaeus, 1758), *Scardinius erytrophthalmus* (Linnaeus, 1758), *Aspius aspius* (Linnaeus, 1758), *Gobio gobio* (Linnaeus, 1758) and other fish species of Cyprinidae. *Stylaria lacustris* (Linnaeus, 1767) (Oligochaeta) is an intermediate host of the parasite (Bauer, 1987; Kakacheva–Avramova, 1983). *St. lacustris* is a bioindicator for β–mesosaposity. The species refers to the relatively tolerant forms (group C) in terms of the conditions in the habitats (Rusev, 1993; Peev and Gerasimov, 1999; Belkinova et al., 2013). *C. fennica* was reported of *B. cyclolepis* in Bulgaria (from Topolnica River – Margaritov, 1965; from Asenica, Harmanlijska, Topolnitsa, Syuyutlijka and Sushitsa rivers, Bedechka – Kakacheva–Avramova, 1965; from Tundzha River – Kakacheva–Avramova, 1972; from Arda River – Kirin, 2003). The Tamrashka River is a new habitat for *C. fennica* from *B. cyclolepis*.

Pomphorhynchus laevis (Müller, 1776) develops as a marita in a lot of freshwater fish species of Cyprinidae, Salmonidae, Percidae, Siluridae, etc. The developmental cycle is related to the participation of an intermediate host – the *Gammarus pulex* crustaceans (Linnaeus, 1758) (Bauer, 1987; Kakacheva–Avramova, 1983). *G. pulex* is a bioindicator for x–β–mesosaprobity as well as relatively tolerant forms (Group C) in terms of environmental conditions in habitats (Rusev, 1993; Peev & Gerasimov, 1999; Belkinova et al., 2013). Small fish species of Cyprinidae have been established as a reservoir host. The species has been reported by *B. cyclolepis* in Bulgaria (from Tundzha River – Kakacheva–Avramova, 1972). The Tamrashka River is a new habitat for *P. laevis* from *B. cyclolepis*.

Rhabdochona hellichi (Šramek, 1901) Chitwood, 1933 is an intestinal parasite species of different species of freshwater fish of the families of Cyprinidae (*B. barbus*, *B. meridionalis*, *B. petenyi*, etc.), Salmonidae (*Salmo trutta fario* Linnaeus, 1758, *Oncorhynchus mykiss* (Walbaum, 1792), *Thymallus thymallus* (Linnaeus, 1758)), Acipenseridae, Siluridae, etc., which are the final hosts of the species (Bauer, 1987; Kakacheva–Avramova, 1983; Moravec, 2013). In the scientific literature, no data on the species development cycle are reported (Moravec, 2013). *Rh. hellichi* has been reported as a parasite of *B. cyclolepis* in Bulgaria (from Tundzha River – Kakacheva–Avramova, 1972; from Arda River – Kirin, 2003). According to the study, the Tamrashka River is a new habitat for *Rh. hellichi* from *B. cyclolepis*.

Rhabdochona gnedini Skrjabin, 1948 is an intestinal parasite of many fish species of Cyprinidae (*B. barbus*, *B. cyclolepis*, *B. meridionalis*, *Luciobarbus bocagei* (Steindachner, 1864), *L. caspius* (Berg, 1914), *R. rutilus*, *Sq.cephalus*, *Sq. svallize (*Heckel and Kner, 1858), Salmonidae (*S. t. fario*, *S. trutta trutta* Linnaeus, 1758, *S. marmoratus* Cuvier, 1829, *S. obtusirostris* (Heckel, 1851), *O. mykiss*, *Hucho hucho* (Linnaeus, 1758), *Th. thymallus*), Percidae (*Zingel streber* (Siebold, 1863), *Z. zingel* (Linnaeus, 1766)), Siluridae (*Silurus glanis* Linnaeus, 1758) and others (Moravec, 2013). The species development cycle has not been studied (Moravec, 2013). The species was reported as a parasite of *B. cyclolepis* in Bulgaria (from rivers Vacha and Maritsa as *Rh. sulaki* – Margaritov, 1965).The Tamrashka River is a new habitat for *Rh. gnedini* from *B. cyclolepis*.

Component communities

With the highest prevalence were distinguished two helminth species of *B. cyclolepis*: *All. isoporum* (P% = 68.18) and *P. laevis* (P% =

40.90). They are core species for the helminth communities of *B. cyclolepis*. The other three species of helminth (*C. fennica*, *Rh. hellichi* and *Rh. gnedini*) are accidental species on these communities. The highest mean intensity was fixed for *Ph. gnedini* (MI = 4.0), followed by those of *All. isoporum* (MI = 3.6), *P. laevis* (MI = 3.0), etc. As a result of this study, only one specimen of *C. fennica* was found. The population of *Rh hellichi* also showed low mean intensity (MI = 1.5) (Table 1).

Infracommunities

All examined fish specimens of *B. cyclolepis* from Tamrashka River were infected. In 15 specimens of fish, one species of helminth was found, and in 7 specimens of Round-scaled barbell, two species of helminths were found. A total of 90 helminth specimens were studied. The helminth infection was presented from one to 12 specimens, the mean of 3.03 specimens per one specimen of infected fish (Table 2).

Table 2. Infracommunities data

Number of helminth species		
Total number of species	5	
Number of fish	15	7
Number of helminth species	1	2
Number of helminth specimens		
Total number of specimens	90	
Mean ± SD	3.03 ± 2.24	
Range (min - max)	1 – 12	
HB ± SD (Brillouin'sdiversity index)	0.929 ± 1.23	
E ± SD (Pielou's evenness index)	0.624 ± 0.25	

No parasites causing dangerous diseases to fish, humans or other hosts have been identified, as reported by some authors from different hosts (Kakacheva-Avramova and Nedeva, 1979; Margaritov, 1959; Pekova et al., 2017a; Pekova et al., 2017b; Mitev et al., 2020). Other authors also reported the helminth species, found in this study in the country except for *Ph. gnedini*. Some of them from Southern Bulgaria are presented in Table 3.

Table 3. Some other species of fish from freshwater ecosystems in Southern Bulgaria, hosts of helminths reported of *Barbus cyclolepis* from the Tamrashka River

Helminth species	Other fish species	Authors
Allocreadium isoporum (Looss, 1894)	*Gobio gobio* (Linnaeus, 1758)	Kakacheva, 65
	Squalius orpheus Kottelat & Economidi, 2006 (*Leuciscus cephalus*)	Kakacheva-Avramova, 65; Margaritov, 65; Kirin, 2000; 2001a,b; 2002c
Caryophyllaeides fennica (Schneider, 1902) Nybelin, 1922	*Alburnus alburnus* Linnaeus, 1758	Kirin, 2003
	Barbus barbus	Margaritov, 1959
	Squalius orpheus Kottelat & Economid, 2006 (*L. cephalus*)	Kakacheva-Avramova, 1965; Kirin, 2002c; Kirin et al., 2013
	Vimba melaniops	Kakacheva-Avramova, 1965
Pomphorhynchus laevis (Müller, 1776)	*Sander lucioperca*	Nedeva & Grupcheva, 1996
	Squalius orpheus Kottelat & Economid, 2006 (*L. cephalus*)	Kirin, 2000; 2001b
	Salmo trutta fario Linnaeus, 1758	Kakacheva-Avramova & Nedeva, 1979
Rhabdochona hellichi (Šramek, 1901) Chitwood, 1933	*Barbus meridionalis petenyi* Heckel, 1852	Kakacheva_Avramova, &Nedeva, 1978
	Salmo trutta fario Linnaeus, 1758	Kakacheva-Avramova & Nedeva, 1978; 1979; 1982; Kirin, 2002a
	Oncorhynchus mykiss (Walbaum, 1792)	Kakacheva-Avramova & Nedeva, 1979; 1982
Rhabdochona gnedini Skrjabin, 1948 (*Rh. sulaki*)	-	-

There are a few types of research of the parasites and parasite communities of *B. cyclolepis* from freshwater ecosystems in other countries. They mainly refer to representatives of Monogenea class (Šimková et al., 2007 – including the materials from Bulgaria; Lambert and El Gharbi, 1995).

The studied Tamrashka River is not exposed to anthropogenic pressure or other negative effects. The adjacent territories are characterized by high biodiversity, a number of protected, endemic and relict species. Determined helminth species of *B. cyclolepis*, total number of species and total number of specimens, parasites life cycles, calculated diversity indices (HB = 0.929, Brillouin's diversity index) and evenness indices (E = 0.624, Pielou's evenness index) is a reliable evidence of β–mesosaprobity conditions in the freshwater ecosystems of the

Tamrashka River and of the very good ecological status of the investigated freshwater biotopes (Tables 1 – 2; Rusev, 1993; Peev and Gerasimov, 1999; Belkinova et al., 2013).

CONCLUSIONS

The Tamrashka River is a new habitat for the helminth species of *Barbus cyclolepis*: *Allocreadium isoporum*, *Caryophyllaeides fennica*, *Pomphorhynchus laevis*, *Rhabdochona hellichi*, *Rhabdochana gnedini*. *All. isoporum* and *P. laevis* are component species of the helminth communities of *B. cyclolepis*. With the highest mean intensity are distinguished the species *Rh. gnedini* and *All. isoporum*. The analyzed results of the studied helminth and helminth communities ofthe Round–scaled barbell showed the very good ecological condition of the investigated freshwater ecosystem.

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