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ENDOHELMITHS AND ENDOHELMINTH COMMUNITIES OF *Rutilus rutilus* (Linnaeus, 1753) FROM ANTHROPOGENIC LOADED ECOSYSTEM OF THE LUDA YANA RIVER, BULGARIA

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Abstract

During 2018, biomonitoring of the Luda Yana River ecosystem was carried out by examining the biological elements of quality: the freshwater fish species common roach (Rutilus rutilus (Linnaeus, 1753)) and its endohelmiths and endohelminth communities as bioindicators. 45 specimens of common roach are examined for parasites and three species of endohelmiths (Caryophyllaeides fennica (Schneider, 1902) Nybelin, 1922; Acanthocephalus lucii (Mueller, 1776); Rhabdochona denudata (Dujardin, 1845) Raillet, 1916) are fixed. New host and locality records are reported. The analysis of the dominant structure of the established intestinal parasite complex was presented to the level of the component community. For an ecological estimation of the freshwater ecosystem, principal biotic indexes are fixed. The bioindicator significance of the identified parasite populations and communities are discussed.

Key words: *bioindication, Luda Yana River, parasite communities, Rutilus rutilus.*

INTRODUCTION

The Luda Yana River is a part of the protected area of the National Network Natura 2000 (BG 0000426 River Luda Yana) according to Directive 92/43/EEC. The protected area is distinguished by great biodiversity. At the same time, the river is influenced by the serious anthropogenic impacts as a result of irrigation, ore mining activities, industrial and municipal waste, etc. (Georgieva et al., 2014). According to other studies, the mining activities, the weathering and the oxidation processes have strong effects on the physicochemical processes in the whole water ecosystem (Rabadjieva et al., 2009). Freshwater fish parasites and its parasite communities are increasing interestbeing used as accumulative bioindicators and for ecological assessment of the ecosystems (Nachev, 2010; Sures et al., 2017; etc.). Parasite communities of *R. rutilus* are indicative of pollutants, eutrophication and fragmentation in the environment (Valtonen et al., 1997; Valtonen et al., 2003; etc.). This paper presents the results from an examination of common roach parasites, dominant structure of fish parasite communities and their bioindicator role for biodiversity condition of the freshwater ecosystem of the Luda Yana River (town of Popinci; Aegean Basin).

MATERIALS AND METHODS

During April-August, 2018 fish and fish parasites are collected and examined from the Luda Yana River (after village of Popinci). The Luda Yana River is of 73.05 km long and is one of the biggest left tributaries of the Maritsa River (Southern Bulgaria). The River springs from 1423 m, from the west of the Bich peak at Sredna Gora Mountain and flows into the Maritsa River at 195 m altitude. It has a catchment's area of 685 km^2, which occupies 1.3% of the Maritsa River catchment (Michev et al., 1980).

The Luda Yana River refers to Type R12: Large Plain Rivers in Ecoregion 7 (Eastern Balkans) (Belkinova et al., 2013). It features a sandy-gravel bottom and a rainy-snowy feeding. It is mainly used for irrigation and industrial water supply, influencing its ecological status (Georgieva et al., 2014).

The studied biotope (42^{0}41'66'67N, 24^{0}28'33'33E, 343 m altitude) is divided into two parts from the river (Michev et al., 1980).A total of 45 freshwater fish specimens belonging

to the species *Rutilus rutilus* (Linnaeus, 1758) are collected and examined for endohelmiths. The fish are caught by angling. The scientific and common names of the fish hostare used according to the FishBase database (Fröse and Pauly, 2018). Helminthological examinations are carried out following recommendations and proceduresdescribed by Petrochenko (1956); Kakacheva-Avramova (1983); Bykhovskaya-Pavlovskaya (1985); Bauer (Ed.) (1987); Protasova et al. (1990); Moravec (2013); etc. Specimens are fixed and preserved in 70% ethyl alcohol. The Cestoda are studied with methods of Georgiev et al. (1986); Scholz and Hanzelová (1998). The acanthocephalans and nematodes are studied on temporary mounts with 5% glycerolin 70% ethanol (Zashev and Margaritov, 1966; Moravec, 2013). The ecological terms prevalence (P%), mean intensity (MI) are presented for each species. Analyses of helminth community structure are carried out in both levels: infracommunity and component community. The infracommunity data are used to calculate the total number of species, the mean number of helminths, etc. (Kennedy, 1993; 1997; Magurran, 1988). The infracommunity data are used to calculate the total number of species, mean number of helminth worms, the Brillouin's diversity index (HB) (Maguran, 1988). The analysis of the dominant structure of the parasite communities is presented to the level of the component communities using the criterion of Bush at al. (1997). The diversity measures are calculated by MS Excel (Microsoft 2010) and Statistica 10 (StatSoft Inc., 2011).

RESULTS AND DISCUSSIONS

Fish communities

A total of 45 fish specimens belonging to the species *Rutilus rutilus* (Linnaeus, 1758) are collected and examined from the Luda Yana River. *R. Rutilus*is estimated as least concern species (LC=LeastConcern; IUCN Red List Status, 2019) and isnot included in Red Data Book of the Republic of Bulgaria (Golemanski (Ed.), 2011). The common roach occurs in fresh and brackish waters but is very adaptable and can be found in different freshwater ecosystems (small ponds, lakes, big rivers). *R. rutilus* feeds mainly on crustaceans, insect larvae, oligochaetes, algae and higher aquatic vegetation (Karapetkova and Zhivkov, 2006; Fröse and Pauly, 2018). *R. rutilus* is a part of biological elements for bioindication of the fresh water ecosystems (Belkinova et al., 2013). *R. rutilus* is one of the dominant fish species of the freshwater ecosystem of the Luda Yana River. Two of the examined fish specimens are free of parasites.

Helminth community structure

From studied 45 specimens of common roach (*Rutilus rutilus* (Linnaeus, 1758)), 3 parasite species are fixed (*Caryophyllaeides fennica* (Schneider, 1902) Nybelin, 1922; *Acanthocephalus lucii* (Mueller, 1776); *Rhabdochona denudata* (Dujardin, 1845) Raillet, 1916). They are belonging to classes Cestoda (1), Acanthocephala (1) and Nematoda (1).

Definitive hosts of *C. fennica* are a number of fish species from Cyprinidae: *Squalius cephalus* (Linnaeus, 1758)*, Leuciscus idus* (Linnaeus, 1758)*, Barbus barbus* (Linnaeus, 1758)*, B. petenyi* Heckel, 1852*, R. rutilus, Abramis brama* (Linnaeus, 1758)*, Scardinius erythrophthalmus* (Linnaeus, 1758)*, Aspius aspius* (Linnaeus, 1758)*, Gobio gobio* (Linnaeus, 1758), etc. (Kakacheva-Avramova, 1983; Moravec, 2001; Bauer, 1987; Protasova et al., 1990;etc.). Development of *C. fennica* is with the participation of an intermediate host – oligochaets of the species *Stylaria lacustris* (Linnaeus, 1767) (Bauer, 1987; Kakacheva-Avramova, 1983; Protasova et al., 1990). *R. rutilus* is a typical host for *C. fennica* (Protasova et al., 1990).

In Bulgaria, the species *C. fennica* was presented of *B. barbus* from Iskarand Tundzharivers; of *B.petrnyi* from rivers Iskarand Palakariya; of *Sq.cephalus* (*Leuciscus cephalus* (Linnaeus, 1758)) from Iskar River (Margaritov, 1959); *Sq. cephalus*, *Barbus cyclolepis* Heckel, 1837 and *Vimba melanops* (Heckel, 1837) from Maritsa and Topolnitsa rivers (Margaritov, 1964); of *B. cyclolepis*, *Sq. cephalus*, *V. melanops* from Asenitsa, Topolnitsa, Syuyutlijka, Sushitsa and Bedechka rivers (Kakacheva-Avramova, 1965); of *B. barbus* and *Sander lucioperca* (Linnaeus, 1758) (*Lucioperca lucioperca* (Linnaeus, 1758)) from the Danube River (Margaritov, 1966); of *B.*

petenyi from Nishava, Ogosta, Vodomerka, Buchinska, Vrabnishka, Barziya, Chuprenska, Iskrecka, Botunya, Bebresh rivers; of *B. barbus* from the Bogovina River; of *Sq. cephalus* from Bogovina, Nishava, Ogosta, Vodomerka, Barziya, Botunya, Bebresh rivers; of *G. gobio* from Botunya and Bebresh rivers; of *Chondrostoma nasus* (Linnaeus, 1758) from the Ogosta River; of *Alburnus alburnus* (Linnaeus, 1758) from the Leva River (Kakacheva-Avramova, 1969); of *Sq. Cephalus* and *R. rutilus* from the Shiposhnitsa River and Reservoir Iskar (Margaritov, 1977); of *Vimba vimba* (Linnaeus, 1758), *A.brama*, *Ballerus sapa* (Pallas, 1814) (*Abramis sapa* (Pallas, 1814)), *Ballerus ballerus* (Linnaeus, 1758) (*Abramis ballerus* (Linnaeus, 1758)), *Blicca bjoerkna* (Linnaeus, 1758), *A. alburnus*, *B. barbus*, *S. lucioperca*, *S.erythrophthalmus*, *Pelecus cultratus* (Linnaeus, 1758) (Kakacheva-Avramova and Menkova, 1978); *Sq. cephalus* and *R. rutilus* from the Palakariya River (Kakacheva-Avramova and Menkova, 1978); of *B. barbus* from the Struma River (Kakacheva-Avramova and Menkova, 1981); of *B. petrnyi* from the Mesta River (Kirin, 2001a); of *Sq. orpheus* from the Arda River (Kirin, 2002a; 2002b; Kirin et al., 2003); of *Sq. orpheus* and *A. alburnus* from the Arda River (Kirin et al., 2002); of *A. alburnus* and *B. cyclolepis* from the Arda River (Kirin, 2003); of *Sq. cephalus* from the Stryama River (Kirin et al., 2005); of *Sq. cephalus* from the Danube River (Cakic et al., 2004); of *B. barbus* from the Danube River (Atanasov, 2012); of *Sq. orpheus* from the Tunja River (Kirin et al., 2013), etc.

Definitive hosts of *A. lucii* are freshwater fish species from Cyprinidae, Percidae, Siluridae, Salmonidae, Esocidae, Gadidae, Cobitidae, Anguillidae. Intermediate host are crustaceans *Asellus aquaticus* (Linnaeus, 1758). (Petrochenko, 1956; Kakcheva-Avramova, 1983; Bauer, 1987).

In Bulgaria, the species *A. lucii* was presented of *Silurus glanis* Linnaeus, 1758 from the Danube River and of *Sq. cephalus* from Iskar and Tundzha rivers (Margaritov, 1959); of *Perca fluviatilis* Linnaeus, 1758 (Margaritov, 1966); of *B. sapa*, *Sq. cephalus*, *R. rutilus*, *S. glanis*, *P. fluviatilis*, *Lota lota* (Linnaeus, 1758), *Acerina schraetser* (Linnaeus, 1758), *Benthophilus stellatus* (Sauvage, 1874), *Proterorhinus marmoratus* (Pallas, 1814) (Kakacheva-Avramova et al., 1978); of *Sq. cephalus* (Cakic et al., 2004); of *L. lota* and *Zingel zingel* (Linnaeus, 1766) (Atanasov, 2012); of *A. brama* (Chunchukova et al., 2017); of *A. alburnus* (Chunchukova et al., 2018), from the Danube River; of *P. fluviatilis* (Shukerova et al., 2010) and *A. brama* (Chunchukova et al., 2016), from the Lake Srebarna etc.

Definitive hosts of *R. denudate* are fish species from Cyprinidae. Intermediate hosts are larvae of representatives of the generas *Heptagenia*, *Ephemerella* and *Hydropsyche* (Kakcheva-Avramova, 1983; Bauer, 1987).

In Bulgaria, the species *R. denudata* was presented of *B. barbus*, *B. petenyi* and *Sq. cephalus* from the Iskar River (Margaritov, 1959); of *Sc. erythrophthalmus* from the Strumeshnitsa River (Kakacheva-Avramova, 1962); of *Sq. cephalus*, *A. alburnus*, *Leuciscus aspius* (Linnaeus, 1758) (*Aspius aspius* (Linnaeus, 1758)), *B. cyclolepis* from Trakian's freshwater ecosystems (Kakacheva-Avramova, 1965); of *Sq. cephalus* from Maritsa, Vacha, Chepinska rivers; of *V. melanops* from the Maritsa River; of *A. alburnus* from Maritsa and Chepinska rivers; of *B. cyclolepis* from Maritsa, Chepinska, Vacha and Topolnitsa rivers (Margaritov, 1964); *Sq. cephalus* from Ogosta, Vrabnishka, Barziya, Nishava, Botunya, Leva, Archar, Berkovska, Chuprenska rivers; of *B. peteny* from Chuprenska, Barziya and Leva rivers; of *B. barbus* from the Leva River; of *G. gobio* from the Barziya River; of *A. alburnus* from Ogosta, Lom and Leva rivers (Kakacheva-Avramova, 1969); of *Sq. cephalus* from the Shiposhnitsa River and Reservoir Iskar (Margaritov, 1977); of *A. alburnus*, *Zingel streber* (Siebold, 1863), *Z. zingel* (Kakacheva-Avramova et al., 1978); of *Sq. cephalus* from the Palakariya River (Kakacheva-Avramova and Menkova, 1978); of *Cobitis taenia* Linnaeus, 1758 from State Fish Farming Blagoevgrad; of *Sq. cephalus* from Zheleznitsa, Blagoevgradska, Bistritsa, Gradevska and Strumarivers (Kakacheva-Avramova and Menkova, 1981); of *Sq. cephalus* and *B. cyclolepis* from the Struma River (Nedeva, 1991); of *C. carpio* (Kirin, 2001a) from the Mesta River; of *Sq. cephalus*

and *A. alburnus* from Kardzhali Reservoir (Kirin, 2001b); of *Sq. orpheus* (Kirin, 2002a; 2002b); of *B. cyclolepis* and *A. alburnus* (Kirin, 2003); of *Sq. orpheus* and *A. alburnus* (Kirin et al., 2002) from the Arda River; of *Sq. orpheus* from the Chepelarska River (Kirin, 2002a; 2002b); of *Sq. orpheus* from the Arda River (Kirin et al., 2003); of *Sq. cephalus* from the Danube River (Cakic et al., 2004); of *S. erythrophthalmus* from Srebarna Biosphere Reserve (Shukerova and Kirin, 2008); of *Sq. cephalus*, *S. erythrophthalmus*, *B. barbus* from the Danube River (Atanasov, 2012); of *Sq. orpheus* from the Tunja River (Kirin et al., 2013), etc. *C. fennica*, *A. lucii* and *R. denudata* are intestinal parasites in the body of fishes. For all fixed endoparasite species, the common roach is a definitive host.

Component communities

The three determined species, *C.fennica, A. lucii, Rh. denudata*, parasitizing in *R. rutilus* are generalists for the helminth communities of the examined freshwater fish species of the Luda Yana River ecosystem.

With the highest prevalence is distinguished *R. denudata* (P%=62.23), followed by those of *C. brachycollis* (P%=26.67) and *A. lucii* (P%=6.67). This tendency is also preserved in terms of mean intensity (MI) (Table 1).

R. denudata and *C. brachycollis* are core species of the parasite communities of common roach of the Luda Yana River. *A. lucii* is an accidental species of these communities (according to the criteria of Bush et al., 1987). The three determined species of endohelminths are autogenic species of the helminth communities of the common roach from the river.

Infracommunities

Established parasite species are presented with a total of 130specimens. *R. denudata* is distinguished with the highest number of specimens (88 specimens), and *A. lucii*, with the lowest (7 specimens).There are no mixed invasions. The low values for the mean number of species, mean number of specimens and Brillouin's diversity index (HB) are due to low species diversity, a small number of specimens of fish and low mean intensity of specimen of *R. rutilus* (Table 1).

For Bulgaria there are a few studies on parasites of *R. rutilus* and a total of 15 species of helminthes from the digestive tract of the roach were reported (Atanasov, 2012; Kakacheva-Avramova, 1983; Shukerova, 2010, etc.). Of these, two species are found in the present study. The third species, *R. denudata*, is a new endoparasite species for *R. rutilus* in the country. Consequently, the roach of the Luda Yana River is represented by 18.75% of the total found for *R. rutilus* intestinal parasite species.

Table1. Species diversity, prevalence (P%), mean intensity (MI) of the established endohelminth species of *Rutilus rutilus* from the Luda Yana River

Species of endoparasites	Ecological indices (N[1]=45)			
	n[2]	p[3]	P%[4]	MI[5] Range
Cestoda				
Caryophyllaeidae				
Caryophyllaeides fennica (Schneider, 1902) Nybelin, 1922	12	35	26.67	2.92 1-7
Acanthocephala				
Echinoderhynchidae				
Acanthocephalus lucii (Mueller, 1776)	3	7	6.67	2.34 1-3
Nematoda				
Rhabdochonidae				
Rhabdochona denudata (Dujardin, 1845) Raillet, 1916	28	88	62.23	3.14 1-10
Total number of species (Mean number of species±SD)	3 (0.95±0.2)			
Total number of specimens (Mean number of specimens±SD)	130 (2.88±2.29)			
Brillouin's diversity index (HB)	0.74±0.79			

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

According to a number of authors, the species developed with intermediate hosts were reported with low indices of infection in ecosystems with negative impacts and parasitological studies of specific parasite species of freshwater fish can be used as bioindicators for environmental conditions (Rakauskas and Blaevièius, 2010; Valtonen et al., 2003, etc.).

The results obtained from the study and the knowledge of the biology of the established parasite species reveals the following main pathways of the parasitic flow: A. Cestoda: 1. Oligohaetes – Fishes (*Caryophyllaeides*

fennica); B. Acanthocephala: 1. Crustaceans – Fishes (*Acanthocephalus lucii*); C. Nematoda: 1. larvaes of Ephemeroptera and Diptera – Fishes (*Rhabdochona denudata*). Determined parasite species and ecological characteristics of parasite communities show that the larvae's of Ephemeroptera and Diptera are dominant in the nutrition of the roach. Probably their populations are well represented in the studied ecosystem of the river ecosystem.

CONCLUSIONS

From studied 45 specimens of common roach, 3 parasite species are fixed: *C. brachycollis*, *A. lucii* and *R. denudata*. The Luda Yana River is a new locality for all of them. *R. rutilus* is a new host record for the endohelminth species *R. denudata*. Poor species diversity and low indices of invasion indicate for negative impacts on biodiversity of the Luda Yana River ecosystem.

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