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HELMINTHS AND HELMINTH COMMUNITIES OF *Perca fluviatilis* (Linnaeus, 1758) AND *Vimba melanops* (Heckel, 1837) FROM MARITSA RIVER, BULGARIA

Dimitrinka KUZMANOVA, Mariya CHUNCHUKOVA, Diana KIRIN

Agricultural University - Plovdiv, Department of Agroecology and Environmental Protection, 12 Mendeleev Blvd, Plovdiv, 4000, Bulgaria

Corresponding author email: dima_kuzmanova@abv.bg

Abstract

As a result of the study of 10 specimens Perca fluviatilis Linnaeus, 1758 and 10 specimens Vimba melanops (Heckel, 1837) from the Maritsa River, Aegean water basin, Bulgaria, infection with 5 helminth species are presented (Allocreadium isoporum (Loos, 1894); Proteocephalus percae (Müller, 1780); Caryophyllaeus laticeps (Pallas, 1781); Contracaecum sp., larvae and Acanthocephalus lucii (Müller, 1776)). All identified parasite species are autogenous to the parasite communities of the perch and Macedonian vimba, except Contracaecum sp.*, which is an allogeneic species. Infection indices are discussed and pathways of helminth flux circulation are traced. The dominant structure of the helminth communities was analyzed. New data on the helminths and their communities in the two species of freshwater fish, as well as on the ecological status of the studied biotopes of the freshwater ecosystem are presented.*

Key words*: Aegean water basin, ecological status, helminth communities, Macedonian vimba, perch.*

INTRODUCTION

The Maritsa River, Aegean water basin, springs from the territory of Bulgaria and is one of the longest rivers (472 km, of which 322 are on Bulgarian territory) in the country, after the rivers Danube, Iskar, and Tundzha. The river begins at 2378 meters above sea level in the Rila Mountain, from the two Marichini lakes, located between the peaks of Mancho (2771 m, a.s.l.), Marishki chal (2765 m a.s.l.) and Bliznacite (2779 m a.s.l.). The Maritsa River flows into the Aegean Sea (Valkanov, 2000). The rapidly increasing anthropogenic impact disrupts the ecological balance in nature. Freshwater ecosystems are particularly sensitive to negative impacts. Maritsa river is also under the such influence (pollution from industry, agriculture, household, etc.). The river is a habitat for many valuable and globally protected animals and plants. The Maritsa River has the role of a hydroclimatic pathway for the penetration of Mediterranean and sub-Mediterranean influence, which favors the presence of rich flora and fauna. Along the river, protected areas BG0000578 "Maritsa River" have been declared under the Habitats Directive (Directive 92/43/EEC), as well as BG0002081 Maritsa-Parvomai and BG0002087 Maritsa-Plovdiv under the Birds Directive (Directive 79/409/EEC) as part of the National and Ecological Network NATURA 2000 (https://natura2000.egov.bg/EsriBg.Natura.Public.Web.App).

Parasites are part of biological diversity. Parasitic organisms are considered to constitute the richest group of species on Earth. They testify to the state of the food chains in the respective habitats because most of them have complex development cycles associated with the participation of free-living organisms and intermediate hosts. Some parasites can accumulate heavy metals and other pollutants, which they biomagnify and transmit through food chains. Many studies have established both losses of fish resources and dangerous parasites for the ichthyofauna and humans but also the need for parasite conservation (Gómez & Nichols, 2013; Auld & Tinsley, 2015; Biswal, 2020; Carlson et al., 2020, etc.).

The study presents new data on the helminths and helminth communities of *Perca fluviatilis* (Linnaeus, 1758) and *Vimba melanosps* (Heckel, 1837), as well as data on the ecological status of the studied biotopes.

MATERIALS AND METHODS

A total of 10 specimens *Perca fluviatilis* Linnaeus, 1758 and 10 specimens *Vimba melanosps* (Heckel, 1837) were examined for helminths. The fish are caught with fishing rods from the river Maritsa in the vicinity of the village of Milevo. The names of the fish are presented according to the FishBase database (Fröse & Pauly, 2022). Helminthological examinations are carried out by the methods described by Petrochenko, 1956; Zashev & Margaritov, 1966; Bauer (Ed.), 1987; Moravec, 2013, etc. Helminth specimens were fixed and preserved with 70% ethyl alcohol in Eppendorf tubes. For the specimens of classes Trematoda and Cestoda, the methods of Georgiev et al., 1986; Scholz & Hanzelová, 1998 were used and for classes, Acanthocephala and Nematoda – the methods of Zashev & Margaritov, 1966; Moravec, 2013 were used. Analyses of the helminth community structure have been performed in both levels: infracommunity (total and mean number of species; total and mean number of specimens; Brillouin's index of diversity - HB) and component community (prevalence (P%) and mean intensity (MI) for each species) (Bushatal, 1997; Kennedy, 1993, 1997; Magurran, 1988). The species are divided into core species (P%>20), component species (P%>10), and accidental species (P%<10) (Kennedy, 1993). The diversity measures are calculated by software products Statistica 10 (StatSoft Inc., 2011) and MS Excel (Microsoft 2010).

RESULTS AND DISCUSSIONS

Model fish species

Macedonian vimba (*Vimba melanops* (Heckel, 1837); Cyprinidae) is a freshwater, brackish, and demersal fish that inhabits rivers of the Aegean watershed (Vardar, Pinios, Struma, Maritsa, etc.) and its tributaries. The species mainly prefer the middle and less often the lower reaches of rivers with sandy and gravelly bottoms. *V. melanops* reproduces during the period of May-June with characteristic migrations to the upper courses of the rivers, reaching sexual maturity at 2-3 years. Macedonian vimba spawns in stony and gravelly places. It feeds mainly on benthic invertebrates and algae. It reaches a maximum of 30-40 cm in length (Kottelat & Freyhof, 2007; Zhivkov & Karapetkova, 2009). The species appears in the IUCN Red list with the category Data Deficient (DD, IUCN) (Froese & Pauly, 2022). *V. melanops* is included in the Red Book of Bulgaria, volume 2. Animals, with a nature conservation status of vulnerable (VU, IUCN) (Golemanski (Ed.), 2011). *V. melanops* is an endemic species to the Aegean watershed (Kolev, 2013).

Perca fluviatilis Linnaeus, 1758 (Percidae) is a freshwater, brackish, and demersal fish species, which inhabits freshwater ecosystems in the temperate and subtropical zones in North America and Eurasia. Perch can be found in rivers, streams, and lakes in almost all of Europe. It is a typical predator. Larvae feed on zooplankton and adults feed on larvae of other fish species, insects, and small fish. The species lives in schools but is not a territorial fish. It hunts during the day. Males reach sexual maturity at 2-3 years, and females at 3-6 years. The perch reproduces in the period April-May, at a temperature not lower than 7-8°C (Kottelat & Freyhof, 2007; Zhivkov & Karapetkova, 2009). The species appears in the IUCN Red list with the category Least Concern (LC, IUCN) (Froese & Pauly, 2022).

Helminths and helminth community structure

As a result of the ecologoparasitological examinations of 10 specimens of perch (*P. fluviatilis*) and 10 specimens of Macedonian vimba (*V. melanops*), infection with 5 taxa of helminths was found: *Allocreadium isoporum* (Loos, 1894); *Proteocephalus percae* (Müller, 1780); *Caryophyllaeus laticeps* (Pallas, 1781); *Contracaecum* sp., larvae; *Acanthocephalus lucii* (Müller, 1776). Found helminth species belong to classes Trematoda (1) and Cestoda (2); Nematoda (1) and Acanthocephala (1), respectively. Macedonian vimba is represented by a bigger number of helminth species in helminth communities (three helminth species) than perch (two helminth species).

The development of *All. isoporum* is carried out with the participation of two intermediate hosts - snails of the genus *Sphaerium* Scopoli, 1777 and insect larvae of the genera *Ephemera* Linnaeus, 1758, *Anabolia* Stephens, 1837,

Chaetopterix Cuvier, 1827. Definitive hosts are mainly carp fish (Cyprinidae). Definitive hosts of *Pr. percae* mainly are *Gymnocephalus cernua* (Linnaeus, 1758), *P. fluviatilis* and *Esox lucius* Linnaeus, 1758, etc. Intermediate hosts are copepods from the genus *Cyclops* Müller, 1785. The development of *C. laticeps* is carried out with the participation of intermediate hosts *Tubifex tubifex* (Müller, 1774), *T. barbatus* (Grube, 1860), *Limnodrilus claparedeanus* Ratzel, 1868. Definitive hosts are freshwater fish from Cyprinidae, with specific hosts from the genus *Abramis* Cuvier, 1816. The intermediate host of *Ac. lucii* is *Asellus aquaticus* (Linnaeus, 1758) and definitive hosts are mainly freshwater fish species from Cyprinidae, rarely from Percidae, Siluridae, Salmonidae, Esocidae, Gadidae, Cobitidae, Anquillidae. Definitive hosts of *Contracaecum* sp. are fish-eating birds from *Ardea* Linnaeus, 1758, and *Nicticorax* Forster, 1817. Intermediate hosts are fish species from Cyprinidae, Percidae, Clupeidae, etc. (Petrochenko, 1956; Zashev & Margaritov, 1966; Kakacheva-Avramova, 1983; Bauer (Ed.), 1987; Moravec, 2013, etc.) (Table 1).

The intermediate hosts of established helminth taxa are detritophages (DF = deposit feeders) (Belkinova et al., 2013). The representatives of the genus *Cyclops* are bioindicators for β-α-mesosaprobity in examined habitats; *T. tubifex* and *T. barbatus* - for p-saprobity; *L. claparedeanus* - for p-α-mesosaprobity, and *A. aquaticus* is a bioindicator for α-mesosaprobity. In addition to being bioindicators of saprobity, some scientific studies show that these intermediate hosts are capable of accumulating heavy metals, which easily biomagnetize and reach higher organisms along the food chain, which poses a danger to freshwater fish and their consumers – fish-eating birds, humans, etc. (Ali & Fishar 2005; Maltby, 1991, etc.; Belkinova et al., 2013; Łuszczek-Trojnar et al., 2014).

Component community

Ac. lucii and *Contracaecum* sp. were infected examined fish hosts in 100%, respectively. The highest mean intensity in helminth communities of *V. melanosps* is distinguished for *Contracecum* sp., larvae (MI=8.5), followed by those of *C. laticeps* (MI=3.0) (Table 1).

Table 1. Biodiversity, mean intensity, the prevalence of parasites

Helminth species	Intermediate hosts	Definitive hosts			
		Perca fluviatilis Linnaeus, 1758 (N=10)		*Vimba melanops* (Heckel, 1837) (N=10)	
		P%	n/p Range MI	P%	n/p Range MI
Trematoda					
1. *Allocreadium isoporum* (Loos, 1894)	Mollusca, Insecta	-	-	10%	½ 2 2.0
Cestoda					
2. *Proteocephalus percae* (Müller, 1780)	Copepoda	30%	3/7 1-4 2.27	-	-
3.*Caryophyllaeus laticeps* (Pallas, 1781)	Oligocheta	-	-	50%	5/15 1-4 3.0
Nematoda					
4. *Contracaecum* sp., larvae	Crustacea, Copepoda, Cyprinidae	-	-	100%	10/85 1-16 8.5
Acanthocephala					
5. *Acanthocephalus lucii* (Müller, 1776)	Amphipoda	100%	10/28 1-4 2.8	-	-

Mean intensities of *Pr. percae* and *Ac. lucii* in helminth communities of perch are almost the same (MI=2.7 and MI=2.8, respectively) (Table 1). All established parasite species are autogenous to the parasite communities of *P. fluviatilis* and *V. melanops*, except for *Contracaecum* sp., which is an allogeneic species. The high mean intensity of infection with *Contracecum* sp. can lead to significant losses of fish resources as well as human health problems (Zashev & Margaritov, 1966; Zaharieva, 2022).

Infracommunity

Ten specimens of perch and ten specimens of Macedonian vimba were infected with one helminth species (100%). There are no uninfected fish specimens from both fish species. Macedonian vimba was infected with three species of helminth and also bigger specimens than perch (104 and 35 specimens, respectively). In perch, specimens infected with one species of helminth predominate (70%), while in Macedonian vimba, specimens infected with two types of helminth predominate (60%). Brillouin's indices of diversity are HB=0,45 (*P. fluviatilis*) and HB=0.48 (*V. melanosps*) (Table 2).

Table 2. Infracommunity data

***Perca fluviatilis* Linnaeus, 1758**		
Number of helminth species		
Total number	2	
Number of infected fish	7	3
Number of helminth species	1	2
Number of helminth specimens		
Total number	35	
Mean ± SD	2.69±1.25	
Range	1-4	
Mean HB ± SD	0.45±0.29	
***Vimba melanops* (Heckel, 1837)**		
Number of helminth species		
Total number	3	
Number of infected fish	4	6
Number of helminth species	1	2
Number of helminth specimens		
Total number	104	
Mean ± SD	6.38±5.86	
Range	1-16	
Mean HB ± SD	0.48±0.31	

Discussions

P. fluviatilis helminths found in this study (*Pr. percae* and *Ac. lucii*) have been reported in previous studies on the freshwater ecosystem and perch (Table 3).

Table 3. Endohelminth species of *Perca fluviatilis and Vimba melanops* reported from other studies Aegean water basin, Bulgaria

Parasite species	Authors	Host (Locality)
***Perca fluviatilis* Linnaeus, 1758**		
Cestoda		
Proteocephalus percae (Müller, 1780)	Todorova-Traykova & Chunchukova, 2018	*P. fluviatilis* (rezervoir Batak)
	Kuzmanova et al., 2019	***P. fluviatilis*** **(river Maritsa)**
Acanthocephala		
Acanthocephalus lucii (Müller, 1776)	Margaritov, 1959	*Silurus glanis*, *Squalius cephalus* (r. Tundhza)
	Kuzmanova et al., 2019	***P. fluviatilis*** **(r. Maritsa)**
	Chunchukova et al., 2019	*Alburnus alburnus* (r. Maritsa)
***Vimba melanops* (Heckel, 1837)**		
Trematoda		
Allocreadium isoporum (Loos, 1894)	Kakacheva-Avramova, 1965	*Barbus cyclolepis*, *Gobio gobio*, *Squalius orphaeus* (rivers Syuyutlijka, Asenitsa, Bedechka, rez. Azmaka, rez. 40-te izvora)
	Margaritov, 1965	*Squalius orphaeus* (**r. Maritsa**), *Barbus cyclolepis* (r. Vacha)
	Kirin, 2000, 2001	*Squalius orphaeus* (**r. Maritsa**)
	Kirin et al., 2019	*Squalius orphaeus* (r. Stryama)
	Kirin et al., 2020	*Barbus cyclolepis* (r. Tamrashka)
Cestoda		
Caryophyllaeus laticeps (Pallas, 1781)	Margaritov, 1959	*Alburnus alburnus* (r. Tundzha)

All. isoporum has been reported for the Maritsa River from other fish species, while *C. laticeps* and *Contracaecum* sp. were not reported for the Maritsa river. *V. melanosps* is reported for the first time as a host of *All. isoporum*, *C. laticeps* and *Contracaecum* sp. in Bulgaria. The helminth species found in this study have been reported also in other studies, mainly referring to the Danube basin: 1). *Pr. percae* of G*ymnocephalus schraetser* (Linnaeus, 1758) (syn. *Acerina schraetser*) *Sander volgensis* (Gmelin, 1789) (syn. *Stizostedion volgense*), *Gymnocephalus cernua* (Linnaeus, 1758) (syn. *Acerina cernua*) (river Danube; Kakacheva-Avramova et al., 1978); of *Perca fluviatilis* Linnaeus, 1758 (lake Srebarna; Shukerova, 2010; Shukerova et al., 2010); of *P. fluviatilis* (r. Danube; Zaharieva, 2022); 2). *Ac. lucii* of *Silurus glanis* Linnaeus, 1758, Squalius cephalus (Linnaeus, 1758) (rivers Danube, Iskar; Margaritov, 1959); of *Ballerus sapa* (syn. *Abramis sapa*) (Pallas, 1814), *Sq. cephalus*, *Rutilus rutilus* (Linnaeus, 1758), S. glanis, *P. fluviatilis*, Lota *lota* (Linnaeus, 1758), *G. schraetser*, *Bentophilus stellatus* (Sauvage, 1874), *Proterorhinus marmoratus* (Pallas, 1814) (r. Danube; Kakacheva-Avramova et al., 1978); of *P. fluviatilis* (lake Srebarna; Shukerova, 2010); of *L. lota*, *Zingel zingel* (Linnaeus, 1766) (r. Danube; Atanasov, 2012); of *Abramis brama* (Linnaeus, 1758) (lake Srebarna; Chunchukova et al., 2016); of *Alburnus alburnus* (Linnaeus, 1758), *Abr. brama* (r. Danube; Chunchukova, 2017); of *Alb. alburnus* (r. Danube; Chunchukova et al., 2019); of *Sq. cephalus* (r. Osym; Kuzmanova et al., 2019); of *Cyprinus carpio* Linnaeus, 1758; of *Neogobius fluviatilis* (Pallas, 1814), *Babka gymnotrachelus* (Kessler, 1857), *Neogobius melanostomus* (Pallas, 1814) (r. Danube; Zaharieva, 2022); 3). *Allocreadium isoporum* of *Gobio gobio* (Linnaeus, 1758), *Barbus petenyi* Heckel, 1852, *Alburnoides bipunctatus* (Bloch, 1782), *Phoxinus phoxinus* (Linnaeus, 1758), *Sq. cephalus* (rivers Vrabniska, Nishava, Iskrecka, Buchinska, Berkovska; Kakacheva-Avramova, 1969); of *B. petenyi*, *Sq. cephalus* (r. Shiposhnitsa, rez. Iskar; Margaritov, 1977); of *Alb. alburnus* (r. Dunav; Kakacheva et al., 1978); of *B. petenyi*, *Barbus barbus* (Linnaeus, 1758) (Kakacheva-Avramova & Nedeva-Menkova, 1981); of *Sq. cephalus* (r. Osym;

Kuzmanova et al., 2019); of *Alb. alburnus*, *Chondrostoma nasus* (Linnaeus, 1758), *Vimba vimba* (Linnaeus, 1758), *R. rutilus* (r. Danube; Zaharieva, 2022); 4). *Caryophyllaeus laticeps* of *B. barbus*, *B. petenyi* (r. Danube, rez. Iskar, respectively; Margaritov, 1959); of *B. barbus* (r. Danube; Margaritov, 1966); of *B. barbus*, *V. vimba*, *Abr. brama*, *Abr. sapa* (r. Danube; Kakacheva et al., 1978); of *B. barbus*, *V. vimba*, *Abr. brama*, *Ch. nasus*, *Esox lucius* Linnaeus, 1758, *S. glanis* (r. Danube; Atanasov, 2012); of *Sq. cephalus* (r. Osym; Kuzmanova et al., 2019); of *V. vimba*, *Abr. brama* (r. Danube; Zaharieva, 2022); 5). *Contracaecum* sp., larvae of Cyprinus carpio (lakeSrebarna; Shukerova, 2006); of *Aburnus alburnus, Aspius aspius, Abramis brama, P. fluviatilis, R. rutilus* (lake Srebarna; Shukerova, 2010); of *Abr. brama* (Srebarna; Chunchukova et al., 2016); of *Alb. alburnus*, *Abr. brama*, *B. barbus* (r. Danube; Chunchukova, 2017); of *B. barbus* (r. Danube; Chunchukova & Kirin, 2018); of *Alb. alburnus* (r. Danube; Chunchukova et al., 2019); of *R. rutilus* (lake Strebarna; Shukerova, Kirin, 2019); of *Abr. brama*, *Alb. alburnus*, *Ch. nasus*, *V. vimba*, *B. sapa*, *Carassius gibelio* (Bloch, 1782), *C. carpio*, *Leuciscus aspius* (Linnaeus, 1758), *Pelecus cultratus* (Linnaeus, 1758), *Scardinius erythrophthalmus* (Linnaeus, 1758), *N. melanostomus*, *P. fluviatilis*, *S. glanis* (r. Danube; Zaharieva, 2022), etc.

CONCLUSIONS

V. melanosps is a new host recors for *All. isoporum*, *C. laticeps* and *Contracaecum* sp. in Bulgaria. Maritsa river is a new locality of *C. laticeps* and *Contracaecum* sp. All taxa of helminths are autogenous for the Maritsa River, except for *Contracaecum* sp.

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REFERENCES

Ali, M.H.H., & Fishar, M.R.A. (2005). Accumulation of trace metals in some benthic invertebrate and fish species relevant to their concentration in water and sediment of Lake Qarun, Egypt. *Egyptian Journal of Aquatic Research*, *31*(1), 289-301.

Atanasov, G. (2012). Fauna, morphology and biology on the endohelminths of fish from Bulgarian part of the Danube River. PhD these, BG: Sofia (In Bulgarian).

Auld, S., Tinsley, M. The evolutionary ecology of complex lifecycle parasites: linking phenomena with mechanisms. *Heredity*, *114*, 125–132

Bauer, O. (Ed.) (1987). *Key to the Parasites of FreshwaterFishes of the USSR*. Leningrad, RU: Nauka (in Russian).

Belkinova, D., Gecheva, G., Cheshmedjiev, S., Dimitrova-Dyulgerova, I., Mladenov, R., Marinov, M., Teneva, I., & Stoyanov, P. (2013). Biological analysis and ecological assessment of surface water types in Bulgaria. Bulgaria: Univ Publ House "P. Hilendarskii" (in Bulgarian).

Biswal, D. (2020). Fish Parasites as Biological Indicators: A Systematic Review. December 2020. *Bioscience Biotechnology Research Communications*, *13*(4), 1743-1755.

Bush, A., Lafferty, K., Lotz, J., Shostak, A. (1997). Parasitology meets ecology on its own terms. *Journal of Parasitology*, *83*, 575-583.

Carlson, C.J., Dallas, T.A., Alexander, L.W., Phelan, A.L., & Phillips, A.J. (2020). What would it take to describe the global diversity of parasites? *Proceedings of Royal Society B. Biologycal Sciences*, *287*, 20201841.

Chunchukova, M., Shukerova, S., & Kirin, D. (2016). Search on the impact of the river Danube on the Srebarna Biosphere reserve the model ecosystem *Abramis brama* – macroinvertebrates – sediments. *Agricultural sciences*, *VIII*(19), 151-158.

Chunchukova, M. (2017). Parasites and parasite communities of fish from the Danube River – ecology, biodiversity and bioindication. PhD these, BG: Plovdiv (In Bulgarian).

Chunchukova, M., & Kirin, D. (2018). New Data on Endohelminth Communities of Barbel *Barbus Barbus* from the Bulgarian Part of the River Danube. *Helminthologia*, *55*(3), 1-8.

Chunchukova, M., Kirin, D. & Kuzmanova D. (2019). Gastrointestinal helminth fauna and helminth communities of bleak (*Alburnus alburnus*, L. 1758) from lower section of Danube River. *Bulgarian Journal of Veterinary medicine*, *22*(3), 344-352.

Chunchukova, M., Kirin, D. & Kuzmanova D. (2019). New data for helminth communities of *Alburnus alburnus* (Linnaeus, 1758) from Maritsa River, Bulgaria. Scientific Papers. *Series D. Animal Science*, *LXII*(1), 439-444.

Georgiev, B., Biserkov, V., & Genov, T. (1986). In toto staining method for cestodes with iron acetocarmine. *Helminthologia*, *23*, 279–281.

Golemanski V. (Ed-in-Chief), 2011. *Red Data Book of the Republic of Bulgaria*. Sofia, BG: Joint edited of the Bulg. Acad. of Sci. and Ministry of Environment and Waters.

Gómez, A. & Nichols, E. (2013). Neglected wild life: Parasitic biodiversity as a conservation target. *Int. J. Parasitol. Parasites Wildl.*, *2*, 222–227.

Froese R., & Pauly D. (Eds.) (2022). *FishBase. World Wide Web electronic publication*. Retrived August, 2022, from www.fishbase.org.

IUCN Red List Status (n.d.). Retrieved from https://www.iucnredlist.org.

Kakacheva-Avramova, D. (1965). Helminthological study of fishes from some water basins in Trakia. *Fauna of Trakia*, *2*, 83-120 (in Bulgarian).

Kakacheva-Avramova, D. (1969). Helminthe on fishes from rivers on western Balkan mountain. II. Trematoda, Cestoda, Acanthocephala, Nematoda. *Rep. Centr. Helm. Lab*., *13*, 61-75 (In Bulgarian).

Kakacheva, D., Margaritov, N., & Grupcheva, G. (1978). Fish parasites of Bulgarian part of the Danube River. *Limnology of Bulgarian part of the Danube River*, *Bulg. Acad. Sci*., 250-271 (In Bulgarian).

Kakacheva-Avramova, D. & Nedeva-Menkova, I. (1981). Contribution to the studies of the helminths of freshwater fishes from Blagoevgrad District. *Helminthology*, *11*, 26-41 (In Bulgarian).

Kakacheva-Avramova, D. (1983). *Helminths of freshwater fishes in Bulgaria*. Sofia, BG: Bul. Acad. Sci. (in Bulgarian).

Karapetkova, M., & Zhivkov, M. (2009). *Fishes in Bulgaria*. Sofia, BG: GeaLibris (in Bulgarian).

Kennedy, C. (1993). The dynamics of intestinal helminth communities in eels *Anguilla anguilla* in a small stream: long-term changes in richness and structure. *Parasitology*, *107*, 71-78.

Kennedy, C. (1997). Freshwater fish parasites andenvironmental quality, an overview and caution. *Parasitologia*, *39*, 249-254.

Kirin, D. A. (2000). Ecologophaunistical study of the helminthological communities of *Leuciscus cephalus* L. from river Maritsa. *Research reports of the Union of scientists in Bulgaria and Humanities*, *1*, 405-408.

Kirin, D. A. (2001). Biodiversity and ecology of the helminths fauna in Leuciscus cephalus from the Maritsa River, Bulgaria. *Trav. Sci. Univ. Plovdiv, Animalia*, *37*(6), 79–84.

Kirin, D., Chunchukova, M., & Kuzmanova, D. (2019). Helminth and helminth communities of Orpheus dace (*Squalius orpheus* Kottelat & Economidis, 2006) from Sryama river, Bulgaria. *Scientific Papers. Series D. Animal Science*, *LXII*(1), 475-480.

Kirin D., Chunchukova, M., Kuzmanova, D., & Paskaleva, V. (2020). Helminths and helminth communities of round-scaled barbel (*Barbus cyclolepis* Heckel, 1837) and its bioindicator role. *Scientific Papers*. *Series D. Animal Science*, *LXIII*(2), 421-426.

Kolev, V. (2013). Species composition of the Ichthyofauna of some tributaries of the Matitza River. *Forestry ideas*, 19, *2*(46), 129-139.

Kuzmanova, D., Chunchukova, M. & Kirin, D. (2019). Helminths and helminth communities of perch (*Perca fluviatilis* Linnaeus, 1758) as bioindicators for ecosystem condition of the Maritsa River. *Scientific Papers. Series D. Animal Science*, *LXII*(1), 463-468.

Kuzmanova, D., Chunchukova, M. & Kirin, D. (2019). Helminth and helminth communities of *Squalius cephalus* (Linnaeus, 1758) from Osym river, Bulgaria. *Scientific Papers. Series D. Animal Science*, *LXII*(1), 456–462.

Łuszczek-Trojnar, E., Sroka, K., Klaczak, A., Nowak, M., & Popek, W. (2014). Bioaccumulation and purification of cadmium in *Tubifex tubifex*. *Turkish Journal of Fisheries and Aquatic Sciences*, *14*, 939-946.

Magurran, A. (1988). *Ecological diversity and its measurement*.London, UK: Cambridge University Press.

Maltby, L. (1991). Pollution as a probe of life-history adaptation in *Asellus aquaticus* (Isopoda). *Oikos*, *61*(1), 11–18.

Margaritov, N., 1959. *Parasites of some freshwater fishes*. Varna, BG: Publishing House NIRRP. (In Bulgarian).

Margaritov, N.M. (1965). Intestinal helminths of fishes of the middle reaches of the R. Maritsa and tributaries. *Yearbook of the Sofia University, Faculty of Biology*, *58*. 129-150 (In Bulgarian)

Margaritov, N., 1966. Helminths of the digestive tract and the abdominal cavity of fishes of the Bulgarian section of Danube River. *Bulletin de L'institut de Zoologie et Musée*, *20*, 157–173 (In Bulgarian).

Moravec, F. (2013). *Parasitic Nematodes of Freshwaterfishes of Europe*. Praha, CZ: Academia.

Petrochenko, V. (1956). *Acanthocephalus domestic and wild animals*. Moskow, RU: AN USSR (in Russian).

Scholz, T., & Hanzelová, V. (1998). Tapeworms of the Genus *Proteocephalus* Wienland, 1858 (Cestoda: Proteocephalidae), parasites of fishes in Europe. Praha, CZ: Academia.

Shukerova, S. (2006). Helminth fauna of the common carp, *Cyprinus carpio* (Linnnaeus, 1758) from the Srebarna Biosphere reserve, Bulgaria. Scientific articles. Ecology 2006, 2, 217–223.

Shukerova, S. (2010). Helmiths and helminth comminities of freshwater fish from Biosphere Reserve Srebarna. Phd these, BG: Sofia (In Bulgarian).

Shukerova, S., Kirin, D., & Hanzelová, V. (2010). Endohelminth communities of the perch, *Perca fluviatilis* (Perciformes, Percidae) from Srebarna Biosphere Reserve, Bulgaria. *Helminthologia*, *47*(2), 99–104.

Statsoft inc. (2011) (n.d.). STATISTICA (dataanalysissoftwaresystem), version 10.retrieved from www.statsoft.com.

Todorova-Traykova, M., & Chunchukova, M. (2018). Helminth fauna of Perca fluviatilis from Batak reservoir. *Agricultural Sciences*, *10*(24), 35-40.

Valkanov, V. (2000). *Maritime history of Bulgaria*. Sofia, BG: Albatross (In Bulgarian).

Zashev, G., & Margaritov, N. (1966). *Diseases of fish*. Sofia, BG: Nauka i izkustvo (in Bulgarian).

https://natura2000.egov.bg/EsriBg.Natura.Public.Web.pp