

NEW DATA FOR HELMINTH FAUNA OF *Esox lucius* (Linnaeus, 1758) FROM MARITSA RIVER, BULGARIA

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Abstract

Twelve specimens of northern pike (*Esox lucius* (Linnaeus, 1758)) were caught from the Maritsa River during 2020-2021 and examined for parasites. Helminth parasites were found in 50% of the examined northern pikes (6 specimens) from the Maritsa River. Two species of parasites were fixed: one trematode species (*Bunodera luciopercae* (Müller, 1776)) and one nematode species (*Raphidascaris acus* (Bloch, 1779)). The dominant structure of the established species is represented at the level of component communities and infracommunities. *R. acus* is a core species for the component community of *Esox lucius* from the Maritsa River. *B. luciopercae* is an accidental parasite species for northern pike helminth communities. The data for the infracommunities were used to determine the basic biotic indices. The bioindicator significance of the established parasite species was discussed for an ecological assessment of the studied river ecosystem. As a result of the conducted research, new data on the parasite fauna of *E. lucius* from the Maritsa River have been presented.

Key words: bioindication, *Esox Lucius*, helminths, Maritsa River.

INTRODUCTION

The longest river on the Balkan Peninsula is the Maritsa River (427 km), the upper and middle reaches of which are located on the territory of Bulgaria (321.6 km). The Maritsa River springs from Rila Mountain. East of Svilengrad, the river flows eastward, forming the border between Bulgaria on the northern coast and Greece on the southern coast and then between Turkey and Greece. The two main tributaries, Tunja and Arda, flow into Edirne (Turkey), and the river pass through Turkish territory on both banks. The Maritsa then turns south and forms the border between Greece on the west coast and Turkey on the east coast reaching the Aegean Sea, which enters near the Enez.

Parasites are considered an essential and necessary component of biodiversity (Dobson et al., 2008). They are also valuable indicators for food web structure and ecosystem state (Marcogliese, 2004; Marcogliese, 2005; Hudson et al., 2006). The complex life cycles of endoparasites reflect relationships with a range of invertebrate and vertebrate hosts. The assemblage of these different parasites in a host organism reflects not only the trophic position of that host in the food web but also the

presence in the ecosystem of any other organisms that participate in the different life cycles of parasites.

This study aims to reveal parasite species biodiversity and structure of parasite communities of *Esox lucius* from the Maritsa River, Bulgaria.

MATERIALS AND METHODS

Twelve specimens of northern pike (*Esox lucius* (Linnaeus, 1758)) were caught from the Maritsa River (Plovdiv region) in the two years 2020-2021 and examined for parasites. The object of the present study is a section of the Maritsa River near the village of Orizari (Rhodopi municipality, Plovdiv region). After the mouth of the Luda Yana River to the state border with the Republic of Greece and the Republic of Turkey, the Maritsa River belongs to river type R12: Large lowland rivers in Ecoregion 7 (Eastern Balkans).

A total of twelve *Esox lucius* (Pisces: Esocidae) are collected from the Maritsa River in the vicinity of the village Orizari and examined for parasites. Fish are caught with a fishing line during the two years of 2020-2021. The studied fish specimens were measured (mean total

length 41.08±2.40 cm; range 39-44 cm) and weighed (mean total weight 1.105±0.38 kg; range 0.950-1.4 kg). The fish specimens were examined with standard techniques immediately after capture. Trematodes were stained with acetic carmine, differentiated in 70% acid ethanol, dehydrated in ascending ethanol series, cleared in eugenol and mounted in Canada balm as permanent slides (Bykhovskaya-Pavlovskaya, 1985; Georgiev et al., 1986). The identification of parasite samples was made using the keys of Bauer et al. (1981), Bauer (1987), and Bykhovskaya-Pavlovskaya (1985). Nematodes were examined in glycerine as temporary microscopic preparations (Moravec, 2013). The common and taxonomic name of fish is used in accordance with Fröse and Pauly (2022). The mean intensity (MI), mean abundance (MA) and prevalence (P%) were used according to Bush et al. (1997). The helminth community structure was studied at levels of component community and infracommunity. Based on the prevalence (P%) as suggested by Kennedy (1993), the parasites are grouped as core (P%>20), component (P%<20) and accidental (P%<10).

RESULTS AND DISCUSSIONS

During 2021-2022, twelve specimens of *Esox lucius* (Linnaeus, 1758) were collected from the Maritsa River and examined for helminths. The northern pike has Holarctic distribution. In Bulgaria, the species is widespread in the Danube River and the lower reaches of its

tributaries, in the Kamchia River, in the rivers of the Aegean Basin, in dams and other stagnant and slow-flowing waters (Karapetkova & Zivkov, 2010). *Esox lucius* is not listed in the Bulgarian Red Data Book (Golemanski (Ed.), 2011) and is rated as LC=Least Concern species (IUCN Red List Status). The northern pike is a freshwater, predatory brackish, potamodromous species (Fröse & Pauly, 2022). *Esox lucius* is typical predatory species that feed on fish and other vertebrates, feeding on zooplankton only in its earliest stages (Karapetkova & Zivkov, 2010). The northern pike is highly territorial and usually solitary (Fröse & Pauly, 2022). Habitat alternations can impact *Esox lucius* locally (Kottelat & Freyhof, 2007).

Parasites were found in 50.00% (Table 1) of the examined northern pikes from the Maritsa River (6 fish). Two parasite species were identified, which were represented by 13 specimens (p), one species belonging to the class Trematoda (*Bunodera luciopercae* (Müller, 1776)) and one to the class Nematoda (*Raphidascaris acus* (Bloch, 1779)). All established species are adults.

In the component community of the northern pike from the Maritsa River, nematodes are represented by the largest number of specimens - 11 belonging to one species. One species with only two specimens represent trematodes. *Raphidascaris acus* is a core species (P%=50.00). The prevalence of *Bunodera luciopercae* (P%=8.30) determines it as an accidental parasite species for the communities of northern pike (Table 1).

Table 1. Parasite diversity of *Esox lucius* from Maritsa River (N - Number of studied fish, n - Number of infected fish)

Helminth species	N = 12					
	n	p	P%	MA±SD	MI±SD	Range
<i>Bunodera luciopercae</i> (Müller, 1776)	1	2	8.3	0.17±0.55	2.0±0.0	0-2
<i>Raphidascaris acus</i> (Bloch, 1779)	6	11	50	0.92±1.19	1.83±1.07	1-4

Species richness in infracommunity of northern pike ranged from 1 to 2 species. Five fish (41.66%) were infected with one helminth species and only one (8.33%) with two helminth species. The largest number of helminths found in one host is 4. The

infracommunity of northern pike is presented by 0.58±0.64 species per host and 1.08±1.32 helminths per host (Table 2). The infracommunities of *E. lucius* from the Maritsa River showed low Brillouin's diversity index, HB=0.278.

Table 2. Parameters of the infracommunities of *Esox lucius*

	Number of endohelminth species				
	0	1	2	Mean±SD	Range
<i>Esox lucius</i>	6	5	1	0.58±0.64	0-2
	Number of endohelminth specimens				
	Total number	Mean±SD	Range	Brillouin's index HB	
<i>Esox lucius</i>	13	1.08±1.32	1-4	0.278	

Bunodera luciopercae is an intestinal parasite of fish, representatives of families Percidae, Gadidae, Siluridae, etc. The development of *Bunodera luciopercae* is accomplished with the precipitation of two intermediate hosts. The first is mollusc - *Sphaerium corneum* (Linnaeus, 1758) and *Sphaerium rivicola* (Lamarck, 1818), and the second are crustaceans - *Thermocyclops oithonoides* (Sars,

1863), *Daphnia pulex* (Leydig, 1860), *Mesocyclops crassus* (Fischer, 1853), *Acanthocyclops vernalis* (Fischer, 1853) (Kakacheva-Avramova, 1983).

For Bulgaria, there is data for seven fish species belonging to 4 families as hosts for *Bunodera luciopercae* (see Table 3). River Maritsa is a new locality for *Bunodera luciopercae* in Bulgaria.

Table 3. Overview of registered in Bulgaria hosts of *Bunodera luciopercae* and their locality

Fish host	Locality	References
Family Esocidae		
<i>Esox lucius</i>	Danube River	Kakacheva-Avramova (1977)
Family Salmonidae		
<i>Salmo trutta fario</i>	Arda River	Kirin (2002)
Family Percidae		
<i>Perca fluviatilis</i>	Danube River	Kakacheva-Avramova (1977)
	Arda River	Kirin (2005)
	Srebarna Lake	Shukerova et al. (2010)
<i>Sander lucioperca</i>	Danube River	Margaritov (1959)
		Margaritov (1966)
		Atanasov (2012)
<i>Zingel zingel</i>	Danube River	Kakacheva-Avramova et al. (1978)
<i>Zingel streber</i>	Danube River	Kakacheva-Avramova et al. (1978)
Family Cyprinidae		
<i>Alburnus alburnus</i>	Danube River	Atanasov (2012)

For the conditions of Europe, the typical definitive host of *R. acus* is the northern pike (*E. lucius*) and quite often also the *Salmo trutta fario* (Moravec, 2013).

Raphidascaris acus has a very complex life cycle that may include: exogenous development, development in invertebrates,

and development in intermediate host vertebrates and also paratenic hosts (both for second-stage larva and for third-stage larva) and postcyclic hosts (see Moravec, 2013). This nematode species was reported from Bulgaria from 10 fish hosts (Table 4) from different localities.

Table 4. Overview of fish hosts of *Raphidascaris acus*

Fish host	Locality	References
<i>Esox lucius</i>	Maritsa River	Kirin (2013)
<i>Abramis brama</i> *	Danube River	Chunchukova et al. (2017)
<i>Neogobius kessleri</i> *	Danube River	Ondračková et al. (2006)
<i>Neogobius melanostomus</i> *	Danube River	Francová et al. (2011)
<i>Cyprinus carpio</i> *	Srebarna Lake	Shukerova (2006)
<i>Perca fluviatilis</i> *	Srebarna Lake	Shukerova et al. (2010)
<i>Barbus barbus</i> *	Danube River	Chunchukova & Kirin (2018)
<i>Carassius gibelio</i> *	Srebarna Lake	Shukerova (2005)
<i>Chondrostoma nasus</i> *	Danube River	Zaharieva & Kirin (2020).
<i>Salmo trutta fario</i>	Tamrashka River	Kirin et al. (2020)

*The fish species reported as host of the larval stage of *Raphidascaris acus*

The parasite fauna of *Esox lucius* in Bulgaria was subject to ecogoparasitological investigation in previous studies only from two river ecosystems – Danube River (Margaritov, 1959; Margaritov, 1964; Kakacheva-Avramova, 1977; Atanasov, 2012), and Maritsa River (Kirin, 2006; Kirin, 2013). The spectrum of northern pike parasites differed depending on the number of examined fish, the study site, and various ecological factors. The number of parasite species reported by the above papers ranged between one (Margaritov, 1964) and seven taxa (Kakacheva-Avramova, 1977) (see Table 5). In the earliest study of the parasite fauna of *Esox lucius* from the same river ecosystem, the fish host was free of parasites (Margaritov, 1965). The number and diversity of parasite species reflect the study site, its ecological factors, and the behaviour of the

examined specimens. It must be taken into consideration that the northern pike is a selective predator that can influence the fish communities and, at the same time, is influenced by other predators and the available prey (see Craig, 2008). For example, some studies reveal the effects of prey size and gape-size limitation on *Esox lucius* behaviour (Nilsson & Brönmark, 1999; Nilsson & Brönmark, 2000).

For Bulgaria, there is also data for myxosporean parasites of *Esox lucius* from the specific ecosystem of the marshes of Belene Island.

Kakacheva-Avramova et al. (1978) reported *Myxidium lieberkuhni* Bütschli, 1882, *Henneguya lobosa* (Cohn, 1895), *Henneguya psorospermica* Thelohan, 1895 with host *Esox lucius* from the marshes of Belene Island.

Table 5. List of parasite species of *Esox lucius* and their locality registered in river ecosystems in Bulgaria

References Parasite species	Margaritov (1959)	Margaritov (1964)	Kakacheva- Avramova (1977)	Atanasov (2012)	Kirin (2006)	Kirin (2013)	This study
<i>Diplostomum pseudospathaceum</i>				*			
<i>Azygia lucii</i>	*		*				
<i>Bunodera luciopercae</i>			*				*
<i>Tetraonchus monenteron</i>	*		*		*	*	
<i>Triaenophorus nodulosus</i>		*	*				
<i>Triaenophorus meridionalis</i>			*	*			
<i>Acanthocephalus tenuirostris</i>					*		
<i>Pomphorhynchus laevis</i>			*	*	*		
<i>Raphidascaris acus</i>						*	*
<i>Contracaecum bidentatum</i>			*				
<i>Caryophyllaeus laticeps</i>				*			
Locality	Danube	Danube	Danube	Danube	Maritsa	Maritsa	Maritsa

CONCLUSIONS

As a result of the examination for parasites of twelve northern pikes from the Maritsa River, thirteen specimens belonging to two classes were found. One from class Trematoda (*Bunodera luciopercae* (accidental species)) and one from class Nematoda (*Raphidascaris acus* (core species)). *Bunodera luciopercae* is reported for the first time in the river ecosystem

of Maritsa. Thereby River Maritsa is a new locality for *Bunodera luciopercae* in Bulgaria.

ACKNOWLEDGEMENTS

This research was carried out with the support of Agricultural University - Plovdiv, providing the laboratory and technical equipment.

REFERENCES

- 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).
- Bauer, O. N., Musselius, V. A. & Strelkov, Yu. A. (1981). *Diseases of pond fish*. Moscow, RU: Legkaya Pishcheyaya Promishlenost' Publishing House (In Russian).
- Bauer, O.N. (1987). *Key to the parasites of freshwater fishes in the fauna of the U.S.S.R.* Leningrad, RU: Academy of Sciences Publishing House.
- Bush, A., Lafferty, K., Lotz, J. & Shostak A. (1997). Parasitology meets ecology on its own terms. *Journal of Parasitology*, 83, 575-583.
- Bykhovskaya-Pavlovskaya, I. (1985). *Parasites of fish. Manual on study*, Leningrad, RU: Nauka Publishing House (In Russian).
- Chunchukova, M., Kirin, D., Kuzmanova, D. & Shukerova, S. (2017). Accumulation of lead in *Abramis brama* and its parasite *Pomphorhynchus tereticollis* from Danube river (Vetren area), Bulgaria. *Scientific Papers. Series D. Animal Science*, LX, 327-332
- Chunchukova, M. & Kirin, D. (2018). New data on endohelminth communities of barbel *Barbus barbus* from the Bulgarian part of the River Danube. *Helminthologia*, 55, 222-229.
- Craig, J. F. (2008). A short review of pike ecology. *Hydrobiologia*, 601, 5–16.
- Dobson, A., Lafferty, K. D., Kuris, A. M., Hechinger, R. F., & Jetz, W. (2008). Homage to Linnaeus: How many parasites? How many hosts? *Proc. Natl. Acad. Sci.*, 105, 11482-11489.
- Francová, K., Ondračková, M., Poláčik, M., & Jurajda, P. (2011). Parasite fauna of native and non-native populations of *Neogobius melanostomus* (Pallas, 1814) (Gobiidae) in the longitudinal profile of the Danube River. *J. Appl. Ichthyol.*, 27, 879-886.
- Fröse, R., & Pauly, D. (Eds.) (2022). *FishBase*. World Wide Web electronic publication, www.fishbase.org (28 November 2022, date last accessed).
- 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, Vol. 2. – Animalia (In Bulgarian).
- Hudson, P.J., Dobson, A.P., & Lafferty, K.D. (2006). Is a healthy ecosystem one that is rich in parasites? *Trends Ecol Evol*, 21, 381-385.
- IUCN Red List Status, (n.d.) www.iucnredlist.org
- Kakacheva-Avramova, D. (1977). Studies on helminths of fishes in the Bulgarian section of the Danube River. *Helminthologia*, 3, 20-45 (In Bulgarian)
- Kakacheva-Avramova, D. (1983). *Helminths of freshwater fishes in Bulgaria*. Sofia, BG: Bulgarian Academy of Sciences (In Bulgarian).
- Kakacheva-Avramova, 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).
- Karapetkova, M., & Zhivkov, M. (2010). *Fishes in Bulgaria*. Sofia, BG: GeaLibris Publishing House (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.
- Kirin, D. (2002). Biodiversity and ecological peculiarities of the helminth fauna in *Salmo trutta fario* from Arda River, Bulgaria. *Comptes rendus de l'Académie bulgare de Sciences*, 55(7), 83-88.
- Kirin, D. (2005). Ecological research of fishes and appraisal of the condition of the Freshwater 468 ecosystems from the Arda river, Bulgaria. *Journal of Environmental Protection and Ecology*, 6 (1), 91-96
- Kirin, D. (2013). Helminth communities and ecological appraisal for the condition of the Maritsa River, Bulgaria. *AgroLife Scientific Journal*, 2(1), 197–202.
- Kirin, D., Chunchukova, M., Kuzmanova, D., & Paskaleva, V. (2020). Helminths and helminth communities of the brown trout (*Salmo trutta fario*, Linnaeus, 1758) from the Tamrashka River, Bulgaria. *Scientific Papers. Series D. Animal Science*, LXIII(1), 489-494.
- Kirin, D.A. (2006). Biodiversity of the helminth species and helminth communities of *Esox lucius* (L., 1758) from Maritsa River, Bulgaria. *The 35th International Scientific Communications Session of the Faculty of Animal Science, Bucharest, Romania*, 135 – 140.
- Kottelat, M. & J. Freyhof, 2007. *Handbook of European freshwater fishes*. Berlin, GE: Publications Kottelat, Cornol and Freyhof Publishing House.
- Marcogliese, D.J. (2004). Parasites: Small players with crucial roles in the ecological theater. *Ecohealth*, 1, 151–64.
- Marcogliese, D.J. (2005). Parasites of the superorganism: Are they indicators of ecosystem health? *International Journal for Parasitology*, 35, 705–16.
- Margaritov, N. (1964). Ichthyoparasitenfauna des Stauesees „Batak“. – *Godishnik na Sofijskya Universitet*, BGGF, Kniga 1, Biologiya (Zoologiya), 56, 105–123 (In Bulgarian).
- Margaritov, N., 1959. *Parasites of some freshwater fishes*. Varna, BG: NIRRP Publishing House. (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).
- Margaritov, N.M. (1965). Intestinal helminths of fishes of the middle reaches of the R. Maritsa and tributaries. *Godshnik na Sofijskya universitet Biologicheski fakultet*, 58, 129-150 (In Bulgarian)
- Moravec, F. (2013). *Parasitic Nematodes of Freshwater fishes of Europe*. Praha, CZ: Academia Publishing House.
- Nilsson, A. & Bronmark, C. (1999). Foraging among cannibals and kleptoparasites: effects of prey size on pike behavior. *Behavioral Ecology*, 10 (5), 557–566.
- Nilsson, A. & Bronmark, C. (2000). Prey vulnerability to a gape-size limited predator: behavioral and

- morphological impacts on northern pike piscivory. *Oikos*, 88 (3), 539–546.
- Ondračková, M., Trichkova, T. & P. Jurajda (2006). Present and historical occurrence of metazoan parasites in *Neogobius kessleri* (Pisces: Gobiidae) in the Bulgarian section of the Danube River. *Acta Zoologica Bulgarica*, 58, 399–406.
- Shukerova S. (2005). Helminth fauna of the Prussian carp, *Carassius gibelio* (Bloch, 1782), from the Srebarna biosphere reserve. *Trakia Journal of Sciences*, 3, 33- 40.
- Shukerova, S. (2006). Helminth fauna of the Common Carp, *Cyprinus carpio* (Linnaeus, 1758), from the Srebarna Biosphere Reserve, Bulgaria. *Scientific Articles. Ecology*, Part 2, 217-223.
- Shukerova, S., Kirin, D., & Hanzelova, V. (2010). Endohelminth communities of the perch, *Perca fluviatilis* (Perciformes, Percidae) from Srebarna Biosphere Reserve, Bulgaria. *Helminthologia*, 42(2), 99-104
- Zaharieva, R. & Kirin, D. (2020). Parasites and parasite communities of the common nase (*Chondrostoma nasus* (Linnaeus, 1758)) from the Danube River. *Scientific Papers. Series D. Animal Science*, LXIII(2), 413-420.