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NEW DATA ON PARASITES AND PARASITE COMMUNITIES OF *ALBURNUS ALBURNUS* (LINNAEUS, 1758) FROM THE DANUBE RIVER

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Abstract

In the spring, summer and autumn of 2019, ecoparasitological studies were conducted on 165 specimens of Alburnus alburnus (Linnaeus, 1758) from the Danube River (Koshava, Kudelin and Novo selo villages, Vidin region). The species diversity of parasites of the bleak was determined. Seven species of parasites were identified: 4 species of Trematoda (Nicolla skrjabini (Iwanitzky, 1928), Allocreadium isoporum (Looss, 1894), Posthodiplostomum cuticola (von Nordmann, 1832), Sphaerostomum bramae (Müller, 1776)), one species of Cestoda (Neogryporhynchus cheilancristrotus (Wedl, 1855), larvae), one species of Acanthocephala (Pomphorhynchus laevis (Müller, 1776)) and one species of Nematoda (Contracaecum sp., larvae). In the spring the dominant species was the trematode A. isoporum, detected in 9 specimens of A. alburnus from the Koshava biotope. The dominant parasite species in summer and autumn was the trematode P. cuticola. A. alburnus is a new host for S. bramae and N. cheilancristrotus, from the studied river ecosystem. The structure of the parasite communities of the bleak from the investigated section of the freshwater ecosystem of the Danube River was presented.

Key words*: Alburnus alburnus, Bulgaria, Danube River, parasites, parasite communities.*

INTRODUCTION

The Danube River crosses Europe in a west-east direction (Keckeis and Schiemer, 2002) for 2,857 km. The river reaches a maximum width of 1.5 km (https://www.icpdr.org). The Danube basin covers the territory of nineteen European countries and can be divided into three parts: Upper, Middle and Lower Danube (Hock and Kovdcs, 1987). The Bulgarian section of the Danube River covers 470 km from the Lower Danube (extending between 845 and 375 km of the river). The Lower Danube, together with the Danube Delta, is distinguished by the highest diversity of fish (Polačik et al., 2008). The ichthyofauna of the Danube River includes fish species of different families like Cyprinidae, Siluridae, Esocidae, Percidae, Acipenseridae, Salmonidae (Juhásová et al., 2019). According to Polacik et al. (2008) in Bulgarian territory of the Danube River with the highest prevalence (more than 88 %) are the following three fish species: *Neogobius fluviatilis*, *Alburnus alburnus* and *Neogobius kessleri*. With the highest abundances are *N. fluviatilis* and *A. alburnus*, as well as *Neogobius melanostomus*. The high species richness of fish in the Danube River is the reason for the great abundance of fish parasites (Juhásová et al., 2019).

Parasitism is the most common way of life. Each organism is a host of at least one type of parasite. Parasites can be established in different trophic levels in different food webs. In this way, they provide information on the location of their hosts in the food web and changes in the ecosystems. Parasites, as well as free-living organisms, respond to environmental changes and can be used as indicators of the state of ecosystems (Marcogliese, 2004; 2005).

Parasites are the subject of increasing research on their bioindicator role and their ability to show changes occurring in the environment as a result of various impacts (Sures, 2001). Investigations on parasites of fish from the Danube River are carried out by multiple authors (Cojocaru, 2003; Cojocaru 2007; Cakić et al., 2008; Cojocaru 2009; Nachev and Sures, 2009; Đikanović et al., 2013; Kirin et al., 2013; Kvach et al., 2013; Kirin et al., 2014; Đikanović et al., 2015; Chunchukova et al., 2016; Kvach et al., 2016; Kvach et al., 2017; Chunchukova and Kirin, 2018; Chunchukova et al., 2018; Đikanović et al., 2018; Juhásová et al., 2019; Radačovská et al., 2019). Few authors provide

information on parasites of *Alburnus alburnus* (Linnaeus, 1758) from the Danube River (Cojocaru, 2007; 2009; 2010; Đikanovic et al., 2011; Kirin et al., 2013; Chunchukova et al., 2018).

The purpose of this study is to provide information on the species composition and structure of the parasite communities of *A. alburnus* from the upper Danube River in the Bulgarian section.

MATERIALS AND METHODS

During the spring, summer and autumn of 2019, fishes were collected and examined from the Danube River (biotopes near Koshava village, Kudelin village and Novo selo village, Vidin region) for endoparasites (Figure 1).

Figure 1. Danube River (Kudelin, Novo selo and Koshava villages)

The village of Kudelin (44°11′30″N, 22°40′5″E) is the first settlement in the Bulgarian section of the Danube River (844 river km) in the country. The village of Novo selo (44°10′0″N, 22°47′0″E) is located on the bank of the river (833 river km) and borders it for 3 km. The village of Koshava (44°4′0″N, 23°2′0″E) is located along the Danube River (807 river km). During the year, a total of 165 specimens of *A. alburnus* were collected, of which 63 in the spring, 64 in the summer and 38 in the autumn (Table 1).

Table 1. Number of *Alburnus alburnus* specimens studied by biotopes from the Danube River

Season (N = 165)	Kudelin	Novo selo	Koshava
Spring (N = 63)	32	–	31
Summer (N = 64)	30	34	–
Autumn (N = 38)	22	16	–

The fish were caught under a fishing permit for scientific research by the Executive Agency for Fisheries and Aquaculture, the Ministry of Agriculture, Food and Forests in Bulgaria. Species belonging to the studied fish specimens were determined by Karapetkova and Jivkov (2006); Kottelat and Freyhof (2007).

The scientific name of the species is represented by Froese and Pauly (2019). Metric data (weight (g) in grams, maximum body length (L) in centimetres and maximum body width (H) in centimetres) for all examined specimens *A. alburnus* were determined (Table 2).

Table 2. Metric data (L, H and g) of the examined specimens *A. alburnus* by biotopes from the Danube River

Alburnus alburnus	Spring		Summer		Autumn	
	Kudelin	Koshava	Kudelin	Novo selo	Kudelin	Novo selo
L Average ± SD	10.54 ± 1.04	12.98 ± 0.58	11.18 ± 2.13	12.65 ± 1.46	11.26 ± 0.81	11.34 ± 1.09
H Average ± SD	2.20 ± 0.26	2.82 ± 0.19	2.25 ± 0.44	2.40 ± 0.37	2.16 ± 0.18	2.12 ± 0.23
g Average ± SD	6.56 ± 2.08	16.42 ± 2.01	9.73 ± 6.89	14.28 ± 5.04	8.73 ± 1.93	8.63 ± 3.61

The collected specimens of bleak from the three biotopes were examined for helminths. Helminthological investigations were performed according to Petrochenko (1956); Zashev and Margaritov (1966); Kakacheva-Avramova (1983); Bauer (Ed.) (1987); Moravec (2013). The isolated endoparasites were fixed and stored in 70% ethyl alcohol. Permanent microscope preparations were prepared by the representatives of the class Trematoda and class Cestoda according to the method of Georgiev et al. (1986) and Scholz and Hanzelova (1998), and temporary microscope preparations were prepared by the representatives of class Nematoda and class Acanthocephala (Zashev and Margaritov, 1966; Moravec, 2013).

Prevalence (P %), mean intensity (MI) and mean abundance (MA) were determined for each parasite species. The structure of the component parasite communities was determined according to the criteria proposed by Kennedy (1993); Bush et al. (1997). Based on prevalence (P%), species are divided into accidental (P%<10), component (10<P%<20), and core (P%>20). The infracommunities are analyzed based on indicators: total number of species, the mean number of endoparasites, the Brillouin's diversity index (HB) (Magurran, 1998). The calculations were performed with MS Excel (Microsoft 2010) and Statistica 10 (StatSoft Inc., 2011).

RESULTS AND DISCUSSIONS

Ecoparasitological studies were conducted on a total of 165 specimens of bleak *Alburnus alburnus* (Linnaeus, 1758) from the Danube River. Bleak is a freshwater fish that is widely distributed throughout Europe. It is also found in the waters of the Danube River. It belongs to the family Cyprinidae. The bleak is a fish with small size – it reaches a maximum length of 17-19 cm and a maximum weight of 80-100 g. The fish prefers the upper water layers, where it feeds mostly on insects (Karapetkova and Jivkov, 2006).

Helminth community structure

Parasites were found in 41 specimens (24.85%) out of all 165 examined specimens *A. alburnus* from the Danube River. In spring, 18 specimens (28.57%) of 63 specimens bleak were infected, in summer 14 specimens (21.88%) of 64 specimens bleak were infected, and 9 specimens (23.68%) of 38 specimens bleak were infected in the autumn. In all 165 studied specimens *A. alburnus*, seven parasite species were established: 4 species of the class Trematoda (*Nicolla skrjabini* (Iwanitzky, 1928), *Allocreadium isoporum* (Looss, 1894), *Posthodiplostomum cuticola* (von Nordmann, 1832), *Sphaerostomum bramae* (Müller, 1776)); 1 species of the class Cestoda (*Neogryporhynchus cheilancristrotus* (Wedl, 1855), larvae); 1 species from the class Acanthocephala (*Pomphorhynchus laevis* (Müller, 1776)); 1 species from the class Nematoda (*Contracaecum* sp., larvae) (Table 3).

Table 3. Species diversity of *Alburnus alburnus* parasites by seasons and biotopes from the Danube River

Parasite species	Spring		Summer		Autumn	
	Koshava	Kudelin	Kudelin	Novo selo	Kudelin	Novo selo
Nicolla skrjabini (Iwanitzky, 1928)	•					•
Allocreadium isoporum (Looss, 1894)	•					
Posthodiplostomum cuticola (von Nordmann, 1832)			•		•	•
Sphaerostomum bramae (Müller, 1776)			•	•	•	•
Neogryporhynchus cheilancristrotus (Wedl, 1855), larvae	•	•		•		
Pomphorhynchus laevis (Müller, 1776)		•	•			
Contracaecum sp., larvae		•			•	

Component community

In the component community of *A. alburnus* from the Danube River (Koshava, Novo selo and Kudelin biotopes), the trematodes (4 species with > 971 specimens) are represented with the largest number of specimens, followed by the nematodes (1 species with 7 specimens), the cestodes (1 species with 4 specimens) and the acanthocephalans (1 species with 2 specimens). Three species of parasites were found in bleak from Koshava biotope: *Nicolla skrjabini*, *Allocreadium isoporum* and *Neogryporhynchus cheilancristrotus*, larvae. The trematode *A. isoporum* (P % = 29.03) is a core, the trematode *N. skrjabini* (P % = 16.13) is a component, and

the cestode *N. cheilancristrotus* (P % = 3.23) is an accidental parasite species in the parasite communities of bleak from Koshava biotope. The highest mean intensity (MI) and the highest mean abundance (MA) has *A. isoporum* (MI = 4.11; MA = 1.19) (Table 4).

Table 4. Species diversity and main ecological terms of parasites and parasite communities of *Alburnus alburnus* from the Danube River, Koshava biotope

Parasite species	Koshava N = 31					
	n	p	MI	MA	P %	Range
Nicolla skrjabini	5	12	2.40	0.39	16.13	1–4
Allocreadium isoporum	9	37	4.11	1.19	29.03	1–12
Neogryporhynchus cheilancristrotus, larvae	1	1	1.00	0.03	3.23	1

N – number of studied fish hosts, n – number of infected fish hosts, p – number of fish parasites, MI – mean intensity, MA – mean abundance, P % – prevalence.

Four species of parasites were found in bleak from Novo selo biotope: *Nicolla skrjabini*, *Sphaerostomum bramae*, *Posthodiplostomum cuticola* and *Neogryporhynchus cheilancristrotus*, larvae. The trematode *S. bramae* (P % = 10.00) is a component parasite species, while *N. skrjabini* (P % = 2.00), *P. cuticola* (P % = 2.00) and *N. cheilancristrotus* (P % = 2.00) are accidental parasite species in the parasite communities of bleak from Novo selo biotope. *P. cuticola* has the highest mean intensity (MI = 100.00) and the highest mean abundance (MA = 2.00) (Table 5).

Table 5. Species diversity and main ecological terms of parasites and parasite communities of *Alburnus alburnus* from the Danube River, Novo selo biotope

Parasite species	Novo selo N = 50					
	n	p	MI	MA	P %	Range
Nicolla skrjabini	1	1	1.00	0.02	2.00	1
Sphaerostomum bramae	5	11	2.20	0.22	10.00	1–5
Posthodiplostomum cuticola	1	> 100	100.00	2.00	2.00	> 100
Neogryporhynchus cheilancristrotus, larvae	1	1	1.00	0.02	2.00	1

N – number of studied fish hosts, n – number of infected fish hosts, p – number of fish parasites, MI – mean intensity, MA – mean abundance, P % – prevalence.

Five species of parasites were found in bleak from Kudelin biotope: *Sphaerostomum bramae*, *Posthodiplostomum cuticola*, *Neogryporhynchus cheilancristrotus*, larvae, *Pomphorhynchus laevis* and *Contracaecum* sp., larvae. All identified parasites, *P. cuticola* (P % = 9.52), *Contracaecum* sp., larvae (P % = 4.76), *S. bramae* (P % = 3.57), *P. laevis* (P % = 2.38) and *N. cheilancristrotus*, larvae (P % = 1.19) are accidental parasite species in the parasite communities of bleak from Kudelin biotope. The highest mean intensity (MI) and the highest mean abundance (MA) has *P. cuticola* (MI = 100.00; MA = 9.52) (Table 6).

Table 6. Species diversity and main ecological terms of parasites and parasite communities of *Alburnus alburnus* from the Danube River, Kudelin biotope

Parasite species	Kudelin N = 84					
	n	p	MI	MA	P %	Range
Sphaerostomum bramae	3	10	3.33	0.12	3.57	1–6
Posthodiplostomum cuticola	8	> 800	100.00	9.52	9.52	>100
Neogryporhynchus cheilancristrotus, larvae	1	2	2.00	0.02	1.19	2
Pomphorhynchus laevis	2	2	1.00	0.02	2.38	1
Contracaecum sp., larvae	4	7	1.75	0.08	4.76	1–4

N – number of studied fish hosts, n – number of infected fish hosts, p – number of fish parasites, MI – mean intensity, MA – mean abundance, P % – prevalence.

Only from Kudelin biotope were taken samples during the three seasons (spring, summer and autumn). During all studied period, parasite species on *A. alburnus* from the Danube River were detected in this biotope. *N. cheilancristrotus* was found in one specimen of *A. alburnus* only in the spring. *P. laevis* was found in one specimen *A. alburnus* in spring and summer. *Contracaecum* sp. occurred in spring and autumn, in 1 and 3 specimens *A. alburnus*, respectively. *S. bramae* and *P. cuticola* were found in summer and autumn. In the component community of bleak from Kudelin biotope, *P. cuticola* has the highest number of specimens (> 700) in summer, the highest mean intensity (MI) in summer (MI = 100.00) and autumn (MI = 100.00), and the highest mean abundance (MA) again in summer (MA = 23.33). In Kudelin biotope, in all three seasons, an equal number of parasite species (3 species) were detected. The highest number of parasite specimens (> 707) were identified during the summer. The dominant parasite species in the summer and autumn is the trematode *P. cuticola* (Table 7).

Table 7. Seasonal differences in invasion indices of *Alburnus alburnus* from the Danube River, Kudelin biotope

Season	Spring (N = 32)				Summer (N = 30)				Autumn (N = 22)			
Parasite species	n/p	MI	MA	P % (Range)	n/p	MI	MA	P % (Range)	n/p	MI	MA	P % (Range)
S. bramae	–	–	–	–	1/6	6.00	0.20	3.33 (6)	2/4	2.00	0.18	9.09 (1–3)
P. cuticola	–	–	–	–	7/>700	100.00	23.33	23.33 (>100)	1/>100	100.00	4.55	4.55 (>100)
***N. cheilancristrotus* larvae**	1/2	2.00	0.06	3.13 (2)	–	–	–	–	–	–	–	–
P. laevis	1/1	1.00	0.03	3.13 (1)	1/1	1.00	0.03	3.33 (1)	–	–	–	–
***Contracaecum* sp., larvae**	1/1	1.00	0.03	3.13 (1)	–	–	–	–	3/6	2.00	0.27	13.64 (1–4)

N – number of studied fish hosts, n – number of infected fish hosts, p – number of fish parasites, MI – mean intensity, MA – mean abundance, P % – prevalence.

Infracommunity

Of all 165 examined specimens *A. alburnus* from the Danube River (Koshava, Novo selo and Kudelin biotopes) – 124 specimens (75.15%) were not infected, and 41 specimens (24.85%) were infected with only one parasite species (Table 8).

Table 8. Infracommunity of *Alburnus alburnus* from the Danube River

Number of specimens *Alburnus alburnus*	Number of parasite species	
	0	**1**
	124	41
Total number of species (Mean number of species ± SD)	7 (0.25 ± 0.43)	
Total number of specimens (Mean number of specimens ± SD)	984 (82.0 ± 227.86)	
Brillouin's diversity index (HB), Mean ± SD	0.60 ± 0.19	

In the infracommunities of *A. alburnus* from the Danube River, Koshava biotope, the number of parasite specimens ranged from 1 to 12. Whereas in infracommunities of *A. alburnus* from the Danube River, Novo Selo and Kudelin biotopes, the number of parasite specimens ranged from 1 to > 100. A total of 984 helminth specimens were studied. The Brillouin's diversity index for the studied sample of the three biotopes is 0.60, ranging from 0.82 to 0.46 for the spring and summer seasons, with an average value of 0.53 for the autumn season (Table 8).

CONCLUSIONS

As a result of the conducted study of 165 specimens *A. alburnus* caught from the Danube River, 7 parasite species were identified: *N. skrjabini*, *A. isoporum*, *P. cuticola*, *S. bramae*, *N. cheilancristrotus*, larvae, *P. laevis* and *Contracaecum* sp., larvae. *A. alburnus* is a new host for *S. bramae* and *N. cheilancristrotus*, from the studied river ecosystem. With the highest parasite species diversity (5 species) are the studied specimens *A. alburnus* from Kudelin biotope. The highest mean intensity has *P. cuticola* (MI = 100.00), established as a parasite of *A. alburnus* from Novo Selo and Kudelin biotopes. With the highest mean abundance is distinguished the trematode *P. cuticola* (MA = 9.52), a parasite of *A. alburnus* from Kudelin biotope. The trematode *A. isoporum* (P % = 29.03) is a core parasite species in the component community of *A. alburnus* from the Danube River, Koshava biotope. In Kudelin biotope, samples were taken in all three seasons. During each season, three parasite species were detected. The highest mean intensity has *P. cuticola* (MI = 100.00) established in *A. alburnus* in the summer and autumn seasons, and the highest mean abundance has *P. cuticola* (MA = 23.33) established in the summer season.

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