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# HELMINTHS AND HELMINTH COMMUNITIES OF *SILURUS GLANIS* (LINNAEUS, 1758) FROM THE TUNDJA RIVER, BULGARIA

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***Abstract***

*Ecologoparasitological research was done based on the helminths and helminth communities of wels catfish (Silurus glanis Linnaeus, 1758) from the freshwater ecosystem of the Tundja, Aegean Water Basin. As a result of the examined seven specimens of wels catfish, three taxa of helminths were found. The dominant structure of the helminth communities was determined. Eustrongylides excisus Jägersiöld, 1909, larvae is a core species for helminth communities of S. glanis (P% = 42.86). S. glanis from the river ecosystem is a new host record for E. excisus. The basic ecological indices of the parasitic populations and communities were determined. The bioindication role of the established parasitic complexes was studied. An assessment of the ecological status of the studied biocenoses was carried out.*

***Key words**: bioindication, helminth communities, river Tundja, Silurus glanis.*

## INTRODUCTION

Tundzha River is the third largest river in Bulgaria (390 km; after the Danube and Iskar rivers) and the Maritsa River's largest tributary, Aegean Water Basin. The river springs from the Balkan Mountains, from 2083 m above sea level. The Tundja River flows into the Maritsa River near Edirne, Turkey, at 32 m above sea level. The waters of the river are used for agriculture, domestic and industrial water supply, electricity, etc. The aquatic ecosystem and its adjacent territories are characterised by great biological diversity, related to the declaration of a number of protected areas and zones. Parasites and parasitic communities reflect the state of the habitat. Most helminths have complex developmental cycles. Therefore, infection indices largely reflect the integrity of food chains, biodiversity, etc. Parasites and parasitic communities have been studied by a number of authors (Margaritov, 1959; Margaritov, 1966; Kakacheva et al., 1978; Soylu, 2005; Goga & Codreanu-Bălcescu, 2013; Kirin & Kuznamova, 2014; Őktener, 2014; Roohi et al., 2014; Abdybekova et al., 2020, etc.). The catfish (*Silurus glanis* Linnaeus, 1758) from the Tundzha River has not been the subject of ecological parasitological research. The study presents data on the endohelminths and helminth communities of catfish (*S. glanis*) from the Tundzha River and discusses the condition of the communities from the studied part of the river.

## MATERIALS AND METHODS

In 2019, seven specimens of wels catfish (*Silurus glanis* Linnaeus, 1758) from the Tundja River, Bulgaria, were examined for helminths. According to permission from the Ministry of Agriculture, Food and Forestry of the Republic of Bulgaria, the fish were caught by angling. The scientific name of the fish was present, according to Froese & Pauly (Eds.) (2020). The fish were caught in the section of the river with coordinates: 42°33′12″N, and 25°38′21″ E; 309 m altitude, located between the Balkan Mountain and the Mountain range Sredna Gora, about 20.5 km far away from the town of Kazanlak, Central Southern Bulgaria. The helminthological studies were carried out according to Zashev & Margaritov (1966); Bauer (Ed.) (1987); Moravec (2013). Helminth specimens were fixed in 70% of ethyl alcohol. Species diversity was determined on temporary slides carried out by the method of Moravec (2013) and Petrochenko (1956). Two levels analysed helminth community structure: 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). In the component community, the found species were divided into 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 Statistica 10 (StatSoft Inc., 2011) and MS Exel (Microsoft 2010).

## RESULTS AND DISCUSSIONS

### Characteristics of the studied fish species

*Silurus glanis* Linnaeus, 1758 (Siluridae) is a brackish, benthopelagic, non-migratory, heat-loving freshwater fish species. The fish species is naturally distributed in Europe and Asia, including in the Aegean Sea and the Maritsa River Basin. The species inhabits the middle and lower parts of the rivers, reservoirs, etc. It prefers slow-flowing and standing waters with shelters and subterranean. *S. glanis* is a typical predator. Only in the first year, he is eating zooplankton organisms and macrozoobenthos. Foods for adult fish are other species of fish, frogs, waterfowl birds and mammals. Due to plants' swift pace, valued qualities of this fish as food, *S. glanis* is subject to artificial breeding and a species for industrial and sport fishing. The wels catfish is protected by the Berne Convention (Annex 3 - Protected Fauna). IUCN Red List Status of the species is Least Concern (=LC, IUCN) (Froese & Pauly, 2020, Eds.). The species is not protected according to the Republic of Bulgaria's national legislation. Of the studied seven specimens of catfish from the Tindja River, two specimens are free of helminths.

### Helminths and helminth community structure

As a result of the ecological-parasitological examinations of 7 specimens of catfish from the Tundja River, infestation with three types of endohelminths was established: *Acanthocephalus lucii* (Müller, 1776) Lühe, 1911; *Eustrongylides excisus* Jägerskiöld, 1909, larvae and *Contracaecum* sp., larvae, belonging to two classes, three orders, three families and three genera (Table 1).

Table 1. Biodiversity and ecological indices of helminths and helminth communities of *Silurus glanis* Linnaeus, 1758 from the Tundja River

| *Silurus glanis* ($N^1 = 7$) Helminth species | $n^2$ | $p^3$ | $P\%^4$ | $MI^5$ |
|---|---|---|---|---|
| **Class** Acanthocephala (Rudolphi, 1808) Skrjabin et Schulz, 1931 **Order** Echinorhynchidae Southwell et Macfie, 1925 **Family** Echinorhynchidae (Cobbold, 1879) Hamann, 1892 **Genus** *Acanthocephalus* Koelreuther, 1771 | | | | |
| *Acanthocephalus lucii* (Müller, 1776) Lühe, 1911 | 1 | 1 | 14.29 | 1.0 |
| **Class** Nematoda Rudolphi, 1808 **Order** Dioctophymida (Skrjabin) Schulz et Gvozdev, 1970 **Family** Dioctophymatidae Castellani et Chalmers, 1910 **Genus** *Eustrongylides* Jägerskiöld, *1909* | | | | |
| *Eustrongylide excises* Jägerskiöld, 1909, larvae | 3 | 10 | 42.86 | 3.34 |
| **Order** Ascaridida Skrjabin et Schulz, 1940 **Family** Anisakidae Skrjabin et Karokhin, 1945 **Genus** *Contracaecum* Railliet et Henry, 1912, larvae | | | | |
| *Contracaecum* sp., larvae | 1 | 2 | 14.29 | 2.0 |

Legend: $^1N$ = total number of examined fish specimens.
$^2n$ = total number of infected fish specimens.
$^3p$ = total number of helminth specimens.
$^4P\%$ = prevalence.
$^5MI$ = mean intensity.

***Ac. lucii*** parasitises as an adult stage in various species of freshwater fish: Cyprinidae, Percidae, Siluridae, Salmonidae, Esocidae, Gadidae, Cobitidae, Anguillidae. Intermediate hosts of this acanthocephalan species are crustaceans *Asellus aquaticus* (Linnaeus, 1758) (Petrochenko, 1956; Bauer, 1987) (Table 4). Bulgaria, *Ac. lucii* was found as the helminth species of *S. glanis* from the Danube River and *Squalius cephalus* (Linnaeus, 1758) from Iskar and Tundzha rivers (Margaritov, 1959); of *Perca fluviatilis* Linnaeus, 1758 (Margaritov, 1966); of *Ballerus sapa* (Pallas, 1814), *Sq. cephalus*, *Rutilus rutilus* (Linnaeus, 1758), *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 *Abramis brama* (Linnaeus, 1758) (Chunchukova et al., 2017); of *Alburnus alburnus* (Linnaeus, 1758) (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; of *R. rutilus* from the Luda Yana River (Kirin et al., 2019); of *Sq. cephalus* from the Ogosta River (Chunchukova et al., 2020), etc. *E. excisus*, larvae are developed with the participation of the first intermediate host oligochaetes (blackworm *Lumbricus variegatus* Linnaeus, 1758, sludge worm *Tubifex tubifex* (Muller, 1774), *Limnodrilus* sp.) and the second fish species, amphibians (Marsh frog, *Pelophylax ridibundus* (Pallas, 1771) (= *Rana ridibunda* Pallas, 1771) and reptiles (Dice snake, *Natrix tesselata* (Laurenti, 1768). The adult nematodes parasitic in the glandular stomach of cormorants [Great Black Cormorant *Phalacrocorax carbo* (Linnaeus, 1758) and Pygmy Cormorant *Microcarbo pygmeus* (Pallas, 1773) (= *Ph. pygmaeus* Pallas, 1773)] (Moravec, 2013) (Table 4).

In Bulgaria, the species is found of *Sander lucioperca* (Linnaeus, 1758) (= *Lucioperca lucioperca* Linnaeus, 1758) (as paratenic host) and of *Gobius* sp. (as intermediate host), of *Aspius aspius* (Linnaeus, 1758) from the Danube River (Kakacheva et al., 1978; Margaritov, 1959); of *P. fluviatilis* from the Zhrebchevo Reservoir (Nedeva & Grupcheva, 1996) and the Srebarna Lake (Shukerova & Kirin, 2007; Shukerova et al., 2010); of *S. glanis*; *L. lota*, *Neogobius melanostomus* (Pallas, 1814) (= *Neogobius cephalarges* Pallas, 1814), *N. kessleri* (Gunther, 1861), *P. fluviatilis* from the Danube River (Atanasov, 2012); of *P. fluviatilis* from the Arda River (Kirin et al., 2013a) from the River Danube and Srebarna lake (Kirin et al., 2013b); of *Rutilus frisii* (Nordmann, 1840) and *Alburnus chalcoides* (Güldenstüdt, 1772) from the Veleka River (Kirin, 2014); of *S. glanis* from the Ivaylovgrad Reservoir (Kirin & Kuzmanova, 2014), etc. *Contacaecum* sp. is reported of *Chondrostoma nasus* (Linnaeus, 1758) and *A. alburnus* from the Danube River (Zaharieva & Zaharieva, 2020a, b; Zaharieva & Kirin, 2020a, b, respectively; Chunchukova et al., 2019), etc. In previous studies, specimens of *Contracaecum* of *S. glanis* were referred to as the species *Contracaecum bidentatum* (Linstow, 1899) (Kakacheva-Avramova, 1977; Kakacheva-Avramova et al., 1978; Kirin & Kuzmanova, 2014) (Table 3).

**Component community**

The presented helminth taxa were found in 5 of the studied seven catfish specimens (71.43%). Prevalence (P%), mean intensity (MI) and rank were determined for each taxa. *E. excisus* (P% = 42.86) is a core species of the endohelminth communities of *S. glanis* from the Tundja River. The other two species are component (both with P% = 14.29). *E. excisus* is also with the highest mean intensity (MI = 33.34), followed by *Contracaecum* sp. (MI = 2.0). Only one specimen of *Ac. lucii* was fixed in the infected specimen of catfish. *Ac. lucii* is autogenic species. *E. excisus* and *Contracaecum* sp. are allogenic species. The established taxa are generalists for the helminth communities of *S. glanis* from the Tundzha River, Bulgaria (Table 1).

**Infracommunity**

**A** total of two examined specimens of *S. glanis* are free of helminths (28.57%). In this study, no mixed invasion was detected. The maximum number of parasites found in a single specimen by the host is four (*E. excisus*). The average number of all endohelminth specimens is low (0.98±0.62), as well as the value of Brillouin's diversity index (HB = 0.45±0.42) (Table 2).

Table 2. Infracommunity data

| Number Of helminth species | | |
|---|---|---|
| Number of infected fish | 2 | 5 |
| Number of helminth species | 0 | 1 |
| Number of helminth specimens | | |
| Total number | 13 | |
| Mean ± SD | 0.98±0.62 | |
| Range | 1-4 | |
| Mean HB ± SD | 0.45±0.42 | |

A total of 13 endohelminth taxa of catfish have been reported in Bulgaria. According to the study, only three taxa were identified (23.08%). Two of the identified species (*A. lucii* and *E. excisus*) have been reported in previous studies as catfish helminths in the country. Detected specimens of the genus *Contraceaceum* have not been identified (Tables 1, 3).

Table 3. Endohelminths of *Silurus glanis* from freshwater ecosystems of Bulgaria

| Species diversity | Authors | Freshwater ecosystems (Biotopes) |
|---|---|---|
| **Trematoda** | | |
| *Orientocreadium siluri* (Bychowski & Dubinina, 1954) Yamaguti, 1964 | Kakacheva-Avramova, 1977 | river Danube (town (t.) Silistra) |
| | Kakacheva, Margaritov, Grupcheva, 1978 | river Danube (t. Silistra) |
| | Atanasov, 2012 | river Danube (village (v.) Archar, v. Botevo, t. Svishov) |
| *Nicolla skrjabini* (=*Crowcrocoecum skrjabini*) | Margaritov, 1966 | river Danube |
| **Cestoda** | | |
| *Triaenophorus nodulosus* (Kuperman, 1968) | Atanasov, 2012 | river Danube (v. Archar) |
| *Silurotaenia siluri* (Batch, 1786) Nybelin, 1942 | Margaritov, 1964 | river Danube (t. Svishov) |
| | Kakacheva-Avramova, 1977 | river Danube (t. Silistra) |
| | Kakacheva, Margaritov, Grupcheva, 1978 | river Danube (t. Svishov, t. Ruse, t. Silistra) |
| | Atanasov, 2012 | river Danube (v. Archar) |
| *Glanitaenia osculata* (=*Proteocephalus osculatus*) | Kabaivanski, 1935 | river Danube |
| | Margaritov, 1959 | river Danube (t. Ruse, t. Svishov) |
| | Margaritov, 1960 | lake Shabla |
| | Kakacheva, Margaritov, Grupcheva, 1978 | river Danube (t. Ruse, t. Svishov) |
| | Kirin, Kuzmanova, 2014 | Reservoir Ivaylovgrad |
| **Acanthocephala** | | |
| *Pomphorhynchus laevis* (Müller, 1776) | | |
| | Margaritov, 1966 | river Danube |
| | Kakacheva-Avramova, 1977 | river Danube (t. Silistra, t. Svishov, t. Lom) |
| | Kakacheva, Margaritov, Grupcheva, 1978 | river Danube (t. Svishov, t. Ruse, t. Vidin, t. Lom t. Tutrakan) |
| | Atanasov, 2012 | river Danube (v. Archar, v. Dobri dol, t. Svishov, v. Botevo, v. Gomotarci, v. Vardim, v. Novo selo, v. Simeonovo, t. Kozloduj) |
| *Acanthocephalus lucii* (Müller, 1776) Lühe, 1911 | Margaritov, 1959 | river Danube (t. Svishov) |
| | Kakacheva, Margaritov, Grupcheva, 1978 | river Danube (t. Vidin, town Silistra, t. Svishov) |
| **Nematoda** | | |
| *Eustrongylides excisus* (Jägerskiöld, 1909) | Atanasov, 2012 | river Danube (v. Archar, v. Dobri dol, v Gomotarci) |
| | Kirin, Kuzmanova, 2014 | Reservoir Ivaylovgrad |
| *Contracaecum bidentatum* (Linstow, 1899) | Kakacheva-Avramova, 1977 | river Danube (t. Svishov) |
| | Kakacheva, Margaritov, Grupcheva, 1978 | river Danube (t. Ruse, t Vidin, t. Silistra, t. Svishov) |
| | Kirin, Kuzmanova, 2014 | reservoir Ivaylovgrad |
| *Rhabdochona* sp. juv | Kakacheva-Avramova, 1965 | rivers Maritsa, Asenitsa |
| *Rhabdochona* sp. | Margaritov, 1966 | river Danube |
| *Rhabdochona* sp., larvae | Kakacheva, Margaritov, Grupcheva, 1978 | river Danube (t. Vidin t. Lom) |
| *Rhabdochona denudate* | Kakacheva-Avramova, 1965 | rivers Maritsa (t. Pazardzhik, v. Ognyanovo, v. Kovachevo, t. Septemvri, v. Sadovo, t. Svilengrad), Topolnitsa (v. Srebrino, t. Pazardzhik), Chepinska (v. Kovachevo), Asenitsa (t. Asenovgrad, v. Katunitsa), Sushitsa (v. Bogdantsi), Syuyutlijka (v. Kiril-Antonievo, v. Starozagorski bani), Bedechka (t. Stara Zagora), Harmanlijska (area "Popov bent" and village Bregovo), reservoir "40-te izvora" |

Researches on catfish parasites are mainly related to the Danube river basin. In most of them, the parasitic communities are not analysed. Registered helminth taxa have complex development cycles involving more than one host. Catfish is the definitive host only for *Acanthocephalus lucii*. The helminth species inhabit the intestine of the host. *E. excisus* and *Contracaecum* sp. are third stage larves with localisation body cavity/ abdominal cavity and mesentery, respectively. Specified invertebrate intermediate hosts are approved bioindicators for the saprobity in the habitats. *Asellus aquaticus* (Linnaeus, 1758) and *Lumbricus variegatus* Linnaeus, 1758 indicated α-mesosaprobity. *Limnodrillus* sp. indicated p-α-mesosaprobity and *Tubifex tubifex* - p-saprobity. The apparent dominance of *E. excisus* and the indicated bioindicator role of the invertebrate intermediate hosts point to p-α-mesosaprobity in the studied section of the river. The values of the prevalence and mean intensity of *E. excisus* (core species in the study) showed very different values as in the studies from Bulgaria and other countries (Table 4). The presented studies do not establish regularities in the values of prevalence and mean intensity related to the type of freshwater ecosystems – lotics or lentics. It is assumed that the obtained values are closely related to the intensity of the first intermediate host populations and those of small fish, frogs, reptiles (second intermediate hosts), which are food for the large predator, the catfish.

Table 4. Prevalence and mean intensity of *Eustrongylides excisus* as a helminth species of *Salmo trutta* from freshwater ecosystems in Bulgaria and other countries

| Author | Localisation | P% | MI (range) |
|---|---|---|---|
| **From Bulgaria** | | | |
| Atanasov, 2012 | river Danube (villages Archar, Dobri dol, Gomotartsi) | 6.38 | 0.15 (3-11) |
| Kirin, Kuzmanova, 2014 | reservoir Ivaylovgrad | 27.82 | 2.0±0.29 (1-2) |
| Kirin, Chunchukova, 2021 – this study | river Tundzha | 42.86 | 3.34± (2-4) |
| **From other countries** | | | |
| Soylu, 2005 | lake Durusu (Terkos), Turkey | 41.8 | 1.37 (2-28) |
| Goga & Codreanu-Bălcescu, 2013 | lake Viktoria, Romania | 10 | - |
| Roohi et al., 2014 | Anzali International wetland, Iran | 69.77 | 5.37±4.65 (1-21) |
| Abdybekova et al., 2020 | Kazakhstan | 33 | 54.33 (72-254) |

## CONCLUSIONS

The study presents the first data on the helminths and helminth communities of the Tundzja River catfish. Of the three found helminth species, *A. lucii* is a core species, and the other two are component species for the helminth communities of *S. glanis*. Only *A. lucii* is an autogenic species in communities. The values of the prevalence and mean intensity are closely related to the intensity of the intermediate host populations and food chains' integrity.

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