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DIVERSITY AND DISTRIBUTION OF PARASITES IN SOME FRESHWATER FISH FROM ROMANIAN SECTOR OF PRUT RIVER

Angelica DOCAN[1], Maria Desimira STROE[2], Iulia GRECU[1], Lorena DEDIU[1], Mirela CREȚU[1], Magda TENCIU[2]

[1]"Dunărea de Jos" University of Galați, 47 Domească Street, Romania
[2]Research - Development Institute for Aquatic Ecology, Fisheries and Aquaculture, 54 Portului Street, Galați, Romania

Corresponding author email: sdesimira.icdeapa@gmail.com

Abstract

Fish parasites represent a major part of aquatic biodiversity. The aim of present investigation was to assess the diversity and distribution of parasites from some fish species from the Prut River. Fish were sampled from three station on Prut River (Rădăuți, Drânceni, Oancea) during the year 2020. The different types of fishes from 5 families Cyprinidae, Esocidae, Percidae, Siluridae, Cobitidae have been examined for analysis of the distribution of parasites from these fish, in order to complete the existing data on metazoan parasites of freshwater fishes in the Romanian sector of Prut River. Parasitological investigations were performed on fresh samples by classic methods and the obtained results were expressed in grades of prevalence and intensity. In the analysed fish, were identified 12 parasitic species belonging to 6 systematic groups: Protozoa, Monogenea, Trematoda, Cestoda, Nematoda, Anelida. The prevalence of the parasitosis varied among examined fish species. The ciliated protozoa and monogenic worms were the most commonly identified parasites, but the intensity of parasitism was low. The effects of parasites on fish hosts in the natural environment may be difficult to isolate and quantify.

Key words*: ecto parasites, endoparasites, freshwater fish, Prut River.*

INTRODUCTION

The Prut River basin is shared by Ukraine, Romania, and Moldova. Its source is in the Ukrainian Carpathians. Later, the Prut forms the border between Romania and Moldova. The diversity of aquatic ecosystems and the richness of freshwater fish species are features of the Prut River. Like other living organisms, fish have parasites either external or internal which cause a host of pathological debilities. Freshwater fish species may be definitive and intermediate hosts of parasites with larvae and mature stages infecting a variety of vertebrates, including humans (Djikanovic et al., 2012). Fishes are susceptible to all the phyla of parasites including annelids and arthropods and can affect the fish biology which tends into diseases, mortality, disordered growth pattern, and ultimately makes the loss to fish values (Lafferty, 2008).

The authors of several studies have revealed large parasitic fauna in freshwater fishes (Cojocaru, 2010, Ejere et al., 2014) ranging from ectoparasites (Kostoingue et al., 2001; Oniye et al., 2004) to endoparasites (Kumar, et al., 2012; Cakic et al., 2008) which can affect fish health, growth and survival. Studies on parasitic communities of wild fish populations increase understanding of the parasite-host-environment interactions, because parasites may be indicators of environmental conditions and of their hosts (Hoshino et al., 2014). The parasites of fish can reflect the life habits of the fish, including their interactions with the benthic, planktonic and fish communities (Landsberg et al., 1998). Parasite species richness and abundance can differ geographically for the same host species and it can be influenced by the ecosystem characteristics and its trophic diversity (Vales et al., 2010).

Our team analyzed the fish community like *Silurus glanis, Stizostedion lucioperca, Cyprinus carpio, Abramis brama,* from the Prut River (Frumușița station, Cotul Chiului area) and the parasites identified do not affect the health status of their hosts (Docan et al., 2019).

This type of information is poorly studied in fish species from Prut River, in particular, and therefore, become the general targets of this study by focusing on the fish types, abundance, and prevalence of parasitic infestation. In this paper we present an analysis of the distribution of parasites from some fish species, in order to complete the existing data on metazoan parasites of freshwater fishes in the Romanian sector of Prut River.

MATERIALS AND METHODS

The fishes were collected randomly between the period of summer (April 1st to July 30th) 2020, from three stations on Prut River (Rădăuți, Drânceni, Oancea). The different types of fishes belong from 5 families: *Cyprinidae, Esocidae, Percidae, Siluridae, Cobitidae*. The scientific fishing activity from every area was carried out over a length of 2 km, with the fishing net wall. The fish were weighed (g) and their total length was measured (cm). Fish were transported in Research Centre MoRAS-UDJ Galati laboratory (http://moras.ugal.ro) and in ICDEAPA Galați laboratory (https://asas-icdeapa.ro/) where parasitological analyses were carried out.

The sampled fish were examined for both ectoparasites and endoparasites using standard parasitological procedures. The external surface of the fish was examined thoroughly using a hand lens for macroscopic ectoparasitic species, including crustaceans and hirudineans. Smear of scrapings from the skin, fins and gills were also examined for ectoparasites. Each fish was sectioned dorso-ventrally and the alimentary canal, liver, kidney, swim bladder and spleen were examined for endoparasites. Parasites were identified to family, genus or species level when possible.

The taxonomic classification and identification of the observed parasites were done on the basis of Munteanu, 2005, Bauer, 1984, 1985, 1987. For isolation, selection and identification of the parasite fauna of freshwater fish from Prut River, we used a Zeiss microscope. The extent of parasite infection was expressed in terms of an individual host as the intensity of infection (the number of individual parasites of a particular species harboured) and in terms of host populations as the prevalence (the proportion of hosts harbouring at least one individual parasite of a particular species) (Bush et al., 1997).

RESULTS AND DISCUSSIONS

The data on freshwater fish parasites are important for the evaluation of health conditions and the general influence of the level of parasitism on the community structure (Djikanovic et al., 2012).

The results of the parasitological examination are presented synthetically in tables 1, 2 and 3: the parasite and their habitat, the prevalence and mean intensity of infestation, in the three analyzed stations.

In the upper sector of the Prut River, Rădăuți station were captured 9 species grouped into 3 families: *Cyprinidae, Esocidae, Percidae*, respectively: *Cyprinus carpio* Linnaeus, 1758*, Carassius auratus gibelio* Bloch, 1782*, Hypophthalmichthys molitrix* Valenciennes, 1844*, Abramis brama danubii* Linnaeus, 1758*, Rutilus rutilus carpathorossicus* Linnaeus, 1758*, Scardinius erythrophtalmus* Linnaeus, 1758*, Sander lucioperca* Linnaeus, 1758*, Acerina cernua* Linnaeus, 1758*, Esox lucius* Linnaeus, 1758.

No infectious and fungal pathogens were identified. In this preliminary study the parasites will be declared by ectoparasite or endoparasite due to the general targets of this study to concentrate on the prevalence of parasitic infection.

Of the total fish species studied, 61% have weak polyparasitosis, of which: 47% ciliate products (*Trichodina domerguei, Apiosoma piscicola*), 43% monogenic worm products (*Dactylogirus vastator and Diplozoon paradoxus*), 5% cestode worm products (*Cysticercercus* sp.) and 5% annelid worm products (*Piscicola geometra*).

The results presented in Table 1 show that ectoparasites predominate in the analyzed fish. Ectoparasites found on body surface and gill, are *Trichodina domerguei, Apiosoma* sp., *Dactylogyrus vastator* and *Diplozoon paradoxus*. In our study, only one specimen of *Cysticercus sp.*was found in a liver, surrounded by a thin connective tissue capsule. This encapsulation of nematode larvae has been

observed in several cyprinids (Moravec, 1994). Cestode and anellida parasites occurred rarely in our samples. Adult worm of *Piscicola geometra* represented the only anellid species found in *Esox lucius.*

Table 1. Prevalence and intensity of metazoan parasites of fish from the Prut River, Rădăuți station

Systematic group	Species of parasite	Fish host	Parasite habitat	N/n	P%	MI
Protozoa/ Ciliata	*Trichodina domerguei*	*Carasus auratus gibelio*	gills	10/4	40	8.25
		Sander lucioperca	gills	6/1	16.66	13
	Apiosoma piscicola	*Acerina cernua*	gills	4/1	25	7
		Hypophthalmichthys molitrix	gills	6/2	33.33	5.5
Monogenea	*Dactylogyrus extensus*	*Carasus auratus gibelio*	gills	10/3	30	4,25
		Abramis brama	gills	8/3	37.5	4.66
		Cyprinus carpio	gills	10/4	40	5.75
	Diplozoon paradoxum	*Abramis brama*	gills	8/2	25	4.5
		Rhutilus rhutilus carpathorossicus	gills	5/1	20	6
		Schardinus erhythrophthalmus	gills	5/1	20	4
Cestoda	*Cysticercus* sp.	*Abramis brama*	liver	8/1	12.5	3
		Sander lucioperca	liver	6/1	16.66	2
Anellida	*Piscicola geometra*	*Esox lucius*	skin	5/1	20	3

N = total number of examined fish specimens
n = total number of infected fish specimens.
P% = prevalence.
MI = mean intensity.

In the middle sector of the Prut River (Rădăuți station) were caught 7 fish species belonging to the families *Cyprinidae* and *Percidae*: *Carassius auratus gibelio, Cyprinus carpio, Abramis brama danubii, Rutilus rutilus carpatorossicus, Scardinius erythrophtamus, Barbus barbus, Sander lucioperca*, which were parasitologically examined.

Of the examined species (Table 2), 57% had weak polyparasitosis, of which: 45% products of ciliated protozoa (*Trichodina* sp.), 35% products of monogenic worms (*Dactylogirus vastator and Diplozoon paradoxus*), 6% products of trematodes (*Neascus cuticola*), 6% products of molluscs (*Glochidia* sp.), 8% produced by cestodes (*Ligula intestinalis*). Prevalence of ectoparasites and endoparasites form freshwater fish species had different values. Regarding the number of parasites belonging to a species, identified in a certain host, there were differences between the studied fish species.

In the C*iprinids* species, the monogenic worm *Dactylogyrus extensus* was identified on the branchial scrapes, the parasitic intensity being reduced to 5-10 specimens/fish. Chubb (1977) had earlier identified temperature as the most important single factor controlling the seasonal prevalence of dactylogyrids. Monogenean trematodes, as flatworms, commonly invade the gills, skin, and fins of freshwater fish from most families of *Teleostei* (Whittington et al., 2000). Monogeneans worms have direct life cycles and they have specificity for the host. *Dactylogyrus extensus*, in the massive invasion, was found to be fatal to both young and adult fish (Munteanu et al., 2003).

Grossly examination of *Rhutilus rhutilus* revealed the presence of some blackspots in the skin on only one individual fish. The low level of *Posthodiplostomum cuticola* could be seen as a positive sign for a fishery because it shows that there is substantial habitat for aquatic gasteropods to survive, and they are an important part of the food chain in fisheries (Munteanu et al., 2003).

Plerocercoids of *Ligula intestinalis* were observed in the body cavity of *Cyprinus carpio* and *Rhutilus rhutilus,* but with low mean intensity. Ligulids have a complex life cycle involving copepods, fishes and birds. It is known to affect especially *Alburnus escherichii, Leuciscus cephalus, Tinca tinca, Cyprinus carpio and Rutilus rutilus,* which are members of the *Cyprinidae* (Înnal, 2007). Molluscae and cestode parasites occurred rarely in our samples. *Glochidia* sp. were found only in two host species, from Drânceni station,

that infected the fins and gills parts (*Abramis brama* and *Barbus barbus*). The presence of these larval stages of unionid mussels in fishes reflects the presence of adult bivalves at that sampling site. All parasite species found in our study are common and most of them occur in many fish species in the Prut River basin.

Table 2. Prevalence and intensity of metazoan parasites of fish from the Prut River, Drânceni station

Systematic group	Species of parasite	Fish host	Parasite habitat	N/n	P%	MI
Protozoa	*Trichodina domerguei*	*Carasus auratus gibelio*	gills, skin	9/3	33.33	9.66
		Sander lucioperca	gills	5/2	40	6,5
Monogenea	*Dactylogyrus extensus*	*Carasus auratus gibelio*	gills	9/4	44,44	5,25
		Abramis brama	gills	7/3	42.86	4.33
		Cyprinus carpio	gills	8/3	37.5	5.66
	Diplozoon paradoxum	*Abramis brama*	gills	7/2	28.6	6.5
		Rhutilus rhutilus carpathorossicus	gills	5/1	20	3
		Schardinus erhythrophthalmus	gills	5/1	20	2
Trematoda	*Posthodiplostomum cuticola*	*Rhutilus rhutilus carpathorossicus*	skin	5/1	20	9
Cestoda	*Ligula intestinalis*	*Cyprinus carpio*	body cavity	8/3	37.5	5.66
		Rhutilus rhutilus carpathorossicus	body cavity	5/2	40	2,5
Mollusca	*Glochidia* sp.	*Abramis brama*	gills, fins	7/1	14.28	4
		Barbus barbus	gills	4/1	25	3

The situation of parasitic agents identified in the fish analyzed on the Prut river, Oancea station, shown in the Table 3, highlights that 78% of fish species have weak polyparasitosis: 25% ciliate (*Trichodina domerguei* and *Apiosoma piscicola*), 25% monogenic worms (*Dactylogyrus extensus* and *Diplozoon paradoxus*), 25% digenic worms (metacercaria larva: *Neascus cuticola* and *Diplostomum spathaceum*), 12.5% nematodes (*Hepaticola* sp.), 12.5% cestodes (*Caryophilaeus* sp.). *Hypophthalmichthys molitrix* was the weakest parasitic species.

The proportion of ecto- and endoparasite specimens infecting fish species was similar in the three analyzed stations.

The ciliated protozoa of *Apiosoma piscicola* were observed in the gills of *Acerina cernua* and *Hypophthalmichthys molitrix* from this station. The pathogenicity of *Apiosoma species* is insufficiently known; ultrastructural observation on attached *Apiosoma* did not reveal any interference with the host cell serving as substrate or peripheral tissue response (Loom, 1973).

Two adults of cestode *Caryophyllaeus* sp. infected the intestine of a *Cyprinus carpio*. Elevated *C. fimbriceps* infection in ciprinids samples may have been caused by a high proportion of oligochaeta species, the intermediate hosts of this parasite, in fish diet.

Chunchukova, 2010 show that cestode species that refer to *A. brama* from Bulgarian part of Danube River belong to order *Caryophyllidea*.

Like the metacercaria larva of *Diplostomum* sp. was identified only in one specimen of *Rhutilus rhutilus.*

A single specimen of nematode *Hepaticola petruschewskii* was found, in the liver of a *Cyprinus carpio* and *Abramis brama*. Nematodes occur worldwide particularly the species utilizing fish as intermediate or transient hosts and can infect all of their organs.

In general, endo-parasites of fish influence fishes negatively in several ways and represent a possible threat to the sustainability of fisheries (Paperna, 1996).

The endo-parasites like tapeworms, nematodes, or acanthocephalans infect the internal organs of fish with their intermediate stages (larvae) and sometimes encysting in various host tissues or most adults mainly affect the digestive systems of their hosts (Luque et al., 2004).

Table 3. Prevalence and intensity of metazoan parasites of fish from the Prut River, Oancea station

Systematic group	Species of parasite	Fish host	Parasite habitat	N/n	P%	MI
Protozoa	*Trichodina domerguei*	*Carasus auratus gibelio*	gills, skin	8/3	37.5	10.33
		Sander lucioperca	gills	5/1	20	12
	Apiosoma piscicola	*Acerina cernua*	gills, skin	4/1	25	8
		Hypophthalmichthys molitrix	gills	3/1	33.33	7
Monogenea	*Dactylogyrus extensus*	*Carasus auratus gibelio*	gills	8/3	37.5	7.66
		Abramis brama	gills	7/2	28.57	6.5
		Cyprinus carpio	gills	8/4	50	6.25
	Diplozoon paradoxum	*Abramis brama*	gills	7/1	14.28	7
		Rhutilus rhutilus carpathorossicus	gills	4/2	50	4.5
		Schardinus erhythrophthalmus	gills	4/1	25	3
Trematoda	*Diplostomum spathaceum*	*Rhutilus rhutilus carpathorossicus*	eyes	4/1	20	2
Cestoda	*Caryophyllaeus fimbriceps*	*Cyprinus carpio*	intestine	8/4	50	5.25
		Rhutilus rhutilus carpathorossicus	intestine	4/1	25	3
Nematoda	*Hepaticola petruschewskii*	*Cyprinus carpio*	liver	8/1	12.5	1
		Abramis brama	liver	7/1	14.28	1

Although the parasitism is common in fish, parasitic diseases are triggered only in environmental conditions that facilitate the multiplication of parasites; therefore, clinical parasitosis is quite rare in freshwaters river. The presence of parasites can provide information about the state of the environment: the ciliates and nematodes should be sensitive indicators of eutrophication and thermal effluent, while digeneans and acanthocephalans should make good indicators of heavy metals and human disturbances (Lafferty 1997).

The establishment of only one intestinal parasite in two cyprinid fishes (*Barbus cyclolepis* and *Squalius orpheus*) indicated poor species diversity within the studied freshwater habitat and negative impacts on the ecosystem (Chunchukova et al., 2020)

CONCLUSIONS

The results from this study show that there is no difference in value prevalence and intensity between the three Prut River stations. But, the total parasite load of analyzed fish was relatively low compared with that observed in the other similar studies (Docan, et al., 2019). Of the eleven parasites identified in fish from the Prut River, the species *Dactylogirus vastator* and *Diplozoon paradoxum* have the highest frequency being found in 37.5% of the species caught. On the other hand, the species *Neascus cuticola* had the lowest frequency being present in 2.5% of captured species.

All captured species had gill parasites but *Abramis brama* was the species in which most parasitic taxa were identified.

In conclusion, none of the parasites collected from analyzed fish species are novel species to the Romanian sector of Prut River. Indeed, all of the observed parasite species are commonly found in native fishes from this river. The abundance of parasites varied among the fish species and station of study, with cyprinids species hosting the richest parasite community.

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