A COMPARATIVE ANALYSIS OF HELMINTH FAUNAS AND INFECTION PARAMETERS OF TEN SPECIES OF GOBIID FISHES (ACTINOPTERYGII: GOBIIDAE) FROM THE NORTH-WESTERN BLACK SEA^{*}

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Background. Fishes of the family Gobiidae may be definitive-, intermediate-, or paratenic hosts of parasites with mature stages infecting a variety of vertebrates, including humans. This group of fishes constitutes a convenient ecological model for studying the processes of colonisation by parasites. Learning these processes may contribute to a better, more complex, understanding of organismal interrelationships within respective habitats. The aim of this study was to compare the helminth infection levels of different gobiid species in the north-western Black Sea (NWBS).

Material and methods. The fishes were sampled within 1996–2003 at different seasons (excepting winter) in the NWBS. A total of 2102 specimens of 10 goby species—the black goby, *Gobius niger*, the knout (toad) goby, *Mesogobius batrachocephalus*, the mushroom goby, *Neogobius eurycephalus*, the monkey goby, *N. fluviatilis*, the round goby, *N. melanostomus*, the ratan goby, *N. ratan*, the syrman goby, *N. syrman*, the marbled goby, *Pomatoschistus marmoratus*, the tubenose goby, *Proterorhinus marmoratus*, the grass goby, *Zosterisessor ophiocephalus*—were examined for helminths. The Czekanowski–Sørensen index (I_{cs}) was used for comparing the helminth faunas. The infection indices were compared using the discriminant analysis.

Results. The fishes examined yielded a total of 24 helminth species. Four parasite species were common for all hosts surveyed: *Cryptocotyle concavum* MET, *C. lingua* MET, *Dichelyne minutus*, and *Acanthocephaloides propinquus*. *Telosentis exiguus* infected six host species, *Eustrongylides excisus*—five of them, while *Pygidiopsis genata Streptocara crassicauda* L3 were found in four gobiids. Five parasites species (*Proteocephalus gobiorum*, *Asymphylodora pontica*, *Acanthostomum imbutiformis* MET, *Raphidascaris* sp. L3, and *Streptocara crassicauda*) were common for three host fish species, while another four helminths (*Bucephalus polymorphus* MET, *Nicolla skrjabini*, *Contracaecum rudolphii* L3, and *Acanthocephalus lucii*) were found in two gobiids only. A total of ten parasite taxa were found to infect single hosts species (*Bothriocephalus gregarius* PL, *Ligula pavlovskii* PL, *Proteocephalus gobiorum* PL, *P. subtilis*, *Proteocephalus* sp. PL, *Paratimonia gobii*, *Aphalloides coelomicola*, *Aphalloides coelomicola* MET, *Contracaecum microcephalum* L3, and Anisakidae gen. sp. L3). The most stable indices of gobiid infection were determined for nematode *D. minutus*.

Conclusion. The observed differences in the species composition of helminth faunas of different gobiids were related to the zoogeographical origin of a host species, the ecological specificity of their habitats (e.g. salinity), and the biology of individual parasites.

Key words: Black Sea, gobiids, fish parasites, helminth fauna, discriminant analysis

INTRODUCTION

The ichthyofauna of the Black Sea is comparatively poor and includes about 200 species (Boltačev 2003). The gobiid fishes (Actinopterygii: Gobiidae) are a large group of approximated by 28 species inhabiting coastal habitats of the Black Sea (Miller 1986, Smirnov 1986, Rass 1993). Five Black-Sea gobiid species are important commercially (Smirnov 1986) and they have also been targeted by anglers.

In the north-western Black Sea (NWBS), gobiids can be found in different salinities ranging from fresh water (0.5% salinity off the Dniester River mouth) to full-salinity sea water (30% -35% in the Tuzly group of lagoons). These fish make up two groups according to their origin: 1) Ponto-Caspian relict species that are endemics of the Sarmatic and Pontic basins; typical representatives of this group are: *Mesogobius batrachocephalus* (Pallas, 1811);

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Neogobius spp.; and *Proterorhinus marmoratus* (Pallas, 1811); 2) Mediterranean immigrants which entered the Black Sea probably within the past 12 000 years (typical representatives are *Gobius bucchichi* Steindachner, 1870; *G. cobitis* Pallas, 1811; *G. niger* L.; *G. paganellus* L.; *Pomatoschistus marmoratus* (Risso, 1810); *P. minutus* (Pallas, 1770); and *Zosterisessor ophiocephalus* (Pallas, 1811); cf. Miller 1965).

Recently some Ponto-Caspian gobiids invaded water areas such as the Danube basin (Bíró 1971, Harka 1990, Guti 2000), the Baltic-Sea basin (Skóra and Stolarski 1993, Danilkiewicz 1998), and the North American Great Lakes (Jude et al. 1992, Charlebois et al. 1997).

Gobiids may be definitive-, intermediate, or paratenic hosts of parasites with mature stages infecting birds, mammals (including humans), and commercially exploited fishes. The Black-Sea gobiids are hosts of metacercariae of *Pygidiopsis genata* (cf. Kvach 2002a, b, 2004) that can infect humans (Youssef et al. 1987). In the 1950s, along the coast of the Gulf of Taganrog (Sea of Azov) an epizooic of nematodes, *Tetrameres fissispina* (Diesing, 1861) and *Streptocara crassicauda*, was fatal to ducklings, which had fed on infected gobiids (Kovalenko 1960).

Because of their ecological plasticity, the gobiids are a convenient model for studying the processes of colonisation by parasites (Zander and Kesting 1998). In the Black Sea the gobiid parasite fauna is more heterogeneous than the gobiid fish fauna itself (Najdenova 1974). It consists of Ponto-Caspian, Mediterranean, Boreal-Atlantic, and limnetic parasite species that are attributed to the different ranges of euryhalinity of the hosts. Therefore, the aim of this study was to compare the helminth faunas and the infection levels of different gobiid species in the NWBS.

MATERIAL AND METHODS

The fish were sampled in different seasons (except winter) during the period of 1996–2003 in the Gulf of Odessa, Tyligul, Hryhorivsky, Khadzhibey, and Dniester estuaries, the Budaki and Tuzly group of lagoons (NWBS; Fig. 1).

A total of 2102 specimens of 10 goby species-the black goby, Gobius niger, the knout (toad) goby, Mesogobius batrachocephalus, the mushroom goby, Neogobius eurycephalus (Kessler, 1874), the monkey goby, N. fluviatilis (Pallas, 1811), the round goby, N. melanostomus (Pallas, 1811), the ratan goby, N. ratan (Nordmann, 1840), the syrman goby, N. syrman (Nordmann, 1840), the marbled goby, Pomatoschistus marmoratus, the tubenose goby, Proterorhinus marmoratus, the grass goby, Zosterisessor ophiocephalus-were examined for the presence of helminths. The numbers (n)of studied hosts are presented in Table 1. Skin, muscles, internal organs, brain, eyes, gills, body cavity, and gut were carefully examined. The helminths found were fixed in 70% ethyl alcohol. To facilitate identification, the trematodes and cestodes were stained with carmine, acetic carmine, or iron carmine following the method of Georgiev et al. (1986) and permanently mounted. The metacercariae were isolated from their cysts. Temporary

Neogobius spp.; and *Proterorhinus marmoratus* (Pallas, glycerine mounts were prepared to study nematodes and 1811); 2) Mediterranean immigrants which entered the acanthocephalans.

For comparison of the helminth fauna the Czekanowski–Sørensen index (I_{cs} , %) was used:

$$I_{cs} = \frac{2c}{(a+b)} \times 100\%$$

where:

a, number of parasite species found in host A

b, number of parasite species found in host B

c, number of parasite species common for both host species.

The following infection indices were calculated: prevalence (P); intensity of infection (presented as the intensity range, IR); mean intensity (MI); abundance (A) (Bush et al. 1997).

Discriminant analysis was used to compare the infection of separate goby species. Three infection indices (P, MI, A) of individual parasite species were grouped according to host species. The squared Mahalanobis distances were presented as a plot of discriminant function.

RESULTS

The helminth fauna of gobiids surveyed consisted of 24 species (Table 1). No monogeneans were found. Four helminth species were common for all goby species studied: digenean metacercariae, Cryptocotyle concavum, C. lingua; nematode Dichelyne minutes; and acanthocephalan, Acanthocephaloides propinguus. These parasites were most widespread (Table 1) and formed the core of the helminth fauna. Telosentis exiguus infected six host species, Eustrongylides excisus-five of them, while Pygidiopsis genata and Streptocara crassicauda L3 were found in four gobiids. Five parasites species (Proteocephalus gobiorum, Asymphylodora pontica, Acanthostomum imbutiformis MET, Raphidascaris sp. L3, and Streptocara crassicauda) were common for three host fish species, while another four helminths (Bucephalus polymorphus MET, Nicolla skrjabini, Contracaecum rudolphii L3, and Acanthocephalus lucii) were found in two gobiids only. A total of ten parasite taxa were found to infect single hosts species (Bothriocephalus gregarius PL, Ligula pavlovskii PL, Proteocephalus gobiorum PL, P. subtilis, Proteocephalus sp. PL, Paratimonia gobii, Aphalloides coelomicola, Aphalloides coelomicola MET, Contracaecum microcephalum L3, and Anisakidae gen. sp. L3).

The species composition of helminth fauna of different gobiid species differed and the magnitude of differences was variable (Table 2). The helminth fauna of the marbled goby, *Pom. marmoratus*, which is of Mediterranean origin, differed markedly from these of the syrman goby, *N. syrman*, the monkey goby, *N. fluviatilis*, and the toad goby, *M. batrachocephalus* (Table 2). A high Czekanowski–Sørensen index, indicating a close similarity, was observed in the helminth fauna of the ratan goby, *N. ratan*, and the toad goby, *M. batrachocephalus* (Table 2).

A completely identical helminth-fauna make up, of four parasite species (*C. concavum*, *C. lingua*, *D. minutus*, and

						Host fis	h species				
Helminth species	Infection parameter	Pomatoschistus marmoratus (n = 56)	Neogobius melanostomus (n = 775)	N. ratan (n = 49)	N. eurycephalus $(n = 83)$	N. syrman $(n = 43)$	N. fluviatilis $(n = 731)$	Mesogobius batrachocephalus (n = 31)	Gobius niger $(n = 15)$	Zosterisessor ophiocephalus (n = 279)	Proterorhinus marmoratus (n = 40)
1	2	3	4	5	6	7	8	9	10	11	12
					CESTOD	A					
Bothriocephalus gregarius PL	P MI IR A	5.4 4.0 2-6 0.2									
Ligula pavlovskii PL	P MI IR A						11.9 1.3 1-3 0.2				
Proteocephalus gobiorum	P MI IR A					14 1.7 1-3 0.2	0.1 3.0 3 0.004			11.1 26.8 1–360 3.0	
P. gobiorum PL	P MI IR A		0.1 1.0 1 0.001			0.2	0.001				
P. subti lis	P MI IR A		0.001							1.4 1.3 1-2 0.02	
Proteocephalus sp. PL	P MI IR A			Ť	REMATO.	2.3 1.0 1 0.02					
D	Р		1.7		KEMAIU.	DA					
<i>Bucephalus polymorphus</i> MET	P MI IR A		1.7 8.2 1–32 0.1	2.0 7.0 7 0.01							
Nicolla skrjabini	P MI IR A			4.1 3.5 1-6 0.1			0.1 4.0 4 0.01				
Asymphylodora pontica	P MI IR A	8.9 6.8 1–15 0.6	4.6 19.3 1-80 0.9							0.4 1.0 1 0.004	
Paratimonia gobii	P MI IR A	17.9 20.2 1–80 3.6									
Aphalloides coelomicola	P MI IR A	5.4 4.7 1–11 0.3									
A. coelomicola MET	P MI IR A	5.4 8.3 1-20 0.4									
Acanthostomum imbutiformis MET	P MI IR A	64.3 9.8 2-61 6.3	2.3 10.4 2–40 0.2							1.1 4.0 1-6 0.04	

The helminth fauna of various gobiid species in the study area

Table 1

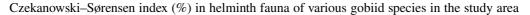
1	2	3	4	5	6	7	8	9	10	11	12
Cryptocotyle	Р	37.5	28.5	81.6	39.8	7.1	4.4	83.9	60.0	44.4	45
concavum MET	MI	6.1	141.4	53.6	7.7	22.0	203.9	15.5	6.1	9.3	17.2
	IR	1-41	1-2000	2-225	2-22	10-40	1-1500	1 - 100	1-29	1-100	1-200
	А	2.3	40.3	43.7	3.1	1.6	8.9	13.0	3.7	4.1	7.8
C. lingua MET	Р	19.6	26.2	40.8	50.6	41.9	4.8	9.7	46.7	31.9	12.5
	MI	3.7	224.9	48.3	5.0	146.2	137.0	13.3	3.6	17.6	2.2
	IR	1-13	1-2500	2-400	1-21	1-573	3-500	10-20	2-5	1-500	1-4
	А	0.7	58.9	19.7	2.5	61.2	6.6	1.3	1.7	5.6	0.3
Pygidiopsis genata	Р		4.0		12.0		3.4			1.1	
MET	MI		105.0		1.3		213.0			30.0	
	IR		1-1000		1-3		1-2500			1-81	
	А		4.2		0.2		7.3			0.3	
					NEMATO	DA					
Eustrongylides	Р		0.9	2.0		21.4	1.0	3.2			
excisus L3	MI		1.0	1.0		1.7	1.1	1.0			
	IR		1	1		1-5	1-2	1			
	А		0.01	0.02		0.4	0.01	0.03			
Raphidascaris	Р		0.3				0.1			1.4	
sp. L3	MI		1.0				1.0			1.5	
	IR		1				1			1-3	
	А		0.003				0.001			0.02	
Contracaecum	Р		0.1								
microcephalum	MI		1.0								
L3	IR		1								
<u> </u>	A		0.001								
C. rudolphii L3	Р		0.1					3.2			
	MI		1.0					1.0			
	IR		1					1 0.03			
Anisakidae gen.	A P		0.001					0.03		0.4	
	r MI									0.4 4.0	
sp. L3	IR									4.0	
	A									0.01	
Dichelyne minutus	P	8.9	25.5	22.4	13.3	16.7	31.1	25.8	13.3	4.3	17.5
Dieneryne minutus	MI	2.8	9.1	6.7	8.8	1.3	9.8	1.1	2.5	3.2	1.9
	IR	1-6	1-80	1-50	1-50	1.5	1-50	1-2	1-4	1-12	1-5
	A	0.3	2.3	1.5	1.2	0.2	3.0	0.3	0.3	0.1	0.3
Streptocara	P	0.0	0.8	14.3	112	0.12	210	9.7	0.0	2.5	012
crassicauda	MI		9.7	2.1				1.7		9.0	
L3	IR		1-50	1-5				1-3		1-50	
	А		0.1	0.3				0.2		0.2	
					NTHOCE	PHALA					
Acanthocephalus	Р		2.1				0.5				
lucii	MI		2.4				2.5				
	IR		1-5				1-6				
	А		0.05				0.01				
Acanthocephaloides propinquus	r P	5.4	6.3	6.1	1.2	19.0	0.6	3.2	93.3	64.2	22.5
	MI	1.0	5.1	2.0	1.0	12.1	2.0	1.0	9.6	46.2	4.4
	IR	1	1 - 70	1-4	1	1-35	1-3	1	2-23	1-500	1-29
	А	0.1	0.3	0.1	0.01	2.3	0.01	0.03	8.9	29.6	1.0
Telosentis exiguus	Р	1.8	2.3	10.2	3.6		0.3			30.8	
	MI	2.0	2.4	1.0	1.3		2.0			6.7	
	IR	2	1-14	1	1-2		1-14			1-63	
	А	0.04	0.1	0.1	0.05		0.01			2.1	
Species number		10	16	9	6	7	12	7	4	13	4

continued Table 1

n, number of examined gobies; PL, plerocercoid; MET, metacercaria; L3, 3rd stage larva

Table 2	
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	Pomatoschistus marmoratus	Neogobius melanostomus	N. ratan	N. eurycephalus	N. syrman	N. fluviatilis	Mesogobius batrachocephalus	Gobius niger	Zosterisessor ophiocephalus	Proterorhinus marmoratus
Pomatoschistus marmoratus	100.0									
Neogobius melanostomus	53.8	100.0								
N. ratan	52.6	64.0	100.0							
N. eurycephalus	62.5	54.5	66.7	100.0						
N. syrman	47.1	52.2	62.5	61.5	100.0					
N. fluviatilis	43.5	69.0	72.7	63.2	60.0	100.0				
Mesogobius batrachocephalus	47.1	60.9	75.0	61.5	71.4	60.0	100.0			
Gobius niger	57.1	40.0	61.5	80.0	72.7	47.1	72.7	100.0)	
Zosterisessor ophiocephalus	60.9	75.9	54.5	63.2	50.0	69.2	50.0	47.1	100.0	
Proterorhinus marmoratus	57.1	40.0	61.5	80.0	72.7	47.1	72.7	100.0	47.1	100.0



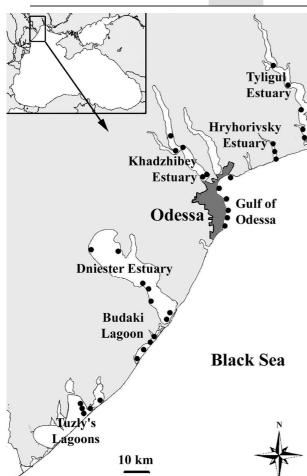


Fig. 1. A schematic map of the study area

A. propinquus) was recorded in the black-, G. niger, and tubenose-, P. marmoratus, gobies ($I_{cs} = 100\%$). Despite the similarities in overall composition of the helminth faunas of black- and tubenose gobies, their infection indices (P, %; MI; A) were significantly different (Fig. 2).

A discriminant analysis of infection indices of the core of gobiid helminth fauna (*C. concavum*, *C. lingua*, *D. minutus*, and *A. propinquus*) shows that those indices were more stable for the nematode *D. minutus* (Fig. 3). The degree of gobiid infection with the acanthocephalan *A. propinquus* and trematodes *Cryptocotyle* spp. MET differed depending on the host species (Fig. 3).

DISCUSSION

The Ponto-Caspian syrman-, *N. syrman*, and monkey-, *N. fluviatilis*, gobies as well as the marbled goby, *Pom. marmoratus* (Mediterranean species), inhabit open sandysilty sites. However, the salinity range of water for this species (6‰–40‰) is greater than that of Ponto-Caspian species (0‰–28‰) (Smirnov 1986). This may be the reason behind the differences in the species composition of the helminth fauna. The presence of specifically marine species (*Paratimonia gobii*, *Aphalloides coelomicola*) is typical for the marbled goby (Table 1). Limnetic parasites (*Bucephalus polymorphus* Met, *Nicolla skrjabini*, *Eustrongylides excisus*, *Acanthocephalus lucii*) were recovered from the Ponto-Caspian gobiids (Table 1).

The toad goby, *M. batrachocephalus*, in contrast to the marbled goby, prefers cold water and therefore lives at greater depths (Smirnov 1986). The Ponto-Caspian

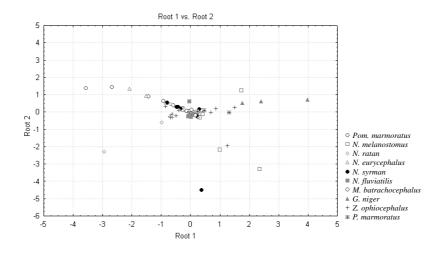


Fig. 2. Plot of discriminant function of indices of gobiid infection (P, %; MI; A) with various helminth species, P < 0.05

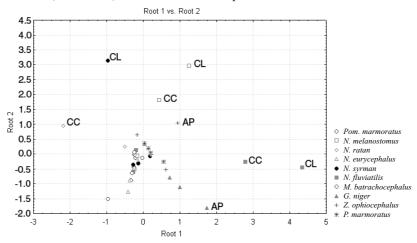


Fig. 3. Plot of discriminant function of indices of gobiid infection (P, %; MI; A) with four parasite species: *Cryptocotyle concavum* MET (CC), *C. lingua* MET (CL), *Acanthocephaloides propinquus* (AP), *Dichelyne minutus* (not marked); P < 0.05

parasites were not found in the toad goby, but the Mediterranean parasites were represented by *A. propin-quus* only. Opposite to the predatory toad goby, the ratan goby, *N. ratan*, is very similar to the mushroom goby, *N. eurycephalus*, when considering its feeding habits and diet (Smirnov 1986, Zamorov and Korenûk 2000). Both ratan and mushroom gobies are lithophilous and inhabit mainly periphyton. Migrations at greater distances (up to 150 m from the coastline) caused by wintering, fattening, and spawning, are typical for the ratan goby. This is similar for the toad goby (Smirnov 1986). It contributes to the richer helminth fauna of the ratan goby, that including common species for both mushroom- and toad gobies.

Both black-, *G. niger*, and tubenose gobies, *P. mar-moratus*, are lithophilous (inhabiting periphyton of stones and shore-protection structures), having the same helminth fauna (Tables 1, 2). The mushroom goby, *N. eurycephalus*, that are also lithophilous, had a very similar helminth fauna, $I_{cs} = 80\%$ (Table 2). All the above-mentioned gobiids feed mainly on crustaceans (Strautman 1972, Smirnov

1986, Zamorov and Korenûk 2000, Kvach and Zamorov 2001). However, the mushroom- and tubenose gobies are Ponto-Caspian relict species, but the black goby is related to the group of Mediterranean immigrants.

Thus, the species composition of the gobiid helminth fauna in the area under study is formed according to the ecology of the host species.

Despite the fact that the helminth fauna of the black goby, *G. niger*, was similar only to those of periphyton inhabitants (mushroom-, *N. eurycephalus*, and tubenose-, *P. marmoratus*, gobies; Table 2), the results of discriminant analysis show that the Mahalanobis distance in the infection indices of mushroom and tubenose gobies was low, but these of the black goby were distanced (Fig. 2). The infection indices of mushroom and tubenose gobies are similar to most of the Ponto-Caspian gobiids, but not to the black goby ones (Fig. 2). This is due to the Mediterranean origin of the black goby, which is immigrant in the Black-Sea fauna. That is why the infection indices of the black goby differed from those of most of the Ponto-Caspian species. The analysis of the core of the gobiid helminth fauna shows that the infection of the Mediterranean immigrants, the black-, *G. niger*, and grass-, *Z. ophiocephalus*, gobies with the Mediterranean marine acanthocephalan *A. propinquus* is greater, then the infection indices distanced from ones of the other gobiids (Fig. 3).

The indices of infection of Ponto-Caspian gobiids (the round goby, *N. melanostomus*, the ratan goby, *N. ratan*, the syrman goby, *N. syrman*, and the monkey goby, *N. fluviatilis*) with *Cryptocotyle* spp. MET are distanced from ones of the other gobiids too (Fig. 3). It is caused by high infection of Ponto-Caspian gobiids with metacercariae.

In my opinion the high infection of black- and grass gobies with A. propinguus was due to the same Mediterranean origin of the parasite and their hosts. It might be affected by remoteness of host-parasite relations. In spite of the fact that A. propinguus infects many hosts, the Mediterranean gobies seem to provide a better habitat for it. The high infection level of Ponto-Caspian gobiids with Cryptocotyle spp. is be related to the absence of a swim bladder. The parthenogenesis stages of Cryptocotyle develop in mud snails of the genus Hydrobia (cf. Zander et al. 2000). The Ponto-Caspian relict goby fishes cannot swim in the water for the great length of time and keep to the bottom which makes them more susceptible to Cryptocotyle cercariae, actively penetrating through the fish skin. In this way the zoogeographical origin of Ponto-Caspian gobiids promotes Cryptocotyle spp. to take up the ecological niche.

Therefore, the species composition of helminth faunas of gobiids depends mostly on the ecology of the host, but the infection indices are determined by its zoogeographical origin. More stable indices of gobiid infection were noted for the nematode *D. minutus*, but infection with other parasite species differs due to zoogeographical origin and biology of both hosts and parasites.

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