

ZOOSPORIC FUNGI GROWING ON THE SPECIMENS OF CERTAIN
FISH SPECIES RECENTLY INTRODUCED TO POLISH WATERS

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Mycoflora developing on some fish species, recently introduced to Polish waters, has not been known. The authors incubated muscles of four fish species (monkey goby, *Neogobius fluviatilis*; racer goby, *N. gymnotrachelus*; Chinese sleeper, *Perccottus glenii*; and stone moroko, *Pseudorasbora parva* caught in the drainage area of the Bug River) in water taken from 6 different places. A total of 59 fungus species were found to grow on fish muscles studied: *Achlya ambisexualis*, *A. americana*, *A. caroliniana*, *A. crenulata*, *A. debaryana*, *A. diffusa*, *A. dubia*, *A. intricata*, *A. klebsiana*, *A. oblongata*, *A. oligacantha*, *A. orion*, *A. polyandra*, *A. prolifera*, *A. proliferoides*, *A. radioasa*, *A. rodrigueziana*, *A. treleaseana*, *Aphanomyces irregularis*, *A. stellatus*, *Blastocladiella britannica*, *Blastocladiopsis parva*, *Catenaria verrucosa*, *Cladolegnia unispora*, *Dictyuchus monosporus*, *Isoachlya monilifera*, *Leptolegnia caudata*, *Leptomitius lacteus*, *Phlyctochytrium aureliae*, *Pythium afertile*, *P. aquatile*, *P. arrhenomanes*, *P. butleri*, *P. dissotocum*, *P. hemmianum*, *P. intermedium*, *P. myriotylum*, *P. ostracodes*, *P. periplocum*, *P. tenue*, *Rheosporangium aphanidermatus*, *Rhizophlyctis hirsutus*, *Rhizophydium laterale*, *R. macrosporum*, *Saprolegnia asterophora*, *S. diclina*, *S. eccentrica*, *S. ferax*, *S. hypogyna*, *S. litoralis*, *S. mixta*, *S. monoica*, *S. parasitica*, *S. pseudocrustosa*, *S. shikotsuensis*, *S. torulosa*, *S. uliginosa*, *Thraustotheca clavata*, *Zoophagus insidians*. The results obtained may be important to ichthyopathologists, because 25 of the above-mentioned fungi are known as necrotroph or parasites of fishes.

Key words: zoosporic fungi, fish, Poland

INTRODUCTION

In the last decade of the 20th century, the Polish ichthyofauna gained four new species which migrated to Poland from the south-east (Danilkiewicz 1997), namely monkey goby, *Neogobius fluviatilis* (cf. Danilkiewicz 1997), racer goby, *Neogobius gymnotrachelus* (cf. Danilkiewicz 1996), Chinese sleeper, *Perccottus glenii* (cf.

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Antychowicz 1994, Danilkiewicz 2000), and stone moroko, *Pseudorasbora parva* (cf. Witkowski 1991). One of the places they all have settled, is the middle stretch of the Bug River with some of its tributaries.

We have been focused, for some time new, on aquatic fungi, paying special attention to their contribution to the ecosystems of the respective bodies of water. Our studies have already covered all Polish freshwater fish species (Czeczuga et al. 1996, Czeczuga and Muszyńska 1997a, 1998, 1999a, b, c). This time, to have a complete picture of fungi growing on fish, found in our country, we decided to examine the species introduced recently to Polish waters.

MATERIAL AND METHODS

This study was based on the muscles of the following fish species: the monkey goby, *Neogobius fluviatilis* (Pallas, 1814); racer goby, *Neogobius gymnotrachelus* (Kessler, 1857); Chinese sleeper, *Perccottus glenii* Dybowski, 1877; and the stone moroko, *Pseudorasbora parva* (Temnick et Schlegel, 1846) which were caught in October 2000 from the Bug River near Janów Podlaski. Specimens of *Pseudorasbora parva* were also obtained from the Cieleśnica River in the same area.

The water for the experiments was collected from six different bodies of water: springs Cypisek and Jaroszkówka, ponds Fosa and Akcent, Supraśl River, and Komosa Lake. Nineteen parameters of these water samples were determined (Table 1) according to the generally accepted methods (Greenberg et al. 1992).

The following procedure was followed while determining the presence of aquatic fungus species on the fish muscles. The muscles from 5 specimens of each species (10–20 fragments) were transferred to thirty 1.0-l vessels (5 in each) and placed in the laboratory at the temperature approaching that of respective body of water. The subsamples from each vessel were observed under a microscope and the presence of mycelium (forming zoospores and oogonia) of aquatic fungi growing on muscles was recorded. The methods were described in detail by Seymour and Fuller (1987). The samples of muscles were examined through one (or one and a half) week. The experiments were carried out for three weeks.

The fungi were identified using the following keys: Johnson (1956), Sparrow (1960), Seymour (1970), Batko (1975), Dick (1990), and Watanabe (2002).

RESULTS

Hydrochemical analyses of water samples used for the experiment showed that the water from both ponds was the most abundant in nutrients, i.e. total nitrogen and phosphates, while the spring water contained fewer nutrients. This tendency was also visible in other hydrochemical parameters of these bodies of water (Table 1).

Fifty-nine zoosporic fungi species were found to grow on the muscles of four fish species introduced to the Polish waters (Table 2, Fig. 1). The fewest fungi developed on the muscles of *Pseudorasbora parva* (18), the most on *Perccottus glenii* (34

species). Such species as *Rhizophydium macrosporum*, *Saprolegnia ferax*, *Saprolegnia parasitica*, and *Saprolegnia torulosa* were observed on the muscles of all the four fish species examined. Of 59 fungus species, 25 are known as fish parasites or necrotrophs.

The most fungi were found on the fish muscles incubated in the water from two springs (Cypisek, 24; Jaroszkówka, 27), the fewest—in the water from ponds (Fosa, 7; Akcent, 11 species). In the water from Supraśl River, 17 fungus species were found on the fish muscles, while in that from Komosa Lake—18 (Table 3).

Table 1
Chemical composition (in mg·l⁻¹) of water from different sampling sites

Specification	Cypisek Spring	Jaroszkówka Spring	Supraśl River	Komosa Lake	Akcent Pond	Fosa Pond
Temperature (°C)	8.2	9.0	7.0	8.5	9.0	10.5
pH	7.17	7.63	7.55	7.87	7.38	7.94
O ₂	9.6	9.4	8.2	12.0	6.8	5.4
BOD ₅	4.2	3.6	4.0	5.8	4.0	7.8
COD	3.6	3.9	6.7	8.6	23.66	24.80
CO ₂	17.6	15.4	11.0	11.0	28.6	25.4
Alkalinity in CaCO ₃ (mval·l ⁻¹)	5.2	5.8	6.5	4.4	7.0	4.8
N (NH ₃)	0.10	0.10	0.14	0.22	2.85	3.40
N (NO ₂)	0.008	0.003	0.0	0.001	0.006	0.006
N (NO ₃)	0.035	0.760	0.0	0.0	0.010	0.010
P (PO ₄)	0.26	0.90	1.04	0.07	5.00	6.11
Sulphates	48.54	51.43	38.26	32.09	72.82	54.72
Chlorides	20.0	14.0	16.0	11.0	48.0	57.0
Total hardness in Ca	85.68	97.92	70.56	54.00	123.12	72.00
Total hardness in Mg	32.68	30.10	11.61	24.94	22.79	31.39
Fe	0.0	0.0	0.20	0.0	0.70	0.50
Dry residue	432	507	205	257	639	466
Dissolved solids	417	460	195	251	601	454
Suspended solids	15	47	10	6	38	12

Table 2

Zoosporic fungi found on the studied fish species (in water: a, Akcent Pond;
c, Cypisek Spring; f, Fosa Pond; j, Jaroszkówka Spring; k, Komosa Lake;
s, Supraśl River; *NF*, *N. fluviatilis*; *NG*, *N. gymnotrachelus*;
PG, *P. glenii*; *PP*, *P. parva*)

	Fungi	<i>NF</i>	<i>NG</i>	<i>PG</i>	<i>PP</i>
1.	<i>Achlya ambisexualis</i> J.R. Raper*			k	
2.	<i>Achlya americana</i> Humphrey*		c, s, k	c, s	c, s, k
3.	<i>Achlya caroliniana</i> Coker*			k	f
4.	<i>Achlya crenulata</i> Pringsheim			f	
5.	<i>Achlya debaryana</i> Humphrey	c	c	k	
6.	<i>Achlya diffusa</i> Harvey et Johnson*		j	j	
7.	<i>Achlya dubia</i> Coker*	k	f	k	
8.	<i>Achlya intricata</i> Beneke*		c	c	
9.	<i>Achlya klebsiana</i> Pieters*		c	c	f
10.	<i>Achlya oblongata</i> de Bary	c	j	s	
11.	<i>Achlya oligacantha</i> de Bary		j	k	
12.	<i>Achlya orion</i> Coker et Couch*			k	
13.	<i>Achlya polyandra</i> Hildebrand	c	c		
14.	<i>Achlya prolifera</i> C.G. Ness*	c	j	s	
15.	<i>Achlya proliferoides</i> Coker*		c	s	
16.	<i>Achlya radioasa</i> Maurizio*		c	f	
17.	<i>Achlya rodrigueziana</i> F.T. Wolf			j	
18.	<i>Achlya treleaseana</i> (Humphrey) Kauffman		c		
19.	<i>Aphanomyces irregularis</i> Scott	c	k		
20.	<i>Aphanomyces stellatus</i> de Bary*	c			
21.	<i>Blastocladiella britannica</i> Horstein et Cantino	k			
22.	<i>Blastocladiopsis parva</i> (Wgiffen) Sparrow			j	a
23.	<i>Catenaria verrucosa</i> Karling			a	
24.	<i>Cladolegnia unispora</i> (Coker et Couch) Johannes	c		j	
25.	<i>Dictyuchus monosporus</i> Leitgeb*		s		
26.	<i>Isoachlya monilifera</i> (de Bary) Kauffman*			c	
27.	<i>Leptolegnia caudata</i> de Bary*				j
28.	<i>Leptomitius lacteus</i> (Roth) Agardth*	s	s	s	
29.	<i>Phlyctochytrium aureliae</i> Ajello			c	k
30.	<i>Pythium afertile</i> Kanouse et Humphrey				j
31.	<i>Pythium aquatile</i> Hohnk	j			
32.	<i>Pythium arrhenomanes</i> Sideris			k	
33.	<i>Pythium butleri</i> Subramaniam				j
34.	<i>Pythium dissotocum</i> Drechsler			a	
35.	<i>Pythium hemmianum</i> Takahashi	k			
36.	<i>Pythium intermedium</i> de Bary			j	a
37.	<i>Pythium myriotylum</i> Drechsler				j
38.	<i>Pythium ostracodes</i> Drechsler				a, k, s
39.	<i>Pythium periplocum</i> Drechsler			a	
40.	<i>Pythium tenue</i> Gobi	a, j			a
41.	<i>Rheosporangium aphanidermatus</i> (Edson) Fitzpatrick				c
42.	<i>Rhizophlyctis hirsutus</i> Karling	k			

Table 2 (cont.)

	Fungi	NF	NG	PG	PP
43.	<i>Rhizophydium laterale</i> (Braun) Rabenhorst	s			
44.	<i>Rhizophydium macrosporum</i> Karling	c	j	a	s
45.	<i>Saprolegnia asterophora</i> de Bary		j, k, s		
46.	<i>Saprolegnia diclina</i> Humphrey*		j		
47.	<i>Saprolegnia eccentrica</i> Coker	c			
48.	<i>Saprolegnia ferax</i> (Gruith.) Thuret*	c, s	j	a, k	f
49.	<i>Saprolegnia hypogyna</i> (Pringsheim) de Bary	c		c	c
50.	<i>Saprolegnia litoralis</i> Coker	c	j	s	
51.	<i>Saprolegnia mixta</i> de Bary*		j		
52.	<i>Saprolegnia monoica</i> Pringsheim*		a, c, s		
53.	<i>Saprolegnia parasitica</i> Coker*	c	j	k	a
54.	<i>Saprolegnia pseudocrustosa</i> Lund		j		
55.	<i>Saprolegnia shikotsuensis</i> Hatai et al.*		f, j	k	
56.	<i>Saprolegnia torulosa</i> de Bary*	c	j	j	c
57.	<i>Saprolegnia uliginosa</i> Johannes		s		
58.	<i>Thraustotheca clavata</i> (de Bary) Humphrey			j	
59.	<i>Zoophagus insidians</i> Sommerstorff		j		
Total number		22	29	34	18

* species known as parasites or necrotrophs of fishes

Table 3

Aquatic fungi found on the fishes studied, in water from different places

Water from	Fungi (see Table 2)	Total number
Cypisek Spring	2,5,8,9,10,13,14,15,16,18,19,20,24,26,29,41,44,47,48,49,50,52,53,56	24
Jaroszówka Spring	6,7,10,11,14,17,22,24,27,30,31,33,36,37,40,44,45,46,48,50,51,53,54,55,56,58,59	27
Supraśl River	2,5,9,10,14,15,25,28,38,43,44,45,48,50,51,52,57	17
Komosa Lake	1,2,3,5,7,11,12,19,21,29,32,35,38,42,45,48,53,55	18
Akcent Pond	22,23,34,36,38,39,40,44,48,52,53	11
Fosa Pond	3,4,7,9,16,48,55	7

DISCUSSION

As revealed by the present study, species composition of aquatic zoosporic fungi growing on the muscles of the four species introduced to the Polish waters does not differ from the species composition encountered on native species (Czeczuga and Muszyńska 1999a, b).

Of 25 fungus species known as fish parasites and found during the present study, some were only found on the specimens of one species, others on two, while 6 fungus species were observed on the specimens of 3 fish species. *Saprolegnia ferax*, *Saprolegnia parasitica*, and *Saprolegnia torulosa* were observed on all the four fish

species examined. The fungus species found to grow on 3 out of 4 fish species included *Achlya americana*, *Leptomitius lacteus*, and *Saprolegnia hypogyna*.

Saprolegnia ferax and *Saprolegnia parasitica* most frequently cause death of fish both in aquaculture, and under natural conditions. At the end of the nineteenth century, initially on the British Isles and then on the European Continent, one of the *Saprolegnia* species caused mass deaths of salmon, *Salmo salar* entering rivers for spawning (Neish and Hughes 1980). At present it is the main cause of fish mortalities in lakes (Meng 1980), ponds (Lartseva and Dudka 1990), and most of all in hatcheries (Florinskaja 1971). *Saprolegnia parasitica* also causes fish losses under natural conditions and in aquaculture. It is especially well-known in Japanese hatcheries (Hatai et al. 1990) and in the aquaculture of *Oncorhynchus kisutch* where it can kill the majority of its population (Hatai and Hoshiai 1992a, b). *Saprolegnia torulosa* has been encountered on eggs of a few coregonid fish species (Czeczuga and Muszyńska 1998) and on dead specimens of young eel *Anguilla anguilla* (Czeczuga 1994, Czeczuga and Muszyńska 2000). This fungus is most frequently encountered as a soil and aquatic saprophyte (Batko 1975). This refers to a number of zoosporic fish species which were first described as saprophytes and then turned out to occur on living plant or animal organisms. For instance, representatives of the genus *Pythium*, known as plant root parasites, particularly of crop plants, can be also found to grow in aquatic conditions on fish eggs (Czeczuga 1996).

Similarly, *Achlya americana*, *Achlya dubia*, *Achlya klebsiana*, and *Achlya prolifera* were described as soil or aquatic saprophytes and as such have been presented in textbooks (Batko 1975). The following species, however, were found to lead a parasitic mode of life on fish: *Achlya americana* (cf. Scott and Warren 1964), *Achlya dubia* (cf. Bhargava et al. 1971), *Achlya klebsiana* (cf. Vishniac and Nigrelli 1957), and *Achlya prolifera* (cf. Srivastava 1976, Srivastava and Srivastava 1977). Our studies have revealed that these zoosporic fungus species grow on the eggs of numerous freshwater fish species (Czeczuga and Muszyńska 1997a, 1999c).

In mycological monographs, *Leptomitius lacteus* has been known as a sewage fungus (Batko 1975, McLaughlin et al. 2001) and a typical mycoflora representative of waters strongly polluted with municipal wastes. However, Lemon (1954) found this fungus to be a parasite of sea lamprey, while Willoughby (1970) encountered it on the specimens of young perch in Windermere. Our investigations observed its growth on coregonid eggs incubated in hatcheries (Czeczuga and Woronowicz 1993).

Saprolegnia hypogyna is known as a soil and aquatic saprophyte, and a facultative parasite on fish eggs. We found this fungus on the eggs of many fish species, including salmonids (Czeczuga and Muszyńska 1996, Czeczuga et al. 1996).

Like in the case of other fish species (Czeczuga et al. 1995, 2001, Czeczuga and Muszyńska 1997b, Czeczuga and Kiziewicz 1999), also in the four species examined, the most aquatic zoosporic fungus species were found to grow on fish living in less eutrophic waters (springs Jaroszkówka and Cypisek), the fewest in polytrophic-type

waters (ponds Fosa and Akcent). This could be associated with the phenomenon, known in hydrobiology, that in oligotrophic type waters the species number of hydrobionts is higher, being decreased in more eutrophic types; in polytrophic waters only one or two and seldom a few hydrobionts grow intensely. As shown in our previous and present study this also refers to aquatic zoosporic fungi.

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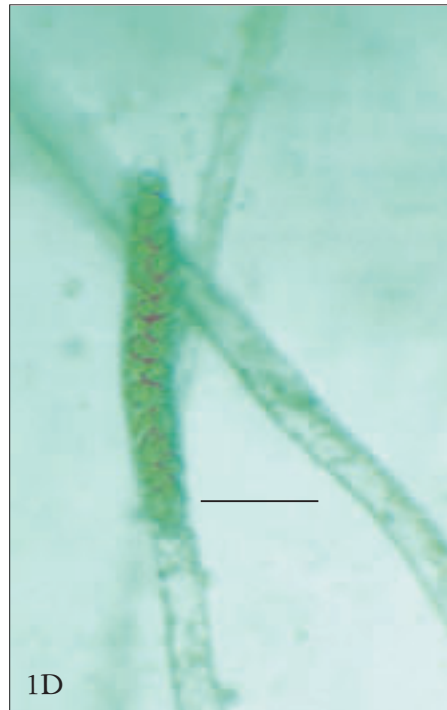


Fig. 1. Some zoosporic fungus species growing on the specimens of fish
 Scale bar = 50 μ m. A – *Achlya debaryana* – oogonia, B – *Achlya orion* – oogonium, C – *Saprolegnia ferax* – proliferating hyphae, D – *Saprolegnia uliginosa* – sporangium.