Polish Journal of Environmental Studies Vol. 8, No. 6 (1999), 377-382

Zoosporic Fungus Species Growing on Dead Benthos Crustaceans

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Received 8 June, 1999
Accepted 24 June, 1999

Abstract

The authors investigated the mycoflora developing on the dead specimens of four species of benthos crustaceans. Ninety-five zoosporic fungus species were found to grow on the fragments of investigated crustaceans. Out of these 95 species, 27 are known as parasites or necrotrophs of fish. Four fungus species were recorded for the first time in Polish waters.

Keywords: benthos crustaceans, aquatic fungi, hydrochemistry

Introduction

Our investigations of zoosporic fungi growing on four crayfish species in the waters of northeastern Poland [9] and on the carapaces of dead zooplankton [7] established a number of rare or new species for Polish hydromycology. Many fungus species growing on crustaceans have long been known as aggressive parasites of economically valuable fish species [8, 15, 20].

Crustaceans present in the so called benthos are a specific environmental group, inhabiting the bottom of water basins - rich in plant residue with numerous species of saprophytic fungi.

Some benthos crustacean species are common in Central Europe [30] and live in running waters [27]; others, such as *Pallasiola quadrispinosa,* are relict crustaceans inhabiting deep lakes [12, 39].

The aim of our study was to establish zoosporic fungus species growing on bottom crustaceans, being a potential source of infection for fish.

Material and Methods

The investigations concerned the following crustacean species:

- *Asellus aquaticus* (Linne, 1758)
- *Gammarus fossarum* (Koch, 1835)
- *Gammarus lacustris* (G.O. Sars, 1863)
- *Gammarus pulex* (Linne, 1758)
- *Pallasiola quadrispinosa* (G.O. Sars, 1867)

which were obtained from water bodies of northeastern Poland {*Pallasiola quadrispinosa* from Wigry Lake).

The water for experiments was collected in April 1998 from seven different water bodies: Cypisek Spring, Jaroszowka Spring, Biata River, Suprasl River, Akcent pond and Fosa in Branicki Park and Lake Komosa. Nineteen parameters of these water samples were determined (Table 1) according to the standard methods [17].

For determining the presence of aquatic fungus species on the crustaceans, the following procedure was employed: a certain amount (10-15 little pieces) of body parts of each species of crustaceans were transfered (two for each water) to a 1.0 litre vessel (together for each species was fourteen vessels) and placed in the laboratory at a temperature approaching the ambient outside given one (about 6-8ºC). A part of the pieces from each vessel was observed under a microscope and the mycelium (zoosporic, antheridia and oogonia forms) of aquatic fungi growing on the pieces of crustaceans was recorded. The methods were described in detail in Czeczuga et al. [10].

The pieces of the various crustacean species were examined for one and a half weeks. The incubation period was three weeks.

The following keys were used to indentify fungi: Batko [1], Johnson [23], Seymour [34], Karling [24] and Dick [13].

Results

Chemical analysis of water samples collected for study from seven basins revealed that spring water was the poor-

Table 1. Chemical composition (in mg l^{-1}) of the different water samples

Specification	Spring		River		Pond		Lake Komosa
	Cypisek	Jaroszówka	Biała	Supraśl	Akcent	Fosa	
Temperature °C	8.2	8.4	13.2	12.8	16.4	15.6	14.2
pH	7.04	7.14	7.13	6.99	7.02	7.08	7.03
O_2	4.52	7.15	4.08	5.12	4.16	3.09	7.12
BOD_5	3.9	5.0	2.0	4.1	0.3	0.5	4.6
Oxidability (COD)	3.24	4.18	10.62	9.36	20.52	15.48	8.42
CO_2	28.6	17.4	30.8	15.6	22.4	11.2	12.5
Alkalinity in $CaCO_3$ (in mval l^{-1})	7.0	6.1	5.8	5.6	7.2	4.7	4.1
$N(NH_3)$	0.03	0.12	0.68	0.14	0.44	0.36	0.02
$N(NO_2)$	0.092	0.025	0.068	0.012	0.008	0.004	0.005
$N(NO_3)$	0.009	0.008	0.124	0.038	0.112	0.096	0.024
PO_4^{3-}	0.124	0.186	0.512	0.306	0.816	0.624	0.215
Cl^-	24.0	25.4	41.8	18.8	50.2	46.4	25.0
Total hardness in Ca	140.4	108.7	106.6	87.1	135.4	84.9	70.6
Total hardness in Mg	35.26	40.4	35.7	24.3	39.6	33.0	33.5
SO_4^{2-}	64.6	61.3	42.8	25.4	83.2	48.4	37.2
Fe (Fe^{+2} and Fe^{+3})	0.32	0.44	0.75	0.74	0.78	0.52	0.45
Dry residue	291.0	131.2	442.0	205.0	563.0	421.0	232.0
Dissolved solids	216.0	130.0	434.0	203.0	548.0	414.0	224.0
Suspended solids	75.0	1.2	8.0	2.0	15.0	7.0	8.0

est while pond water (Fosa and Akcent) the richest in biogenes. The same also refers to oxidability, an indicator of organic matter content in water (Table 1). Ninety-five aquatic fungus species belonging to Chytridiomycetes (17 species), Hyphochytriomycetes (1), Oomycetes (76) and Zygomycetes (1) were found on 5 benthos crustacean species. Most fungus species were found on *Pallasiola quadrispinosa* (60), fewest on *Asellus aquaticus* and *Gammarus lacustris* (34 on each). Such fungus species as *Achlyaa apiculata, Achlya klebsiana, Aphanomyces irregularis, Saprolegnia delica, Saprolegnia ferax* and *Saprolegnia parasitica* were noted on all crustaceans examined (Table 2). Some species displayed fungi new to Polish waters: *Saprolegnia pseudocrustosa* and *Saprolegnia richteri* were found on *Gammarus fossarum, Saprolegnia stagnalis* on *Gammarus lacustris* and *Pythium ostracodes* on *Pallasiola quadrispinosa.*

Fewest aquatic fungus species were found to develop on the dead benthos crustaceans in pond water (Table 3).

Discussion

The present study has proved that the growth of respective aquatic fungus species on dead benthos crustaceans depends on the water body from which water has been collected for the experiment. The fewest species developed on crustaceans in pond water. Chemical analysis of the water collected from these seven water bodies showed differences regarding the content of chemical compounds. Pond water showed considerably more oxidability, phosphorus and chlorides.

The present study revealed a relatively large number of zoosporic fungi on fragments of 5 benthos crustacean species, including four new to Polish waters as well as many species which cause damage on fish farms as parasities or necrotrophs. *Saprolelegnia pseudocrustosa* is a new species. It was first found in Danish waters and described by Lund [26]. *Saprolegnia richteri* was first isolated from swamp meadow soil in the vicinity of Marburg in Germany as *Isoachlya terrestris* by Richter [31]. Later, Johannes [22] included it into the genus *Cladolegnia* and then Seymour [34] placed it among the genus *Saprolegnia. Saprolegnia stagnalis,* first described by Tiesenhausen [38] from the waters of Switzerland, belongs to aquatic saprophytes [1]. *Pythium ostracodes* was first isolated by Drechsler [14] from radicibus *Tritici aestivi* in Texas.

Aphanomyces astaci, a frequent cause of plague among noble crayfish [9], was found on *Asellus aquaticus, Gammarus fossarum* and *Pallasiola quadrispinosa* individuals. Fungus species growing on benthos crustaceans and fish include *Achlya polyandra, Achlya prolifera, Aphanomyces laevis, Dictyuchus monosporus, Saprolegnia diclina, Saprolegnia ferax, Saprolegnia parasitica* and *Saprolegnia shikotsuensis.* Sometimes damage is great, e.g. on a fish farm in England the whole fish fry of *Anguilla anguilla* died of saprolegniosis [2]. *Achlya polyandra* is known as a parasite of the eggs [29] and grown-up individuals of salmonids [16], while *Achlya prolifera* frequently causes total loss of eggs in hatcheries [32]. *Aphanomyces laevis* attacks both eggs [25] and adult individuals of many economically valuable fish species [15]. *Dictyuchus monosporus* causes damage to acipenserids, salmonids and cyprinids in hatcheries [15, 16, 25]. Fungus species of the genus *Pythium* has been long known to infect many fish species [33]. Because of difficulty in defining species status in this group, most investigators use only generic terms [35, 37]. Two *Pythium* species - *Pythium proliferum* [16] and *Pythium ultimum* [33] are most common for fish.

Our detailed studies of fungi of the genus *Pythium* revealed several species on fish eggs [3]. *Saprolegnia diclina* frequently affects the eggs of salmonids in hatcheries [11, 21]

Table 2. Aquatic fungi found on the crustacean specimens

Classis, Ordo and species fungi	*A. aquaticus*	*G. fossarum*	*G. lacustris*	*G. pulex*	*P. quadrispinosa*
Chytridiomycetes					
Chitriodiales					
1. *Asterophlyctis irregularis* Karling		x			x
2. *Chytriomyces aureus* Karling	x	x	x		x
3. *Chytriomyces hyalinus* Karling	x	x			x
4. *Karlingia chitinophila* Karling					x
5. *Obelidium mucronatum* Nowakowski			x		
6. *Phlyctochytrium aureliae* Ajello	x			x	
7. *Podochytrium chitinophilum* Willoughby		x			x
8. *Rhizoclosmatium hyalinum* Karling		x			
9. *Rhizidium chitinophilum* Sparrow		x	x	x	x
10. *Rhizidium nowakowskii* Karling					x
11. *Rhizophydium laterale* (Braun) Fisher		x			
12. *Rhopalophlyctis sarcoptoides* Karling					x
13. *Siphonaria variabilis* Petersen			x		x
Blastocladiales					
14. *Allomyces arbuscula* Butler	x				x
15. *Blastocladiella britannica* Horenstein et Cantino		x			x
16. *Blastocladiopsis parva* (Whiffen) Sparrow	x			x	
17. *Catenaria anguillulae* Sorokin			x		
Hyphochytriomycetes					
Hyphochytriales					
18. *Rhizidiomyces bivellatus* Nabel	x				x
Oomycetes					
Lagenidiales					
19. *Myzocytium zoophthorum* Sparrow		x			x
Saprolegniales					
20. **Achlya americana* Humphrey		x	x	x	
21. *Achlya apiculata* de Bary	x	x	x	x	x
22. **Achlya bisexualis* Coker et A. Couch					x
23. **Achlya caroliniana* Coker	x	x		x	x
24. *Achlya colorata* Pringsheim	x			x	
25. *Achlya debaryana* Humphrey		x	x		x
26. **Achlya diffusa* Harvey ex Johnson		x			x
27. **Achlya dubia* Coker	x			x	
28. *Achlya hypogyna* Coker et Pemberton				x	
29. **Achlya klebsiana* Pieters	x	x	x	x	x
30. *Achlya megasperma* Humphrey	x				
31. *Achlya oblongata* de Bary					x
32. *Achlya oligacantha* de Bary					x
33. **Achlya orion* Coker et Couch	x		x	x	x
34. *Achlya polyandra* Hildebrand	x			x	x
35. **Achlya prolifera* Nees			x	x	x
36. **Achlya proliferoides* Coker	x	x	x	x	
37. **Achlya racemosa* Hildebrand	x			x	
38. *Achlya treleaseana* (Humphrey) Kauffman	x			x	
39. *Aphanomyces amphigynus* Cutter	x		x		
40. *Aphanomyces astaci* Schikora	x	x			x
41. *Aphanomyces irregularis* Scott	x	x	x	x	x
42. **Aphanomyces laevis* de Bary	x	x		x	x
43. **Aphanomyces stellatus* de Bary				x	x
44. *Aplanes androgynus* (Archer) Humphrey		x	x	x	x
45. *Apodachlya pyrifera* Zopf				x	
46. *Calyptralegnia basraensis* Muhsin				x	
47. *Cladolegnia unisospora* (Coker et Couch) Johannes		x			x
48. **Dictyuchus monosporus* Leitgeb				x	
49. **Dictyuchus sterillis* Leitgeb	x		x	x	x
50. **Isoachlya monilifera* (de Bary) Kauffman		x	x		x
51. **Leptolegnia caudata* de Bary			x	x	x
52. **Pythiopsis cymosa* de Bary	x				
53. *Saprolegnia anisospora* de Bary			x		x

Table 2. Continuation

Classis, Ordo and species fungi	*A. aquaticus*	*G. fossarum*	*G. lacustris*	*G. pulex*	*P. quadrispinosa*
54. Saprolegnia asterophora (de Bary) Johannes	x	x		x	x
55. **Saprolegnia delica* Coker	x	x	x	x	x
56. **Saprolegnia diclina* Humphrey	x				
57. *Saprolegnia eccentrica* (Coker) Seymour					x
58. **Saprolegnia ferax* (Gruith.) Thuret	x	x	x	x	x
59. *Saprolegnia glomerata* (Tiesenhausen) Lund		x	x	x	x
60. *Saprolegnia latavica* Apinis				x	x
61. *Saprolegnia litoralis* Coker			x		
62. **Saprolegnia monoica* Pringsheim			x	x	
63. **Saprolegnia parasitica* Coker	x	x	x	x	x
64. *Saprolegnia pseudocrustosa* Lund		x			x
65. *Saprolegnia richteri* Richter ex Seymour		x			
66. **Saprolegnia shikotsuensis* Hatai et al.		x			x
67. *Saprolegnia stagnalis* Tiesenhausen			x		
68. *Saprolegnia torulosa* (de Bary) Cejp	x		x	x	x
69. *Saprolegnia uliginosa* Johannes			x		x
70. *Saprolegnia unispora* Coker et Couch	x			x	
71. **Thraustotheca clavata* (de Bary) Humphrey		x	x	x	
Leptomitales					
72. **Leptomitus lacteus* (Roth) Agardh	x			x	
Peronosporales					
73. *Pythium acanthicum* Drechsler		x			x
74. *Pythium afertile* Kanouse et Humphrey		x			x
75. *Pythium aquatile* Höhnk		x		x	x
76. *Pythium butleri* Subramaniam					x
77. *Pythium catenulatum* Matthews				x	x
78. *Pythium debaryanum* Hesse	x	x		x	
79. *Pythium dissotocum* Drechsler		x			x
80. *Pythium elongatum* Matthews			x		x
81. *Pythium helicandrum* Drechsler		x	x		x
82. *Pythium inflatum* Matthews				x	x
83. *Pythium intermedium* de Bary		x			
84. *Pythium jirovecii* Cejp		x			x
85. *Pythium myriotylum* Drechsler					x
86. *Pythium oligandrum* Drechsler		x			
87. *Pythium ostracodes* Drechsler					x
88. *Pythium polysporum* Sorokin					x
89. **Pythium proliferum* de Bary non Schenk		x		x	x
90. *Pythium pulchrum* Minden		x			
91. *Pythium rostratum* Butler	x	x	x	x	x
92. *Pythium torulosum* Coker et Patterson			x		
93. **Pythium ultimum* Trow			x	x	
94. *Zoophagus insidians* Sommerstorff				x	
Zygomycetes					
Entomophthorales					
95. *Zoophthora rhizospora* (Thaxfer) Batko	x			x	
Total number	34	43	34	45	60

* Known in literature as parasites or necrotrophs of fish

and also salmonid fish fry in hatcheries [18]. *Saprolegnia shikotsuensis* was first described from *Oncorhynchus nerka* var. *adonis* individuals from Shikotsu lake of Hokkaido Island, Japan [19].

Our further studies confirmed its presence on the eggs of European fish species [8], lampreys [4] and on eggs of certain amphibians [10]. The growth of *Saprolegnia shikotsuensis* was also observed on the carapaces of several species of higher crayfish [9]. However, the greatest damage in fish farming is due to *Saprolegnia ferax* and *Saprolegnia parasitica* [28]. Since the previous century *Saprolegnia ferax* has been known to cause epizooties of Atlantic salmon in the rivers of Europe [36]. In recent decades *Saprolegnia ferax* has caused major damage to acipenserid farming in the river Volga [25]. *Saprolegnia parasitica* causes huge losses in various hatcheries, affecting both eggs [33] and fish fry [35]. Known are the cases when half of the fish-breeding die due to *Saprolegnia parasitica-induced* saprolegniosis. In the 1980s on salmonid farms in Miyagi Prefecture in Japan this fungus frequently caused mass

Table 3. Aquatic fungi found on specimens of benthos crustacean in different waters (numbers as in Table 2)

Water from	Fungi (see Table 2)	Only in one water	Number of fungi
Cypisek Spring	1,9,14,17,18,21,23,25,29,31,33,34,35,36,39,40,41,42,44,46,47,49,50,51,52,53,55, 57,58,59,61,62,63,64,67,68,71,73,74,75,78,80,81,82,84,91,95	17,52,67,95	47
Jaroszówka Spring	1,2,5,6,9,14,24,25,29,31,33,34,35,36,37,40,41,42,44,47,49,50,53,54,55,58,59,62, 63,64,65,66,68,69,70,74,77,78,81,82,83,85,87,89,90,93	24,37,65,66,83,85,87	46
Biała River	2,3,7,9,15,16,19,20,21,23,25,29,32,33,34,35,36,40,41,42,44,46,47,50,51,54,55,56, 57,58,59,60,63,68,69,70,72,74,75,78,79,81,82,88,89,91,94	16,56,88	47
Supraśl River	1,2,3,7,8,9,10,13,18,19,20,21,23,25,27,29,33,34,35,36,38,40,41,42,43,44,45,47, 50,53,54,55,57,58,60,62,63,68,69,70,71,72,73,74,75,79,82,84,86,89,90,91,92,93	8,10,27,86,90,92	54
Akcent Pond	1,2,4,5,6,9,11,13,15,18,19,25,29,34,38,39,40,41,42,44,48,50,51,55,58,59,60,62, 63,68,70,71,72,74,75,78,81,91,93	4	39
Fosa Pond	1,2,3,9,11,20,21,25,26,29,32,33,34,36,40,41,42,43,44,48,49,50,51,53,55,58,59, 61,63,64,74,75,78,82,91,94	26	36
Komosa Lake	2,7,9,12,15,18,19,21,22,25,28,29,30,31,32,33,34,35,36,40,41,42,44,45,47,49,50, 51,53,54,55,57,58,59,62,63,64,68,69,70,71,73,74,76,77,78,80,89,94	12,22,28,30	49

death of Pacific salmonid *Oncorhynchus kisutch* [20]. Also *Thraustotheca clavata* was observed on various fish species **[15].**

Moreover, worth noting is the finding of *Aphanomyces astaci* on fragments of the three benthos crustacean species examined. This fungus is known to cause the so-called crayfish plague among Decapoda representatives [9].

The present study has revealed that benthos crustaceans are the vectors of many fungus species, being fish parasites and of a fungus which induces crayfish plague.

Acklowledgement

The authors wishes to thank Professor S. Niewolak from University of Agriculture and Technology at Olsztyn-Kortowo and MSc W. Mikulski from University at Biatystok for their kind help in obtaining some species of investigated crustaceans.

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