# A C T A I C H T H Y O L O G I C A E T P I S C A T O R I A Vol. XXV, Fasc. 2 Szczecin 1995

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Fish eggs fungi

### AQUATIC FUNGI GROWING ON THE EGGS OF SEVERAL SPECIES OF ACIPENSERID FISHES \*

## GRZYBY WODNE ROZWIJAJĄCE SIĘ NA IKRZE KILKU GATUNKÓW RYB JESIOTROWATYCH

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The authors investigated the mycoflora developing on the eggs of six species of acipenserid fishes.

#### INTRODUCTION

Acipenserid fish species which live mainly in the basins of the Black Sea and the Caspian Sea provide valuable meat and eggs, the latter commonly known as caviar. Sturgeon is a migratory two-environmental fish, which only occasionally lives in one environment—river or lake.

River dams form an obstacle for migratory species to reach their natural spawninggrounds. Therefore, in order to maintain the proper stock of economically valuable species, breeding of the young has been commenced in hatcheries. The literature devoted to artificial propagation of acipenserid fishes reports considerable mortality rate of eggs due to saprolegnia fungus infection. Frequently, this loss amounts to 70–90% of the incubated eggs (Lartseva 1986b; Lartseva and Altufiev 1987).

The studies on the occurrence of zoosporic fungi in acipenserids go back to the end of the previous century. Clinton (1894) reported the occurrence of *Saprolegnia ferax* fungus on eggs of *Acipenser sturio* L. specimens. In the years to follow, this subject was not investigated. Only when the intensive breeding of acipenserid fishes started in artificial conditions, where a great loss of eggs due to aquatic fungus infection occured (Lartseva and Altufiev 1987), the interest in saprolegnosis in the acipenserids increased. A number of reports have been issued lately on the occurrence of zoosporic fungi on the incubated eggs in the basin of the Caspian Sea, referring mainly to such sturgeons as *Huso huso, Acipenser güldenstädti* and *Acipenser stellatus* (cf. Lartseva 1986; Lartseva and Altufiev 1987; Lartseva and Dudka 1990). Also in Poland, an interest has developed in acipenserid fishes, expressed by eggs import, hatching and breeding of a variety of acipenserid fish species (Kolman 1993).

Part 41 in the series "Studies of Aquatic Fungi"

Thus, we have decided to publish the data concerning the development of lower zosporic fungi on certain acipenserid species in the conditions of our inland waters.

### MATERIAL AND METHODS

The investigations included the eggs of the following fish species: Acipenser güldenstädti Brandt, Acipenser güldenstädti persicus Borodine, Acipenser rudiventris Lovetzky, Acipenser ruthenus L., Acipenser stellatus Pallas and Huso huso (L.) which were obtained from hatcheries of the Kaspijskij Naučno-issledovatelskij Institut Rybnogo Choziajstva, Astrachań, Russia and from hatcheries of the Department of Ichthyology, University of Tehran, Iran. The materials were transported in a thermos flask by air mail.

 Table 1

 Chemical composition (in mg-l<sup>-1</sup>) of the different water (October, 1994)

Specifcation	Pond	Lake	River
Temperature °C	6.8	6.2	6.1
pH	7.1	7.6	7.5
<b>O</b> <sub>2</sub>	4.6	8.2	11.4
BOD <sub>5</sub>	4.2	1.6	2.2
Oxidability	9.5	7.2	7.6
CO <sub>2</sub>	12.1	13.2	17.8
Alkalinity in CaCO3 *	4.6	3.9	4.1
N (NH <sub>3</sub> )	0.78	0.10	0.20
N (NO <sub>2</sub> )	0.05	0.01	0.02
N (NO <sub>3</sub> )	0.70	0.06	0.12
PO <sub>4</sub>	7.12	0.18	0.36
Cl	69.0	23.0	28.0
Total hardness in Ca	84.96	68.40	76.52
Total hardness in Mg	28.81	28.82	31.00
SO <sub>4</sub>	56.77	12.75	19.85
Fe	1.50	0.20	0.50
Dry residue	394.0	317.0	373.0
Dissolved solids	362.0	305.0	312.0
Suspended solids	32.0	12.0	61.0

\* in mval l<sup>-1</sup>

Water for the experiments was collected from three different water bodies: the Supraśl River, pond in Branicki Park and Lake Komosa. Nineteen parameters of these water samples were determined (Tab. 1) according to the generally accepted methods (Golterman and Clymo 1969).

For the determinations of the presence of aquatic fungus species on the eggs, the following procedure was employed: a certain amount of eggs of each species of fish (100–200) was transferred to a 1.0-litre vessel and placed in the laboratory at temperature approaching that of the given hatchery. Then part of the eggs from each vessel was observed under a microscope and the mycelium (form zoospore, oogonia and condidia) of aquatic fungi growing on the eggs

was recorded. The methods were described in detail in the paper of Fuller and Jaworski (1986). The eggs of the various fish species were examined for one, up to one and a half weeks. The eggs were mostly live but sometimes dead.

For determinations of the fungi the following keys were used: Johnson (1956), Seymour (1970), Kreger van Rij (1984) and Dick (1990).

#### RESULTS

Forty-two species of zoosporic fungi and *Candida albicans* conidial fungus were detected on the eggs of six species of acipenserid fish (Tab. 2–3). The fewest species were observed on the eggs of *Huso huso* (7) and on the eggs of *Acipenser güldenstädti persicus* (8). The remaining four species had twice more zoosporic fungi (14–16 species). The data obtained show that among the forty-two fungi noted on the eggs of acipenserid fishes, only nine fungus species were previously found in these fishes; thus, the remaining thirty-three are new to the acipenserids and they still include eight species new to fishes in general (Fig. 1).

#### Table 2

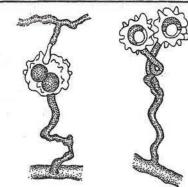
Species	A.gülden-		A.nudi-	A.ruthe-	A.stella-	H.huso
-	städti	sicus	ventris	nus	tus	
Achlya ambisexualis Raper			×			
Achlya americana Humphrey					×	
Achlya bisexualis Coker et Couch	×				×	
Achlya caroliniana Coker				×	×	
Achlya colorata Pringsheim		×				
Achlya diffusa Harvey et Johnson	×		×			×
Achlya dubia Coker				×	×	
Achlya glomerata Coker	×					
Achlya hypogyna Coker et Pemberton	×		×	×		8
Achlya klebsiana Pieters	×			1		i i i
Achlya megasperma Humphrey			×			
Achlya oligacantha de Bary				×		
Achlya orion Coker et Couch	×	- N	×			
Achlya polyandra Hildebrand	×					
Achlya racemosa Hildebrand	×					
Achlya radiosa Maurizio	×					
Achlya treleaseana (Humph.) Kauffman	×		×			
Allomyces anomalus Emerson				×	×	(
Allomyces arbuscula Butler		×				
Aplanes androgynus (Archer) Humphrey		×		×		
Blastocladiopsis parva (Whiffen) Sparrow	×		×			1
Calyptralegnia achlydoides (Coker et Couch) Coker					×	
Dictyuchus sterilis Coker	×	×		×		
Isoachlya anisospora (de Bary) Coker		×		×		
Isoachlya monilifera (de Bary) Kauffman					×	
Isoachlya toruloides Kauffman et Coker				1	×	
Leptolegnia caudata de Bary	1		×	×		
Leptomitus lacetus (Roth) Agardh	×	×	×	×	×	×
Protoachlya paradoxa (Coker) Coker			1		×	
Pythiopsis cymosa de Bary				×		
Pythium artotrogus de Bary	· · 6		×	1		
Pythium ultimum Trow			×		1	
Saprolegnia australis Elliott		×	×		×	
Saprolegnia diclina Humphrey					×	
Saprolegnia ferax (Gruith) Thurnet			×	1	×	×
Saprolegnia mixta de Bery						×
Saprolegnia monoica Pringsheim					×	
Saprolegnia parasitica Coker	×	×	×	×		×
Saprolegnia shikotsuensis Hatai et al.	×		x	×		×
Saprolegnia unispora (Coker et Couch) Seymour				×		
<i>Thraustotheca clavata</i> (de Bary) Humphrey				×		
Zoopage phanera Drechsler	×					
Candida albicans (Robin) Berkhout (conidial funei)						×
		1			in the second se	

#### Aquatic fungi found on the eggs of acipenserid fishes

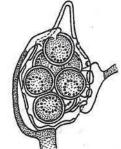
# Table 3

Species	Pond	Lake	River
Achlya ambisexualis Raper	×		
Achlya americana Humphrey	×	×	
Achlya bisexualis Coker et Couch		×	
Achlva caroliniana Coker	×		×
Achlya colorata Pringsheim	×		×
Achlya diffusa Harvey et Johnson	×		×
Achlya dubia Coker		×	
Achlya glomerata Coker	×		
Achlya hypogyna Coker et Pemberton	×		×
Achlya klebsiana Pieters	×		
Achlya megasperma Humphrey	×		
Achlya oligacantha de Bary	×		
Achlva orion Coker et Couch	×		×
Achlya polyandra Hildebrand		1	×
Achlya racemosa Hildebrand	×		
Achlva radiosa Maurizio	×		×
Achlya treleaseana (Humph.) Kaufiman			×
Allomyces anomalus Emerson	×		
Allomyces arbuscula Butler	×		
Aplanes androgynus (Archer) Humphrey	×	×	×
Blastocladiopsis parva (Whiffen) Sparrow	×	×	
Calyptralegnia achlydoides (Coker et Couch) Coker			×
Dictvuchus sterilis Coker	×	×	×
Isoachlya anisospora (de Bary) Coker	×	×	~
Isoachlya monilifera (de Bary) Kauffman	×	×	×
Isoachlya toruloides Kaufinan et Coker	×		, A
Leptolegnia caudata de Bary		×	×
Leptomitus lacetus (Roth) Agardh	×	×	×
Protoachlya paradoxa (Coker) Coker	×	A .	
Pythiopsis cymosa de Bary	^	1	×
Pythium artotrogus de Bary	×.	×	^
Pythium ultimum Trow	×	~	
Saprolegnia australis Elliott	×	×	×
Saprolegnia diclina Humphrey	Î Â	^	×
Saprolegnia ferax (Gruith) Thurnet	×	×	
Saprolegnia mixta de Bery	^	^	× ×
Saprolegnia monoica Pringsheim		N N	×
Saprolegnia parasitica Coker	×	×	×
Saprolegnia shikotsuensis Hatai et al.	×	×	
Saprolegnia unispora (Coker et Couch) Seymour	×	×	
Thraustotheca clavata (de Bary) Humphrey			×
Zoopage phanera Drechsler	3	×	×
Candida albicans (Robin) Berkhout (conidial fungi)			×
Number of species	30	17	24

Aquatic fungi found on the eggs of acipenserid fishes in the different waters

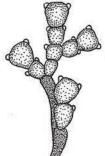


Achlya glomerata: gametangium (12-22 µm)

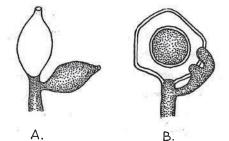


Achlya colorata: gametangium (17-38 µm)

Achlya megasperma: gametangium (36–62 µm)



Allomyces arbuscula: gametangium (14–24 µm) Isoachlya toluroides: gametangium (10–30 µm)

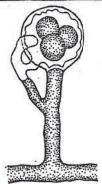


Pythiopsis cymosa: A. sporangium (20–25 µm) B. gametangium (20–30  $\mu$ m)

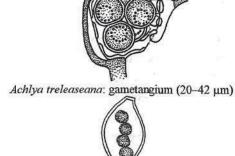


Saprolegnia unispora: gametangium (42-50 µm)

Fig. 1. Aquatic fungi new to fishes









#### DISCUSSION

The studies hither to conducted on the aqaatic fungi growing on the eggs of acipenserid fishes have revealed three species of Achlya (Achlya bisexualis, Achlya hypogyna, Achlya klebsiana), Leptolegnia caudata and five species of the genus Saprolegnia (Saprolegnia declina, Saprolegnia ferax, Saprolegnia mixta, Saprolegnia monoica and Saprolegnia parasitica). The remaining species mentioned in this paper should be treated as new to acipenserid fish.

The present study has revealed the fewest fungi on the eggs of *Huso huso* and *Acipenser güldenstädti* persicus. It should be stressed that Larceva (1986), while studying the growth of saprolegnia fungi on the eggs of three acipenserid species, observed a similar phenomenon. The authors observed only four aquatic fungus species on the eggs of *Huso huso*, while the eggs of *Acipenser güldenstädti* had seven and *Acipenser stellatus*—eleven species of saprolegnia fungi.

The most common fungus species found on the eggs of the acipenserid fishes studied include Leptomitus lacteus, found on the eggs of all the species, and Saprolegnia parasitica, growing on five, exept for the eggs of Acipenser stellatus. Saprolegnia shikotsuensis occurred on the eggs of four species. Leptomitus lacteus, commonly known as a sewage fungus, was found on perch specimens (Perca fluviatilis) in Windermere in England by Pickering and Willoughby (1977) and Willoughby and Roberts (1991). The growth of the eggs of this fungus was observed on eggs of European whitefish (Coregonus albula) and lavaret (Coregomus lavaretus) in the hatchery of Wegorzewo (Czeczuga and Woronowicz 1993). It should be noted that in five hatcheries investigated, Leptomitus lacteus was found to infect the eggs of European whitefish and lavaret only in Wegorzewo, and only there it occurred in the water of the Wegorapa River that supplies the hatchery (Czeczuga 1991a). Saprolegnia parasitica is one of the most common species observed on fishes in various latitudes (Neish and Hughes 1980; Srivastava 1980); on eggs (Scott and O'Bier 1962; Czeczuga and Woronowicz 1993) and on the young and full-grown individuals of various species (Florinskaja 1969, 1971; Osipian et al. 1988; Dudka et al. 1989; Hatai et al. 1990; Hatai and Hoshiai 1992). It was also found in acipenserid fishes (Larceva and Altufiev 1987; Larceva 1986; Larceva and Dudka 1990). Saprolegnia shikotsuensis was first described in 1974 from specimens of Oncorhynchus nerka var. adonis from Lake Shikotsu on Hokkaido in Japan (Hatai et al. 1977). The present study has revealed new to acipenserid fishes growth of fungi on the eggs. This group includes Achlya colorata, Achlya glomerata, Achlya megasperma, Aehlya treleaseana, Allomyces arbuscula, Isoachlya toruloides, Pythiopsis cymosa and Saprolegnia unispora (Fig. 1). The species of the genus Achlya, new to fishes, have been already observed in different types of water in northeastern Poland (Czeczuga 1991b, c, 1994a, b, 1995; Czeczuga and Woronowicz 1992). Up to now, these fungi have been presented as

phytosaprophytes, and only *Achlya megasperma*, is a zoosaprophyte included among keratynophylic species, usually found in water (Czeczuga and Muszyńska 1994). *Allomyces arbuscula* has been encountered as a soil and aquatic saprophyte (Batko 1975), and as a chitinophylic fungus growing on wing cases of insects (Czeczuga and Godlewska 1994), *Isoachlya toruloides* has been regarded as an aquatic saprophyte, while, *Pythiopsis cymosa* as a phytosaprophyte. *Saprolegnia unispore*, according to Seymour (1970) lives in water and wet soil.

*Candida albicans* is not frequently encountered in waters of north-eastern Poland (Czeczuga 1991a, b, c, 1994a, b). In fish *Candida albicans* was found to occur on the European whitefish eggs in Węgorzewo hatchery (Czeczuga and Woronowicz 1993) and on the eel fry monteé (Czeczuga 1994c).

The present study has proved that growth of the respective aquatic fungus species on fish eggs depends on the water body from which water has been collected for the experiment (Tab. 3). Most species developed on eggs in pond water, fewest in Lake Komosa. The chemical analysis of the water collected from these three water bodies found water differentiation with regard to the content of chemical compounds. Pond water had considerably more biogenic compounds, mainly phosphorus. The water from Lake Komosa was very poor in biogenes. This would confirm once again our earlier assumptions (Czeczuga and Woronowicz 1993) that the degree of infection of fish eggs in hatcheries depends largely on the state of cleanness and trophicity of water that supplies a given hatchery.

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### GRZYBY WODNE ROZWIJAJĄCE SIĘ NA IKRZE KILKU GATUNKÓW RYB JESIOTROWATYCH

#### STRESZCZENIE

Autorzy badali w warunkach laboratoryjnych występowanie grzybów wodnych na ikrze 6 taksonów ryb jesiotrowatych. Badaniami objęto ikrę Acipenser güldenstädti, Acipenser güldenstädti persicus, Acipenser nudiventris, Acipenser ruthenus, Acipenser stellatus oraz Huso huso. Do doświadczeń używano wody ze stawu, z jeziora i rzeki, uwzględniając w niej poszczególne parametry hydrochemiczne.

Ogólnie stwierdzono na ikrze rozwój 42 gatunków grzybów zoosporowych oraz 1 gatunek konidialny (*Candida albicans*). Najmniej gatunków rozwijało się na ikrze *Huso huso* (7), najwięcej na ikrze *Acipenser güldenstädti* (16). Wśród stwierdzonych gatunków grzybów 33 okazały się gatunkami nowymi dla ryb jesiotrowatych. Ponadto wśród nowych dla ryb jesiotrowatych grzybów 8 gatunków: *Achlya colorata, Achlya glomerata, Achlya megasperma, Achlya treleaseana, Allomyces arbuscula, Isoachlya toruloides, Pythiopsis cymosa oraz Saprolegnia unispora* okazało się gatunkami nowymi dla ryb w ogóle.

Woda ze stawu była najzasobniejsza w związki biogenne, zaś woda z jeziora Komosa zawierała ich najmniej. Toteż najwięcej gatunków grzybów rozwijało się na ikrze badanych ryb w wodzie ze stawu, najmniej zaś — w wodzie z jeziora.

Received: 30 October 1995

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