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**EXPERIMENTAL INFECTION OF THE BLACK-HEADED GULL
LARUS RIDIBUNDUS (L.) WITH EYE-FLUKES PARASITES OF THE
THREE-SPINED STICKLEBACK *GASTEROSTEUS ACULEATUS* (L.)**

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Abstract

Metacercariae of *Diplostomum* spp. have been detected in the eyes (lens, humour and under retina) of many species of fish. Adult forms occur in the intestine of piscivorous birds, mostly Laridae. Eight young black-headed gulls, *Larus ridibundus* (L.) were infected with eye-flukes from the eyes of three-spined stickleback, *Gasterosteus aculeatus* (L.): five with flukes from the eye-lenses and three from the vitreous humour of eyes. After few days birds were euthanised with an ether overdose and the intestines were studied. In the intestine of experimentally infected birds were found adult stages of *Diplostomum* sp.

Key words: *Diplostomum* sp., Gulf of Gdańsk, stickleback, seagull

INTRODUCTION

Eye-flukes of the genus *Diplostomum* von Nordmann, 1832 are widely dispersed in Holarctic region. Their life-cycles may involve two intermediate hosts and a definitive host: aquatic snails (first intermediate host), fish (second intermediate host) and piscivorous birds (definitive host) (Karvonen et al. 2006). Cercariae of *Diplostomum* spp. were detected in lymnaeid snails, *Lymnea stagnalis*, *L. peregra*, *L. auricularia* and *L. ovata* (cf. Lyholt and Buchmann 1995, Niewiadomska 1996, Niewiadomska et al. 1997, Valtonen and Gibson 1997, Väyrynen et al. 2000). Metacercariae occurred in the eyes (lenses, vitreous humour and retina) of many species of freshwater and brackish water fish (Leno and Holloway 1986, 1989, Höglund 1991, Chappell 1995, Niewiadomska 1996, 2003, Valtonen and Gibson 1997, Marcogliese et al. 2001). These metacercariae were known from 50 (Niewiadomska 1996) to 125 species of fish (Höglund 1991, McKeown and Irwin 1995). Infection with *Diplostomum* spp. associated with blindness, cataracts can lead to emaciation including mortality (Graczyk 1988, Chappell 1995, Grabda-Kazubska et al. 1996, Valtonen and Gibson

1997, Marcogliese et al. 2001, Niewiadomska 2003). Adult forms occur in the intestine of fish-feeding birds, mostly Laridae (Sitko 1968, Leno and Holloway 1986, Höglund and Thulin 1992, Valtonen and Gibson 1997, Rolbiecki et al. 1999, Marcogliese et al. 2001). *Apatemon gracilis* has similar life cycle which include snails (*Lymnea*), fish (perch and stickleback, sometimes Cyprinidae) and birds (Anatidae) hosts (Niewiadomska 2003).

The identification of many *Diplostomum* species is very difficult due to their high morphological similarity and variability in dependence of species or specimen of host and age or density of metacercariae (Graczyk 1991, 1992, Chappell 1995, Niewiadomska 1996, 2003, Niewiadomska and Laskowski 2002, Marcogliese et al. 2001).

Metacercariae of *Diplostomum* spp. were detected in many species of fish from the Gulf of Gdańsk (south Baltic Sea) (Rokicki and Morozińska 1994, Rolbiecki et al. 1999, Morozińska-Gogol 2006). The three-spined stickleback was one of the most infected fish.

In this study young black-headed gulls were used as the experimental definitive host for identification of metacercariae from the three-spined stickleback.

MATERIALS AND METHODS

Three-spined sticklebacks were caught with a dip-net in the Gdynia Yacht Port (Gulf of Gdańsk) in 1996 and examined at the Department of Invertebrate Zoology, University of Gdańsk. Metacercariae were collected in two samples: 1st – flukes from the eye-lenses (*Diplostomum* spp.), 2nd – from the vitreous humour (*Diplostomum* spp. and encysted *Apatemon gracilis*).

Ten juvenile (about one week old) black-headed gulls were taken out from the abandoned nests from the major colony on the Vistula Lagoon by ornitologist from the University of Gdańsk. Before the infection, the birds were starved for 24 hours. Gulls were fed with pieces of fish (herring). Five birds were fed with fish and a dose of approximately 50 metacercariae from lens, three birds with approximately 25 metacercariae from the vitreous humour containing both parasites (*Diplostomum* sp. and *A. gracilis*) and two gulls with pieces of herring without metacercariae. All birds were fed on fish filets in next days.

After 10 days gulls were euthanised with an ether overdose and examined. Parasites were removed and clean by delicate shaking in physiological saline and after that fixed in 70% alcohol. After dehydration in ethanol, specimens were cleared in creosote and mounted in Canada balsam.

Digeneans were measured. The metric data (length and width of body, forebody, hindbody, oral sucker, ventral sucker, pharynx, Brande's organ, ovary and testis) were given in micrometers in table 1. The identification based on the morphology of adults and on the metric data showed by Bychovskaja-Pavlovskaja (1962), Niewiadomska (1984, 1987), Sonin (1986) and Höglund and Thulin (1992).

RESULTS

Adult forms were detected in the gulls infected with metacercariae from the lenses and only a few specimens from the vitreous humour. A total of 98 *Diplostomum* spp. from gulls infected with eye flukes from the lens and 6 from the vitreous humour were collected. Adults of *Apatemon gracilis* were not found.

Based on the metric data and the morphology, 97 digeneans from birds infected with lens flukes and all from vitreous humour were very similar with *Diplostomum spathaceum* (Rudolphi, 1819). One specimen, from seagull infected with eye-fluke from the lens was different (Tab. 1). Parameters like length and width of body, forebody, hindbody, oral sucker, ventral sucker, pharynx, Brande's organ and hindbody to forebody ratio are similar with metric data shown by Sonin (1986) and Höglund and Thulin (1992) for *Diplostomum mergi* Dubois, (1932) but the metric dimensions of the ovary and the testis were different.

Table 1
The metric data (μm) of adults *Diplostomum* from the intestine of experimental infected black-headed gulls with metacercariae from the eyes of stickleback

Characters		Metric data mean (range)		
		alike <i>D. spathaceum</i>		alike <i>D. mergi</i>
Body	length	2523	(1540-3115)	706
Forebody	length	1079	(750-1510)	468
	width	523	(490-579)	349
Hindbody	length	1444	(1215-1890)	238
	width	349	(295-470)	206
Oral sucker	length	63	(55-107)	29
	width	60	(53-97)	31
Ventral sucker	length	87	(63-115)	70
	width	91	(70-143)	70
Pharynx	length	53	(43-75)	76
	width	45	(31-47)	45
Brande's organ	length	232	(180-255)	131
	width	266	(235-310)	131
Ovary	length	129	(87-156)	29
	width	129	(98-170)	16
Anterior testis	length	211	(185-240)	41
	width	232	(190-257)	43
Posterior testis	length	246	(200-283)	49
	width	278	(213-320)	53
Hind – to forebody ratio		1.34		0.51

DISCUSSION

Metacercariae of *Diplostomum* noted in the 20th century in Poland were identified as *D. spathaceum*, but this is a collective species and quite often determination are questionable (Niewiadomska 2003). *G. aculeatus* are known as a host of *D. gasterostei* (Höglund and Thulin 1992), *D. spathaceum* (Graczyk 1991), *D. pseudospathaceum* (Graczyk 1992) and *D. paracaudum* (Niewiadomska 1987). Except *D. gasterostei* (live at the retina) all mentioned above occurred in the lenses of fish (Höglund and Thulin 1992). All parasites from gulls infected with specimens from the lenses and five from the vitreous humour resembled *D. spathaceum*. It is possible that the same species were found in the lenses and vitreous humour, because of heavy infection, the lens could degenerate (Leno and Holloway 1989) and parasites got out from the lens or could be in the way to the lens. One specimen resembled *D. mergi*. Smaller measure of the ovary and testis of these specimens then mentioned by Sonin (1986) and Höglund and Thulin (1992), can be a result of the “young age”.

Metacercariae of *Apatemon* sp. occurred in freshwater fish (Williams and Jones 1994; Paperna 1995) and adults in birds (Vojtek 1972). Adults of *Apatemon gracilis* were not found, because of host specificity – occurred mostly in piscivorous Anatids and sporadically in Larids. Vojtek (1972) observed, that encysted metacercariae of *Apatemon* often were not invasive for definitive host.

CONCLUSIONS

To conclude, *D. spathaceum* is the most abundant parasite of the eyes of the three spined-stickleback from the Baltic Sea.

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EKSPERYMENTALNE ZARAŻENIE MEWY ŚMIESZKI *LARUS RIDIBUNDUS* (L.) PRZYWRAMI Z OKA CIERNIKA *GASTEROSTEUS ACULEATUS* (L.)

Streszczenie

Przedstawiona praca zawiera wyniki eksperymentalnego pozyskania postaci dorosłej przywr z rodzaju *Diplostomum*. Przywry z rodzaju *Diplostomum* są szeroko rozprzestrzenione na półkuli północnej. W ich cyklu życiowym występuje trzech żywicieli: ślimaki, głównie z rodzaju *Lymnea* (I żywiciel pośredni), ryby (II żywiciel pośredni) oraz ptaki rybożerne, gł. z rodzaju *Larus* (żywiciel ostateczny). Identyfikacja metacerkarii z oka ryb jest niezwykle trudna ze względu na ich podobieństwo oraz ogromną zmienność. Przywry żyją w oku wielu gatunków ryb słodkowodnych, występujących w wodach słonawych oraz ryb hodowlanych. Licznie występują u ryb z Zatoki Gdańskiej. Metacerkarie pozyskano z cierników złowionych w porcie jachtowym w Gdyni. Zebranymi metacerkariami zarażono młode mewy śmieszki. Ptakom podano kawałki ryb (śledzi) wraz z metacerkariami. W eksperymencie uzyskano postaci dorosłe przywr. Uzyskane osobniki dorosłe zostały zmierzone (tab. 1). Większość pomiarów odpowiadała danym literaturowym dla *D. spathaceum* i w jednym przypadku *D. mergi*. Jedynie narządy rozrodcze były mniejsze niż to wynika z danych literaturowych. Można próbować to powiązać np. z młodym wiekiem osobników. Wyniki wskazują, że *D. spathaceum* jest najliczniejszym pasożytem oka cierników z Bałtyku.