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Occurrence of Nematodes in the alimentary tract of great cormorants [*Phalacrocorax carbo* (Linnaeus, 1758)] in colonies located in the upper and lower Vistula River

by

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Abstract

The occurrence of nematodes in the alimentary tract of great cormorant *Phalacrocorax carbo* (L., 1758) nestlings was studied in the Włocławek Reservoir located on the Vistula in central Poland and in adult birds wintering in the south of Poland, with the food base in the upper Vistula. Mixed (max 4 species) nematode infections were found in all birds. Apart from Contracaecum rudolphii Hartwich, 1964 found in all studied birds, the thirdand fourth-stage larvae as well as adult individuals of the following species were also present: Eustrongylides excisus Jägerskiöld, 1909, Anisakis simplex (Rudolphi, 1809), Hysterothylacium aduncum (Rudolphi, 1802), Desmidocercella numidica Seurat, 1920, Synquaria squamata (Linstow, 1883), Cosmocephalus obvelatus (Creplin, 1825), Baruscapillaria carbonis (Dubinin et Dubinina, 1940). The overall mean intensity of infection with nematodes was 23.4 \pm 11.6 in nestlings and 175.7 \pm 110.9 in adult cormorants. The most common species was C. rudolphii with the mean infection intensity of 12.5 ± 5.9 in nestlings and 154.6 ± 111.4 in adults. The second most common species was E. excisus. S. squamata, C. obvelatus and D. numidica were less common in both age groups. A. simplex and H. aduncum were found only in nestlings, while B. carbonis only in adults.

Key words: great cormorant; parasite; Nematoda

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Introduction

Cormorants (Phalacrocorax carbo L., 1758) are migrating birds. They stay in Poland from April to October and migrate to the south of Europe for winter. They were observed in Tunisia, Egypt and Algeria (Mokwa et al. 2005; Bzoma 2011). Poland is also a resting site for cormorants flying from the north, mainly from Sweden and Denmark, as well as from the White Sea, the Barents Sea and the Sea of Azov. After a brief period of feeding, the birds fly to the south of Europe. Some cormorants, mainly those from the Baltic Sea basin and sporadically from northern Scandinavia and western Europe, spend winter in Poland, predominantly in the Szczecin Lagoon and the Gulf of Gdańsk and in the Vistula and Oder drainage basins (Bzoma 2011). The birds staying in the Vistula region feed mainly in dam reservoirs and smaller water bodies contiguous to the river (Wziątek 2015).

The effect of cormorants on aquatic ecosystems is considered in two aspects: the impact of birds on fisheries and the role they play in the biocenosis. Cormorants are typical ichthyophages that feed on many different fish species. Due to their top position in the trophic chain in aquatic ecosystems, cormorants are particularly exposed to contact with the larval stages of parasites occurring in fish. Many of them develop and mature in cormorants. A large portion of cormorant feces, containing different parasites at different development stages, are dropped into the water where parasites meet the intermediate or final hosts. Migrations of cormorants from their wintering areas to the nesting grounds are of great importance to the transmission of parasites, and the colonization of new ecosystems by parasites is favored, for example, by climate changes, including mainly an increase in average annual temperature (Marcogliese 2008; Klimpel, Palm 2011). The nesting success of cormorants depends on the number of eggs laid, on the age of parents and their experience, as well as on the food abundance, the presence of predators and weather conditions (Bzoma 2011). Females lay 3 to 6 eggs in 2-3-day intervals. The incubation lasts 23-24 days. The cormorant is a typical altricial species - when being fed, the young reach deep into the parent's throat from where they collect partly digested food, which can result in transferring the parasites living in the alimentary tract of adult birds. Cormorants are capable of flying about 8-9 weeks after hatching, but they are still fed by their parents for another 2-4 weeks. This way of acquiring the nematode Contracaecum has been described for very young cormorants (Kuiken et al. 1999) and according to the authors, it may be responsible for the high mortality rate in this age

group. Cormorants lay more eggs than they can raise young. Dead young cormorants are sometimes found in colonies; some of them fell out of a nest, others – the weakest ones – were thrown out by their parents.

Contracaecum rudolphii Hartwich, 1964 is the most common nematode in the cormorant alimentary tract (Abollo et al. 2001; Torres et al. 2005; Dziekońska-Rynko, Rokicki 2008; Kanarek 2011; Švažas et al. 2011; Monteiro et al. 2011). Adult individuals and the third- and fourth-stage larvae inhabit mainly the stomach, but they are also found in the esophagus, the duodenum and in farther parts of the small intestine. Nematodes from the genera *Syncuaria, Eustrongylides, Baruscapillaria, Cosmocephalus, Cyathostoma* occur much less frequently (Frantová 2001; Kanarek, Rolbiecki 2006a; Monteiro et al. 2006, 2011; Kanarek 2009; Švažas et al. 2011; Kanarek, Zaleśny 2014).

Apart from individual features, such as the age, sex or resistance, the composition of the parasite fauna is affected by environmental factors, such as season and the food base (Monteiro et al. 2011; Kanarek, Zaleśny 2014). The majority of studies on parasites that infect cormorants in Poland were conducted in summer and they concern adult birds living on the Masurian Lakes or the Vistula Lagoon (Kanarek, Rolbiecki 2006b; Dziekońska-Rynko, Rokicki 2008). There are no reports on parasites of young cormorants, birds wintering in Poland and/or individuals dwelling close to lotic waters. The aim of the presented study was to investigate the parasites of the alimentary tract of young cormorants collected in spring in the vicinity of the Włocławek Reservoir on the Vistula River in central Poland, as well as adult birds collected in winter on the upper Vistula in the south of Poland.

Materials and methods

The parasitological study was conducted on 20 dead, young cormorants (nestlings = pulli and fledglings) collected in May 2013 under the nests in a colony situated on the island close to the right bank of the Włocławek Reservoir, near the village of Murzynowo (52°35'09"N, 19°30'27"E) and 20 adult birds feeding on the upper Vistula near Tyniec (50°01'09"N 19°48'02"E), obtained in February 2014. The shooting of birds was conducted in accordance with the permission of the Regional Director for Environmental Protection in Kraków (number OP-64.01.30.2014). The alimentary tract was divided into sections (esophagus, stomach, intestine) and placed in separate dishes. The sections were then cut up lengthwise and decanted with 0.9% sodium chloride solution NaCl. Parasites were identified according to Baruš et al. (1978),



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Moravec et al. (1988), Okulewicz (1989), Moravec and Scholz (1994), Grabda-Kazubska and Okulewicz (2005), Frantová (2001), Monteiro et al. (2006), Kanarek and Rolbiecki (2006a) immediately after decantation or after being fixed in 70% ethanol with an addition of glycerol (5%).

<u>Results</u>

Mixed (max. 4-species) nematode infections were found in all studied birds (Table 1). Apart from C. rudolphii found in all birds, the third- and fourth-stage larvae and adult individuals of the following species were also found: Eustrongylides excisus Jägerskiöld, 1909, Anisakis simplex (Rudolphi, 1809), Hysterothylacium aduncum (Rudolphi, 1802), Desmidocercella numidica Seurat, 1920, Synguaria squamata (Linstow, 1883), Cosmocephalus obvelatus (Creplin, 1825), Baruscapillaria carbonis (Dubinin et Dubinina, 1940). The overall intensity of infection with nematodes was 23.4 ± 11.6 (range 10-45) in nestlings and 175.7 \pm 110.9 (range 81-565) in adult cormorants. The most common species, C. rudolphii, constituted 53.2% of all nematodes in the alimentary tract of nestlings and 87.9% in adult individuals. The mean intensity of infection was 12.5 ± 5.9 in nestlings and 154.6 ± 111.4 in adults; mature nematodes dominated in both age groups with 59.8% and 51.9% contribution, respectively. Females C. rudolphii prevailed in young and mature birds and constituted 37.7% and 35.0% of all nematodes, respectively. The second most common nematode was *E. excisus* with the prevalence of 75% in nestlings and 80% in adults, and the mean infection intensity of 4.2 ± 3.9 and 7.0 ± 6.3 , respectively. *S. squamata* was found in 65% of the studied birds. *C. obvelatus* and *D. numidica* were less common in cormorants in both age groups. *A. simplex* and *H. aduncum* were found only in nestlings, while *B. carbonis* was found only in adult birds (Table 1).

Discussion

One of the factors determining the susceptibility to parasites is the host's age. During their intensive growth, young birds consume large amounts of food, much larger than adult birds, which makes the parasite infection more likely, and the lack of fully developed immunity favors the increased intensity of infection (Sitko 1993). Apart from individual features of the host, species diversity of parasite fauna is greatly affected by environmental factors. The acquisition of new parasites by cormorants is facilitated by migration to different climate zones and changing the food base within a year. The community of metazoan parasites in cormorants that fed in the brackish waters of the Vistula Lagoon was clearly more heterogeneous (30 species) than in birds that fed in the Masurian Lakes (18 species). The number of nematode species also depends on the season and age of birds (Kanarek, Zaleśny 2014).

Table 1

Nematodes in great cormorants (*Phalacrocorax carbo*) from colonies in the area of the lower (Murzynowo) and upper (Tyniec) Vistula River. P – prevalence, MI – mean intensity, RI – range of intensity, F – female, M – male, L – third- and fourth-stage larvae, SD – standard deviation

	Nestlings from Murzynowo					Adults from Tyniec				
Nematoda	Р	MI ± SD (RI)	Contribution (%)			Р	MI ± SD	Contribution (%)		
	(%)		F	М	L	(%)	(RI)	F	М	L
Contracaecum rudolphii	100	12.5 ± 5.9 (2-22)	37.7	22.1	40.2	100	154.6 ± 111.4 (39-545)	35.0	16.9	48.1
Anisakis simplex	20	0.7 ± 1.3 (0-4)	0.0	0.0	100	0.0	0.0	0.0	0.0	0,0
Hysterothylacium aduncum	30	0.9 ± 1.5 (0-5)	0.0	0.0	100	0.0	0.0	0.0	0.0	0.0
Cosmocephalus obvelatus	30	1.2 ± 2.1 (0-6)	0.0	0.0	100	30	1.3 ± 2.5 (0-7)	0.0	0.0	100
Syncuaria squamata	65	2.3 ± 2.0 (0-6)	80.4	19.6	0.0	65	6.2 ± 7.6 (0-28)	78.4	21.6	0.0
Eustrongylides excisus	75	4.2 ± 3.9 (0-16)	59.5	40.5	0.0	80	7.0 ± 6.3 (0-20)	85.7	14.3	0.0
Baruscapillaria carbonis	0.0	0.0	0.0	0.0	0.0	60	5.3 ± 8.6 (0-34)	79.4	20.6	0.0
Desmidocercella numidica	45	1.7 ± 2.3	0.0	0.0	100	25	1.2 ± 2.4	0.0	0.0	100





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To date, parasitological studies of cormorants have been conducted before or after the nesting season, on birds feeding in lentic waters. The present study focuses on birds of different age, feeding in the upper and lower sections of the Vistula River, examined in spring or in winter.

C. rudolphii occurred in stomachs of all studied cormorants, with the intensity of infection much lower in nestlings than in adult birds (12.5 and 154.6, respectively). Mature nematodes dominated in the stomachs of nestlings, which confirms that parasites can be transferred from adult to young birds with food. This way of acquiring *Contracaecum* nematodes was described by Kuiken et al. (1999) for very young cormorants in a colony at Lake Dore (Saskatchewan, Canada). The prevalence increased with age and reached 50%, 71% and 83% in one-, two- and three-week-old birds, respectively. According to the authors, infection with these nematodes is the cause of high mortality in the above-mentioned age groups.

Dziekońska-Rynko and Rokicki (2008) found mainly the 3rd and 4th stage larvae in young birds, the same way as in adults examined in the summer. The results of this study are different - mature nematodes dominated both in young and adult cormorants, which can be attributed to the study period, i.e. winter or early spring. At that time, the food amount may be limited and the foraging intensity may be significantly reduced, thus the number of nematode larvae transmitted directly from food (infected fish) can also be limited. Similar results were obtained for adult birds from Lake Selment Wielki before the nesting season, when mature nematodes of C. rudolphii dominated (Dziekońska-Rynko, Rokicki 2008). The pathogenicity of this nematode is low, but it can digest the gastrointestinal mucosa of the host, with damage leading to digestion and absorption disorders which - combined with other stressors, such as bacteria or xenobiotics may cause the birds to die (Abollo et al. 2001; Rokicki et al. 2011). With extremely high intensity of infection (max 545) detected in adult birds from the Vistula River near Tyniec, one can expect negative effects on their health.

The stomachs of young birds under study were also found to contain other nematodes of the *Anisakidae* family, such as the third stage larvae of *A. simplex* and *H. aduncum*. The final hosts of *A. simplex* are marine mammals, whereas of *H. aduncum* –predatory fish, both saltwater and freshwater ones. These larvae occur quite frequently in fish in the Vistula Lagoon and in the Baltic Sea (Rolbiecki, Rokicki 2000; 2002; Rolbiecki 2006; Rolbiecki, Rokicki 2008). They were also found in cormorants in the Vistula Spit (Kanarek, Rolbiecki 2006b). The presence of larvae of those nematodes in cormorants in the colony in Murzynowo may indicate that the birds feed not only in the Vistula, but also in the Baltic or in the Vistula Lagoon. No wonder that larvae of *A. simplex* or *H. aduncum* were not found in birds caught in the south of Poland, near Tyniec.

E. excisus was the second most frequent nematode, found in 75% of the young and in 80% of the adult birds. The fourth-stage larvae are noted in different benthic fish (intermediate host II) infected by eating oligochaetes (intermediate host I) with the secondand third-stage larvae. Predatory fish can acquire the parasite with their fish prey and become a paratenic host (Bjelić-Čabrilo et al. 2013). Larvae of E. excisus have been found both in freshwater and marine fish (Rolbiecki et al. 1999; Rolbiecki 2003; Mierzejewska et al. 2012), while the mature nematodes have been commonly reported in fish-eating birds (Kanarek, Rokicki 2005; Švažas et al. 2011; Kanarek, Zaleśny 2014). The nematode is highly pathogenic for fish as well as for fish-eating birds. High intensity of infection can be lethal, especially for young individuals. This nematode is also potentially pathogenic for humans (Coler 2009; Bielić-Čabrilo et al. 2013) and reports on its presence in natural environment are of particular interest.

S. squamata is a cosmopolitan parasite of cormorants, found in these birds in the Czech Republic, Russia and Poland (Moravec, Scholz 1994; Kanarek, Rolbiecki 2006a; Švažas et.al. 2011). In Poland, the first report on this species in cormorants comes from 2000-2001 (Kanarek, Rolbiecki 2006a). The prevalence in the Vistula Spit exceeded 80% (87% in nestlings). The highest intensity of infection was determined in fledglings chicks, even though the nematode was not found in adult birds from this colony. On the other hand, in the Masurian Lakes, this species was found only in adult birds (Kanarek, Rolbiecki 2006a). The prevalence of S. squamata in cormorants in the Curonian Spit was also high (100%), with the mean intensity of 38.5 (Švažas et al. 2011). In our study, the prevalence in nestlings and adult cormorants was guite similar, whereas the intensity of infection in young birds was three times lower than in adults (Table 1).

The nematode *B. carbonis* was found only in adult birds from the colony in the upper Vistula. The species was first found in Poland in cormorants from the Stawy Milickie Nature Reserve in 1988 (Okulewicz 1989) and then (in 2001-2005) in cormorants from the Vistula Lagoon and Lake Selment Wielki (Kanarek, Zaleśny 2014). This nematode is usually found in adult cormorants and its pathogenicity has not been precisely determined (Frantová 2001).

C. obvelatus and *D. numidica*, cosmopolitan species – parasites of many fish-eating birds – are found sporadically in cormorants (Baruš et al. 1978).



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Previously D. numidica was found in Poland in grey heron Ardea cinerea Linnaeus, 1758 in Podlasie and in the Lublin Uplands (Bezubik 1956) and then (2001-2005) in cormorants from the Vistula Split, the Vistula Lagoon and the Masurian Lake District (Kanarek, Rokicki 2005). In our study, the prevalence in nestlings from the lower Vistula was 45%, in adult birds from the upper Vistula - 25%, with quite low intensity of infection: mean 1.7 and 1.2, respectively. Data on adult cormorants from the Curonian Spit were comparable: the prevalence exceeded 70%, while the mean intensity of infection was 2.1 (Švažas et al. 2011). There are few reports in the available literature on the occurrence of these nematodes in fish in Poland. The third-stage larvae of D. numidica were found in twaite shad, Alosa fallax (Lacepéde, 1803) from the southern Baltic Sea (Rokicki at al. 2009), in roach Rutilus rutilus (Linnaeus, 1758) from Lake Żarnowiec and in the European perch Perca fluviatilis Linnaeus, 1758 from Lake Raduńskie Dolne (Rolbiecki 2010). Whereas the third-stage larvae of C. obvelatus were found only in the European perch from Lake Raduńskie Dolne (Rolbiecki 2010).

The results indicate that the species diversity of nematodes in the alimentary tract of cormorants depends on the age of birds and their food base. The presented study on birds with the main foraging areas in the Vistula River basin revealed smaller heterogeneity of the nematode community compared to birds with the main foraging areas in the Masurian Lakeland and the Vistula Lagoon (Kanarek, Zaleśny 2014). This may be due to less complex and less abundant assemblage of potential intermediate hosts in lotic ecosystems compared to lentic ones. Moreover, it may also be connected with the sampling season – in winter and early spring, when the foraging intensity of birds is lower, the transmission of parasites via food is also significantly limited.

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