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Intestinal parasites in five species of the genus *Acrocephalus*

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Abstract. The occurrence of intestinal parasites was studies in adults and nestlings of *Acrocephalus paludicola*, *A. schoenobaenus*, *A. palustris*, *A. scirpaceus* and *A. arundinaceus*. The most common taxa in all these species were Coccidia and Ascaridia, and in some species also *Ornithostrongylus* and other Nematoda. In *A. arundinaceus* parasitized males were significantly heavier than nonparasitized ones, whereas in females the opposite was found. Moreover parasite-free males *A. schoenobaenus* and *A. scirpaceus* and females *A. paludicola* tended to be lighter compared to parasitized ones, but the difference was not significant. Infected *A. scirpaceus* females had significantly larger fat deposits than noninfected ones. Parasite prevalence also varied significantly between the promiscuous *A. paludicola* and the related monogamous species.

Key words: genus *Acrocephalus*, intestinal parasites, mating systems, body weight, fat deposit

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INTRODUCTION

Intestinal parasites of passerine birds, including representatives of the genus *Acrocephalus*, have been relatively frequently studied (Frank 1980, 1987, Sulgostowska & Czaplińska 1987, Czapliński et al. 1992). However, almost all these studies were conducted on dead individuals. Results of these studies were restricted to listing the parasite species, but no effort has been made to determine the influence of parasites on their hosts.

This study was carried out on five species of the genus *Acrocephalus* sympatrically inhabiting Central Europe: Aquatic Warbler *A. paludicola*, Sedge Warbler *A. schoenobaenus*, Marsh Warbler *A. palustris*, Reed Warbler *A. scirpaceus* and Great Reed Warbler *A. arundinaceus*. Their breeding biology has been relatively well studied (Koskimies 1991, Leisler 1991, Schulze-Hagen 1991a,b,c). Within this small group there is considerable variation in mating systems (Leisler & Catchpole 1991), from strict monogamy in Reed Warbler to specific form of promiscuity in Aquatic Warbler.

According to Hamilton & Zuk hypothesis (1982) bird females assess the genetic quality of males on the basis of their resistance to endoparasites. Resistance is correlated with certain secondary sex characters, such as feathers or comb, which are less developed in parasitized individuals. The authors suggest that these traits function to inform females about genetic resistance of a male to endoparasites, and hence they reflect current physical condition. Across species, the degree of ornamentation should be positively correlated with parasite prevalences. In dull-coloured species, the secondary sex character correlated with the parasite load may be song complexity (Zuk 1984), which should be related to male condition measured by body weight and fat deposit. Such a relationship was found in Sedge Warbler: males infected with blood parasites had significantly lower repertoire sizes compared to nonparasitized males (Buchanan et al. 1999). Representatives of genus *Acrocephalus* are dull-coloured and do not have elaborated ornaments, but their song is relatively complex. Song complexity varies among the species: the simplest songs have Reed Warbler and Aquatic Warbler, and the most variable and complicated one is Marsh Warbler song.

This study aimed to determine whether variability in mating systems corresponds with variability in prevalence of parasites of the digestive system.

STUDY AREA AND METHODS

Field study was carried out in May-July during the breeding seasons 1996–1998 in three study areas:
1) The South Basin of the Biebrza River (NW Poland, 53°20′N, 22°40′E), where Aquatic and Sedge Warblers were studied. The area was an open sedge meadow with the dispersed small *Salix* spp. bushes. Dry parts were covered with some patches of reedbed as well as small trees of alder *Alnus glutinosa* and willow;
2) Milicz ponds (51°33′N, 17°23′E) ca. 60 km N of Wrocław, where Reed and Great Reed Warbler were studied. Study plots were situated in reedbeds on fish-ponds near Stawno village. The ponds are extensive (20–283 ha) and resemble natural eutrophic lakes. Some of them are within the nature reserve;
3) water-bearing areas near Wrocław, where Marsh Warbler was studied. The species breeds here in high and dense herbaceous vegetation covering the areas between drinking water reservoirs with concrete walls.

Nests of the studied species were searched for in the field and faeces of nestlings were collected, the exception was Reed Warbler whose nestling faeces were not gathered. Adult birds were caught in nylon nets at their nests, as well as rows of nets standing on dikes within breeding sites. In order to collect faeces the caught birds were kept for a few minutes in cotton bags with the bottom made of a wire net. Each faecal bag was stored separately in a receptacle with 4% dichromic potassium solution. Birds were sexed on the basis of brood patch, present only in females. Fat deposit (range 0–5) was estimated with the method used in studies of bird migration (Busse 1970). Birds were weighed to the nearest 0.25 g with a Pesola spring balance, and their wing length was measured to the nearest 0.5 mm (Svensson 1992). In nestlings only fat deposit was estimated. The collected faeces were taken to the lab, where slides were prepared and subsequently examined at × 100–400 magnification to determine the presence of parasites. Parasites were generally classified to the genus level, or to the higher level when identification of found eggs posed difficulties. Apart from that, the number of oocysts or eggs in faecal samples was also estimated using the five grades scale: 1 — absence of oocysts/eggs, 2 — single oocysts/eggs in a slide, 3 — single oocysts/eggs within a field of view, 4 — 5–10 oocysts/eggs within a field of view, 5 — more than 10 oocysts/eggs within a field of view.

Faeces analysis do not allow for clear assessment of parasite load. The absence of eggs in the faeces may have various reasons, such as collection of faeces before maturation of parasite stage, or uneven distribution of eggs in faeces. On the other hand, all species were studied with the same method (e.g. the same time of day and year) and relatively large samples of individuals were analysed. Hence, we think that the mentioned factor should not have biased our results.

In statistical analysis we used chi-square test of independence (with Yates' correction for tables with one degree of freedom), Mann-Whitney U-test (Łomnicki 1995), and Kruskal-Wallis test (Fowler et al. 1998). If the same data were cross-tested several times, the Bonferroni correction was applied to judge the significance of p-values (Sokal & Rohlf 1995). P-values, which remained significant after Bonferroni's correction, were marked with asterics (e.g. p* = 0.01).

RESULTS

Proportion of various groups of parasites

The classified parasites belonged to six taxonomic groups. In all studied bird species, both adults and nestlings, dominated Coccidia and Ascaridia (Table 1 and 2). In adult individuals *Ornithostrongylus* were also abundant, whereas representatives of Nematoda were common in nestlings. Other group of parasites (Trematoda, Cestoda, Nematoda in adults, *Ornithostrongylus* in nestlings and other taxonomic groups) constituted less than 10% of all collected items.

Proportion of various parasites of digestive system found in faeces was similar in adults of Sedge Warbler, Reed Warbler and Great Reed Warbler ($\chi^2 = 6.755$, df = 6, ns). Statistically significant differ-

Table 1. Percentage distribution of various parasite taxa in adult birds. *Ornith.* — *Ornithostrongylus*.

Bird species	Coccidia	Ascaridia	*Ornith.*	Others
A. paludicola (n = 44)	38.6	31.8	–	29.5
A. schoenobaenus (n = 32)	75.0	12.5	6.2	6.2
A. palustris (n = 28)	21.4	60.7	10.7	7.2
A. scirpaceus (n = 27)	55.6	18.5	25.9	–
A. arundinaceus (n = 49)	63.3	16.3	14.3	6.1

Table 2. Percentage distribution of various intestinal parasite taxa in nestlings. Nemat. — other Nematoda.

Bird species	Coccidia	Ascaridia	Nemat.	Others
A. paludicola (n=140)	41.4	15.7	35.7	7.2
A. schoenobaenus (n=25)	36.0	48.0	16.0	–
A. palustris (n=16)	25.0	43.7	25.0	6.2
A. arundinaceus (n=37)	43.2	27.0	16.2	13.5

ences in composition of digestive system parasites were found between Aquatic Warbler and Marsh Warbler ($\chi^2 = 13.741$, df = 3, p* = 0.003), Marsh Warbler and Reed Warbler ($\chi^2 = 13.989$, df = 3, p* = 0.003), and Marsh Warbler and Sedge Warbler ($\chi^2 = 15.303$, df = 3, p* = 0.002). In nestlings (Table 2) similar parasite composition was found between Sedge Warbler, Marsh Warbler and Great Reed Warbler ($\chi^2 = 7.167$, df = 6, ns), and between Aquatic Warbler, Marsh Warbler and Great Reed Warbler ($\chi^2 = 12.996$, df = 6, ns). Significant difference was detected only between Sedge Warbler and Aquatic Warbler ($\chi^2 = 15.189$, df = 3, p* = 0.002).

Effect of parasites on bird body weight

In two studied bird species mean body weight of males and females differed significantly (Reed Warbler — males: $\bar{x} = 12.76 \pm 1.321$ g, n = 19; females: $\bar{x} = 11.97 \pm 0.527$ g, n = 30; Mann Whitney U-test, p = 0.015; Great Reed Warbler — males: x = 34.10 ± 1.570 g, n = 47; females: $\bar{x} = 29.86 \pm 2.215$ g, n = 11; Mann Whitney U-test, p < 0.0001). Hence the influence of parasites on body weight were analysed separately for males and females (Table 3). Significant differences were found only in Great Reed Warbler: parasitized males were on average heavier that nonparasitized ones; in females the opposite was found (Table 3). Similar tendency was found in male Sedge Warblers and Reed Warblers, and female Aquatic Warblers: parasitized individuals had higher body weight compared to healthy ones, but the differences were not significant (Table 3).

Effect of parasites on fat deposit

Males-females differences in fat deposit were significant in the Aquatic Warbler (Dyrcz 1993), Sedge Warbler (females: $\bar{x}=1.17 \pm 1.043$, n=18; males: $\bar{x}=0.61 \pm 0.761$, n=31; Mann-Whitney U-test, p = 0.05), and Reed Warbler (females: $\bar{x} = 1.47 \pm 0.513$, n = 19; males: $\bar{x} = 0.73 \pm 0.583$, n = 30; p = 0.0001). Therefore, also in this case both sexes were analysed separately. Parasitized

Table 3. Mean body weight (g) and the presence of intestinal parasites in adult males and females of four studied species (statistical significance: Mann-Whitney U-test).

Bird species	Parasites absent			Parasites present			p
	$\bar{x}$	SD	n	$\bar{x}$	SD	n	
Males							
A. paludicola	12.29	0.68	28	12.27	0.58	20	ns
A. schoenobaenus	12.36	0.72	16	12.79	0.59	14	ns
A. scirpaceus	11.82	0.46	17	12.17	0.56	13	0.055
A. arundinaceus	33.46	1.60	17	34.46	1.46	30	0.05
Females							
A. paludicola	11.92	0.625	12	12.59	1.002	11	ns
A. schoenobaenus	12.64	0.936	9	13.07	1.264	10	ns
A. scirpaceus	12.55	1.544	11	13.06	0.952	8	ns
A. arundinaceus	31.5	1.291	4	28.93	2.130	7	0.05

Table 4. Mean fat deposit expressed as a median (Q_1–Q_3 interquartile ranges) and the presence of intestinal parasites in adult females of four studied species (statistical significance: Mann-Whitney U-test).

Bird species	Parasites absent			Parasites present			p
	median	(Q_1–Q_3)	n	median	(Q_1–Q_3)	n	
A. paludicola	1	(0.5–1.5)	12	1	(1–2)	11	ns
A. schoenobaenus	1	(0.5–1.5)	8	1	(0.5–1.5)	10	ns
A. scirpaceus	1	(1–1)	11	2	(1.5–2)	8	0.05
A. arundinaceus	0.5	(1.5–2)	4	1	(1–1)	6	ns

Reed Warbler females had on average larger fat deposit that nonparasitized ones (Table 4). However, such a relationship was not found in other species, either in adults or nestlings.

DISCUSSION

Despite close relationship between the studied bird species, (Leisler et al. 1997) they are separated ecologically by occupying different habitats from reedbeds extending into deep water neighbouring an open water surface (Great Reed Warbler), to herbaceous and ruderal vegetation growing on dry land (Marsh Warbler). One may expect that taxonomic composition of intestinal parasites is associated to some degree with type of habitat. The three species (Great Reed Warbler, Reed Warbler and Sedge Warbler) whose parasite composition was similar inhabit flooded areas. In contrast, Aquatic Warbler and Marsh Warbler, whose parasite composition was different, may nest in dry areas. Monogamy is the main form of mating system in Marsh Warbler with low incidence of polygyny recorded in some populations (Dowsett-Lemaire 1981). Aquatic Warbler is a promiscuous species (Heise 1970, Schulze-Hagen 1991, Dyrcz & Zdunek 1993, Schulze-Hagen et al. 1993). These differences may explain the variability in parasite composition between Marsh Warbler and Aquatic Warbler, and between these species and other studied ones. It should also be noticed that Marsh Warbler song is most complex among the species of the genus *Acrocephalus*, and the song of Aquatic Warbler is one of the simplest. A relationship between song complexity and parasitofauna requires however further studies, considering individual differences.

A comparison of the two closely related species studied within the same study area, Aquatic Warbler (promiscuous species) and Sedge Warbler (predominantly monogamous species with some proportion of polygynous males) seems to support a relationship between the incidence of parasites and mating system. This study revealed that the two species belonged to two separate homogenous groups considering their parasitofauna composition. The data suggest that invasions of intestinal nematodes are much more frequent in promiscuous Aquatic Warbler compared to monogamous Sedge Warbler (Table 1). In the case of Coccidia the opposite relationship was found: infections were more frequent in the Sedge Warbler. However, coccideans are much less pathogenic than nematodes in adult birds; e.g. even 100% of adult passerines of some bird species (e.g. House Sparrow and Tree Sparrow, Kruszewicz 1995) may be parasitized with Coccidia.

The positive relationship between body weight, fat deposit and parasite load, found in this study, is difficult to interpret. From ecological point of view a bird organism may be treated as a parasite environment, hence well nourished organism may be able to sustain more parasites. The necessary condition is low pathogenicity of the parasite resulting in a stable parasite-host association. In such a case the presence of a parasite should not affect condition but rather e.g. song intensity, sexual selection, reproductive success or offspring condition (through feeding frequency).

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STRESZCZENIE

[Występowanie pasożytów przewodu pokarmowego u pięciu gatunków ptaków z rodzaju *Acrocephalus*]

Badaniami objęto: wodniczkę, rokitniczkę, łozówkę, trzcinniczka i trzciniaka. Prowadzono je w latach 1996–1998 w Kotlinie Biebrzańskiej (wodniczka i rokitniczka), na stawach rybnych w pobliżu Milicza (trzciniak, trzcinniczek i rokitniczka), oraz na terenach wodonośnych pod Wrocławiem (łozówka). Kał od piskląt pobierano w gniazdach, a ptaki dorosłe chwytano w sieci stylonowe w ich terytoriach lęgowych. Oznaczano też płeć na podstawie plamy lęgowej, oceniano otłuszczenie w skali 0–5 (Busse 1970), ważono i mierzono skrzydło. U piskląt oceniano tylko otłuszczenie.

Stopień zarażenia ptaków pasożytami badano przyżyciowo na podstawie analizy kału. Zebrany kał, umieszczony w roztworze dwuchromianu potasu, był przewożony do laboratorium, gdzie wykonywano preparaty bezpośrednie, badane mikroskopowo przy powiększeniu 100–400-krotnym. Pasożyty oznaczano co do rodzaju, a w przypadku braku możliwości bliższego rozpoznawania jaj — do wyższej jednostki systematycznej. Jednocześnie oceniano liczebność oocyst lub jaj w próbkach kału, stosując 5-stopniową skalę: brak, pojedyncze w preparacie, pojedyncze w polu widzenia, 5–10 sztuk w polu widzenia, powyżej 10 w polu widzenia.

Pasożyty zaliczono do sześciu grup systematycznych. U wszystkich badanych gatunków, zarówno ptaków dorosłych, jak i piskląt, dominowały Coccidia i Ascaridia. U ptaków dorosłych występowały też liczne *Ornithostrongylus*, natomiast u piskląt Nematoda. Pozostałe grupy pasożytów (Trematoda, Cestoda, Nematoda u dorosłych, *Ornithostrongylus* u piskląt oraz nieliczne pasożyty z innych grup systematycznych) stanowiły mniej niż 10 % ogółu stwierdzonych.

Wśród ptaków dorosłych rokitniczka, trzcinniczek i trzciniak miały podobny udział ($\chi^2 = 6.755$, df = 6, ns) różnych pasożytów przewodu pokarmowego w kale (Tab. 1). Istotne różnice w składzie zespołu pasożytów przewodu pokarmowego stwierdzono między wodniczką a łozówką ($\chi^2 = 13.741$, df = 3, p = 0.003), łozówką a trzcinniczkiem ($\chi^2 = 13.989$, df = 3, p = 0.003) i łozówką a rokitniczką ($\chi^2 = 15.303$, df = 3, p = 0.002). U piskląt (Tab. 2) grupę o podobnym składzie pasożytów stanowiły zarówno rokitniczka, łozówka i trzciniak ($\chi^2 = 7.167$, df = 6, ns), jak i wodniczka, łozówka i trzciniak ($\chi^2 = 12.996$, df = 6, ns), a istotna statystycznie różnica wystąpiła tylko między rokitniczką i wodniczką ($\chi^2 = 15.189$, df = 3, p = 0.002).

U dwóch gatunków stwierdzono istotne statystycznie różnice między średnim ciężarem samic i samców: trzcinniczek samce $\bar{x} = 12.76$ g ± 1.32 g, n = 19; samice $\bar{x} = 11.97$ g ± 0.53 g, n = 30 (U-test Manna-Whitneya, p = 0.015). Trzciniak: samce $\bar{x} = 34.10$ g ± 1.57 g, n = 47; samice $\bar{x} = 29.86$ g ± 2.21 g, n = 11, (U-test Manna-Whitneya, $p < 0.0001$). Dlatego wpływ pasożytów na ciężar ciała badano oddzielnie dla samców i samic (Tab. 3 i 4). Istotne różnice stwierdzono tylko u trzciniaka, u którego zarażone samce były przeciętnie cięższe niż wolne od pasożytów, podczas gdy u samic było odwrotnie (Tab. 3 i 4). Tendencja do przeciętnie większej masy ptaków zarażonych w porównaniu z ptakami nie zarażonymi zaznaczyła się także u samców rokitniczki i trzcinniczka oraz samic wodniczki (Tab. 3 i 4).

Różnice w otłuszczeniu między samicami i samcami były istotne u wodniczki (Dyrcz 1993), rokitniczki (samice: $\bar{x} = 1.17 \pm 1.043$, $n = 18$; samce: $\bar{x} = 0.61 \pm 0.761$, $n = 31$; $p = 0.05$, U-test Manna-Whitneya) i trzcinniczka (samice: $\bar{x} = 1.47 \pm 0.513$, $n = 19$; samce $\bar{x} = 0.73 \pm 0.583$, $n = 30$; $p = 0.0001$). A zatem i w tym przypadku rozpatrywano obie płcie oddzielnie. Zarażone samice trzcinniczka miały przeciętnie wyższe zapasy tłuszczu niż nie zarażone (Tab. 5). Takiej zależności nie stwierdzono jednak u pozostałych gatunków, a także w przypadku samców i piskląt wszystkich badanych ptaków.

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