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A Sarcocystis Species from Goshawk (Accipiter gentilis) with Great Tit (Parus major) as Intermediate Host

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Summary. One hundred and seventy five passerine birds from the Czech Republic were examined for the presence of sarcosporidia; only 5 (all great tits) were positive for muscle cysts. Three great tits experimentally fed sporocysts (12.2×8.4 , $11-13 \times 7.5-9 \mu m$) from a goshawk developed cysts while 2 house sparrows (*Passer domesticus*) were not infected. The cysts from great tits muscles were morphologically similar to those found in natural infections. The thin-walled cysts contained banana-shaped cystozoites $6.9 \times 1.8 (5.9-8.5 \times 1.3-2.3)\mu m$ in size. The natural life cycle of this *Sarcocystis* species involves goshawks and great tits. The relation of this species to *S. accipitris* is discussed.

Key words: Apicomplexa, birds, coccidia, host specificity, life cycle, Sarcocystis, transmission.

INTRODUCTION

Although birds have been known to harbour Sarcocystis cysts in their muscles since 1865 (Kühn ex Erickson 1940), it was only in 1977 that Munday and colleagues described the first sarcosporidian life-cycle with a bird (Gallus gallus) as the intermediate host and a carnivore (Canis familiaris) as the final host. Sarcosporidia have since been found in dozens of bird species (Kalyakin and Zasukhin 1975) but their life-cycles have been elucidated only in a few cases, mostly with a carnivore as final host (Levine 1986). In his abstract, Ashford (1975) was the first to suggest the existence of a sarcosporidian life cycle involving birds as both final (Accipiter nisus) and intermediate (Serinus canaria) host. The only named species involving birds as both final (A. gentilis) and intermediate (*S. canaria*) hosts in its life cycle is *Sarcocystis accipitris* (Černá and Kvašňovská 1986); the life cycle was investigated only in laboratory experiments.

During our investigation of sarcosporidiosis in passerine birds, the great tit was the only species found to be infected; the morphology of the cysts and cystozoites suggested this *Sarcocystis* species could have a bird of prey as final host (Munday et al. 1977). Among raptors living in the investigated area the genus *Accipiter* most often preys on birds (Hudec and Černý 1977), and the existence of a sarcosporidian life cycle with the goshawk as final and the great tit as natural intermediate host therefore seemed probable.

MATERIALS AND METHODS

Birds examined for the presence of muscular stages of sarcosporidia were unintentionally caught during trapping of mice or were collected for other investigations (house sparrows). They originated from Prague or from other localities in Czech Republic. The inoculum used in experimental infections originated from a single goshawk obtained from a

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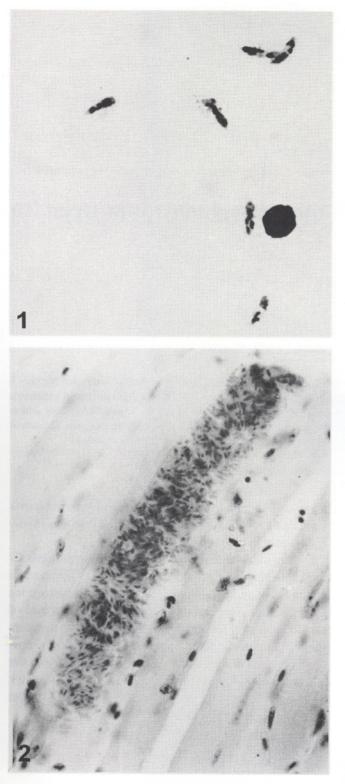


Fig. 1. Individual cystozoites from the muscle homogenate of a naturally infected great tit. Giemsa staining, x 1500

Fig. 2. Muscular cyst of *Sarcocystis* sp. in the great tit. Note the thin wall without any protrusions. Histological section, Harris hematoxylin, x 1000

taxidermist workshop DIPRA, Prague; the raptor was probably shot on an unknown locality in Czech Republic. Three great tits and 2 house sparrows were net-caught, each species caged separately and reared in an animal room. The birds were fed with a combined animal and plant feed (live insect larvae, seeds, vegetables, eggs) and subsequently infected per os with decanted gut contents of the goshawk containing an unknown number of sarcosporidian sporocysts. The probability that all the 3 tits were infected with sarcosporidia before the experiment is $(5/79)^3 = 0.00025$ (based on our data concerning the prevalence in great tits). Two tits were killed 2 months, 2 sparrows 4 months and 1 tit 5 months after infection. The muscles from half of the body of experimental as well as free-living birds were homogenized in saline using a grinding mortar, filtered through gauze and centrifuged for 15 min at approx. 1000 g. The sediment was smeared on slides, Giemsa-stained and examined for the presence of cystozoites under immersion objective. In the case of positive result, cystozoites (Fig. 1) were measured (n=10 in each specimen), their lengths compared using the multiple range test (ANOVA); muscles from the second half of the body were fixed in Bouin's fixative, embedded in paraplast, histologically processed and stained with Harris hematoxylin (Fig. 2).

RESULTS

One hundred and seventy five passerine birds were examined for the presence of muscular sarcosporidia. Among them, only 5 great tits were found to be infected, representing 2.9 % of all birds and 6.3 % of great tits examined (Table 1). These tits originated from different localities in the Czech Republic (2 from Prague, 50.05 N, 14.20 E; 3 others from the vicinity of Mělník, Central Bohemia, 50.21 N, 14.29 E; Choceň, East Bohemia, 50.00 N, 16.13 E; and Mikulov, South Moravia, 48.48 N, 16.38 E, resp.)

Table 1. Passerine birds examined for the presence of muscular sarcosporidiosis

| Bird species | Examined | Infected |
|--------------------------------------|----------|----------|
| Carduelis carduelis Goldfinch | 1 | 0 |
| Emberiza citrinella Yellowhammer | 3 | 0 |
| Erithacus rubecula Robin | 25 | 0 |
| Fringilla coelebs Chaffinch | 2 | 0 |
| Hippolais icterina Icterine Warbler | 1 | 0 |
| Parus caeruleus Blue Tit | 1 | 0 |
| Parus major Great Tit | 79 | 5 |
| Parus palustris Marsh Tit | 2 | 0 |
| Passer montanus Tree Sparrow | 2 | 0 |
| Passer domesticus House Sparrow | 47 | 0 |
| Phyloscopus trochilus Willow Warbler | 1 | 0 |
| Prunella modularis Hedge Sparrow | 3 | 0 |
| Serinus serinus Serin | 1 | 0 |
| Sitta europaea Nuthatch | 4 | 0 |
| Turdus merula Blackbird | 2 | 0 |
| Turdus philomelos Song Thrush | 1 | 0 |
| Total | 175 | 5 |

All 3 great tits fed experimentally with sporocysts, which measured 12.2 x 8.4 µm (11-13 x 7.5-9), n=20, SE 0.12 and 0.13, resp.), developed muscular cysts, while the 2 sparrows remained uninfected. The morphology of cystozoites and muscular cysts was similar in naturally and experimentally infected individuals; after Giemsa staining the cystozoites were banana-shaped with light blue cytoplasm and violet, slightly out of the center nucleus; their length and width was 6.9 x 1.8 µm (5.9-8.5 x 1.3-2.3, n=80, SE 0.07 and 0.02, resp.); fresh thin-walled muscular cysts measured 57 µm (45-84, n=4) in width. The cystozoites from experimental infection measured 7.0 x 1.7 µm (5.9-8.5 x 1.3-2.0, n=30, SE 0.13 and 0.04, resp.), those from natural infections 6.9 x 1.8 µm (5.9-8.5 x 1.3-2.3, n=50, SE 0.07 and 0.03, resp.) The similarity of cystozoites from natural and experimental infection was confirmed using the multiple range test (ANOVA).

DISCUSSION

The successful experimental inoculation of sarcosporidian sporocysts isolated from the goshawk to the great tit, and the similarity of sarcocysts and cystozoites derived from experimental and natural infections suggests this passerine bird acts as natural intermediate host of this *Sarcocystis* species.

Although goshawks usually prey upon larger birds, small passerines are also found in their diet (Goszczyński and Piłatowski 1986), especially in the nesting period (Hudec and Černý 1977); and the great tit was also reported from goshawk diet (Mañosa 1994).

The great tit was the only bird species in which sarcocysts were found, although a relatively high number of sparrows (47) were also examined; after being fed sporocysts the sparrows were not infected, and they are probably not involved in the life cycle of this parasite.

Oocysts/sporocysts isolated from the goshawk were reported to be infective for the canary (*Serinus canaria*) (Černá and Kvašňovská 1986). The canary, however, cannot act as intermediate host in central European area due to its range (the Canaries, the Azores and Madeira (Heinzel et al. 1983)); on the contrary, the ranges of goshawk and great tit overlap in the whole Europe (Heinzel et al. 1983). Therefore, it would be tempting to conclude that the *Sarcocystis* species in our work is *Sarcocystis accipitris* and that the great tit is the natural intermediate host of this species; the morphology of cystozoites and muscular cysts being similar to those reported by Černá and Kvašňovská (1986) further supports this hypothesis. However, sporocysts used for inoculation differ in their morphology and size, being relatively wide and measuring 15-17 x 13-15 μ m in the case of *S. accipitris* (Černá and Kvašňovská 1986), while those used in our experiment measured 11-13 x 7.5-9; they resemble sporocysts from sparrowhawk (*A. nisus*) which were not infective for the canary (Černá and Kvašňovská 1986) but it is unclear if in their experiment the sporocysts were still viable. Unfortunately, we had no sparrowhawk available for transmission study.

The fact that the great tit and the canary belong to different families (Paridae and Fringillidae, resp.) also does not help to answer the question if those findings are identical or not. From the species with well-known life cycles, Sarcocystis falcatula, a species with opossum (Didelphis virginiana) as final host, develops muscle cysts in members of 2 different bird orders (Box and Smith 1982); oocysts of Sarcocystis dispersa from barn owl (Tyto alba) are infective for all the members (7 genera tested) of the family Muridae except the laboratory rat (Rattus norvegicus) but not for family Cricetidae and Caviidae (Votýpka, unpub. obs.) The question is which of these two Sarcocystis species should we chose for comparison with "Sarcocystis accipitris"? Oocysts/sporocysts from the goshawk are also known to be infective for laboratory mice (Černá 1977, confirmed by Kolářová ex Černá 1986) but, to our knowledge, there is no Sarcocystis species with an intermediate host spectrum including both mammals and birds.

For the above reasons, the species identity of *Sarcocystis* species using great tit as intermediate and goshawk as final host remains unclear.

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226 M. Svobodová

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