

Three new species of *Isospora* Schneider, 1881 (Apicomplexa: Eimeriidae) from the lesser seed-finch, *Oryzoborus angolensis* (Passeriformes: Emberizidae) from Brazil

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Three new coccidian (Apicomplexa: Eimeriidae) species are reported from the lesser seed-finch, *Oryzoborus angolensis* from Brazil. Sporulated oocysts of *Isospora curio* n. sp. are spherical to subspherical; 24.6×23.6 ($22-26 \times 22-25$) μm , shape-index (SI, length/width) of 1.04 (1.00-1.15). Oocyst wall is bilayered, ~ 1.5 μm thick, smooth and colourless. Micropyle and oocyst residuum are absent. The sporocysts are ovoid, 13.2×10.9 ($15-17 \times 10-13$) μm , SI = 1.56 (1.42-1.71), with a small Stieda body and residuum composed of numerous granules scattered among the sporozoites. Sporozoites are elongated and possess a smooth surface and two distinct refractile bodies. Oocysts of *Isospora braziliensis* n. sp. are spherical to subspherical, 17.8×16.9 ($16-19 \times 16-18$) μm , with a shape-index of 1.06 (1.00-1.12) and a smooth, single-layered wall ~ 1 μm thick. A micropyle, oocyst residuum and polar granules are absent. Sporocysts are ellipsoid and slightly asymmetric, 13.2×10.8 ($12-14 \times 9-12$) μm , SI = 1.48 (1.34-1.61). Each sporocyst contains a barely visible Stieda body and a residuum composed numerous of granules. Sporozoites are elongated and each of them contains two distinct refractile bodies. Oocysts of *Isospora paranaensis* n. sp. are subspherical to broadly ellipsoid 24.3×19.8 ($22-26 \times 18-22$) μm , SI = 1.22 (1.15-1.38) with smooth single-layered wall ~ 1.5 μm thick. A micropyle and oocyst residuum are absent, but one distinct ellipsoid polar granule ($2.5-3.5 \times 1.5-2.5$ μm) is present. Sporocyst are ovoid, 15.7×10.1 ($14-18 \times 8-12$) μm , SI = 1.46 (1.31-1.72), with distinct Stieda and sub-Stieda bodies. Each sporocyst contains a spherical sporocyst residuum, 4 μm in diameter. All described isosporan species represent a possible cause of acute coccidiosis for *O. angolensis* in captivity.

Key words: *Isospora* - *curio* - lesser seed-finch - *Oryzoborus angolensis* - Brazil

The lesser seed-finch *Oryzoborus angolensis* (Linnaeus, 1766) (Passeriformes: Emberizidae) is a bird distributed in open woodland, savanna, bushes, and shrubs in forest edge of Latin America. They are found from North of the Lesser Antilles through Venezuela, Brazil, and Bolivia to South Argentina (Sick 1993). The lesser seed-finches are highly coveted as pet birds and are considered to be among the noblest singing birds. This feature has made this species one of the most highly prized songbirds in Brazil and many birds are bred in captivity. These finches are also frequently trapped from the wild and, consequently, wild lesser seed-finch populations have declined.

No species of *Isospora* have previously been described in emberizid birds of the genus *Oryzoborus*. The present study provides descriptions of three new species of *Isospora* from *O. angolensis*.

MATERIALS AND METHODS

A total of 675 *O. angolensis* were examined in this study. The birds originated from 48 private aviaries localized in the state of Mato Grosso do Sul, Brazil. They were reared in suspended cages and individual faecal samples were collected immediately upon defecation on tinfoil on the bottom of the cages at the different day-time. Faecal material was placed in vials containing 2.5% (w/v) potassium dichromate solution and shipped to the University of Veterinary and Pharmaceutical Sciences in Brno for follow-up examination. Faecal samples were 3-8 weeks old when screened for coccidia using flotation in Sheather's sugar solution (specific gravity 1.30). The sporulated oocysts were examined and photographed with an Olympus AX 70 microscope using Nomarski interference contrast optics (DIC). All measurements were made using a calibrated ocular micrometer and reported as means in micrometers (μm), followed by the range in parentheses.

RESULTS

During the coprological examination of the 675 *O. angolensis*, we detected the presence of oocysts of genus *Isospora* in 178 birds. The morphological evaluation of sporulated oocysts revealed three different species of *Isospora* that are described and discussed below.

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Isospora curio n. sp.

Figs. 1, 2, 7

Description - Oocysts are spherical to subspherical; 24.6×23.6 ($22-26 \times 22-25$) ($n = 30$), shape-index (SI, length/width) 1.04 (1.00-1.15). Micropyle and oocyst residuum are absent. Oocyst wall is bilayered, ~ 1.5 thick, smooth and colourless. Sporocysts are ovoid, 13.2×10.9 ($15-17 \times 10-13$) ($n = 30$), with a smooth, single-layered wall ~ 0.5 thick, sporocyst SI = 1.56 (1.42-1.71). Stieda body is present, ~ 0.7 high $\times 2.0-2.5$ wide, sub-Stieda body is absent. Sporocyst residuum is composed of numerous granules scattered among sporozoites. Sporozoites are elongated, 18.2×3.5 ($16-19 \times 3-4$) ($n = 12$), possessing a smooth surface and two distinct refractile bodies. Anterior refractile body is spherical, ~ 3 in diameter, the posterior one is elongated, 6.2×3.2 ($5-7 \times 3.0-3.5$) ($n = 10$). The spherical nucleus is situated between the refractile bodies.

Type host - *Oryzoborus angolensis* (Linnaeus 1766) (Aves: Passeriformes: Emberizidae), the lesser seed-finch.

Type locality - Material was isolated from faeces of captive birds in Mato Grosso do Sul.

Prevalence - 14.66 %.

Site of infection - Unknown, oocysts were recovered from faeces.

Sporulation - Exogenous. Time required unknown.

Type material - Photo-syntypes deposited under the depository number A 224 in the Department of Parasitology, University of Veterinary and Pharmaceutical Sciences Brno, Czech Republic.

Etymology - The specific name is derived from the common local name for *O. angolensis*.

Isospora braziliensis n. sp.

Figs 3, 4, 8

Oocysts are spherical to subspherical, 17.8×16.9 ($16-19 \times 16-18$) ($n = 50$), SI = 1.06 (1.00-1.12). Micropyle and oocyst residuum are absent. Oocyst wall is single-layered, ~ 1.0 thick, smooth and colourless. Sporocysts are ellipsoid and slightly asymmetric, 13.2×10.8 ($12-14 \times 9-12$) ($n = 30$), with a smooth, single-layered wall ~ 0.5 thick, SI = 1.48 (1.34-1.61). Stieda body is present, appearing as a fine plug, ~ 1 wide. Sub-Stieda body is absent. Sporocyst residuum composed of numerous granules scattered among sporozoites. Sporozoites are elongated, 13.2×3.1 ($12-14 \times 2.5-3.5$) ($n = 10$), possessing a smooth surface and two distinct refractile bodies. Anterior refractile body is spherical, ~ 2 in diameter, the posterior one is elongate, 5.4×2.4 ($4-6 \times 2.0-3.0$) ($n = 10$). The spherical nucleus is situated between the refractile bodies.

Type host - *Oryzoborus angolensis* (Linnaeus 1766) (Aves: Passeriformes: Emberizidae), lesser seed-finch.

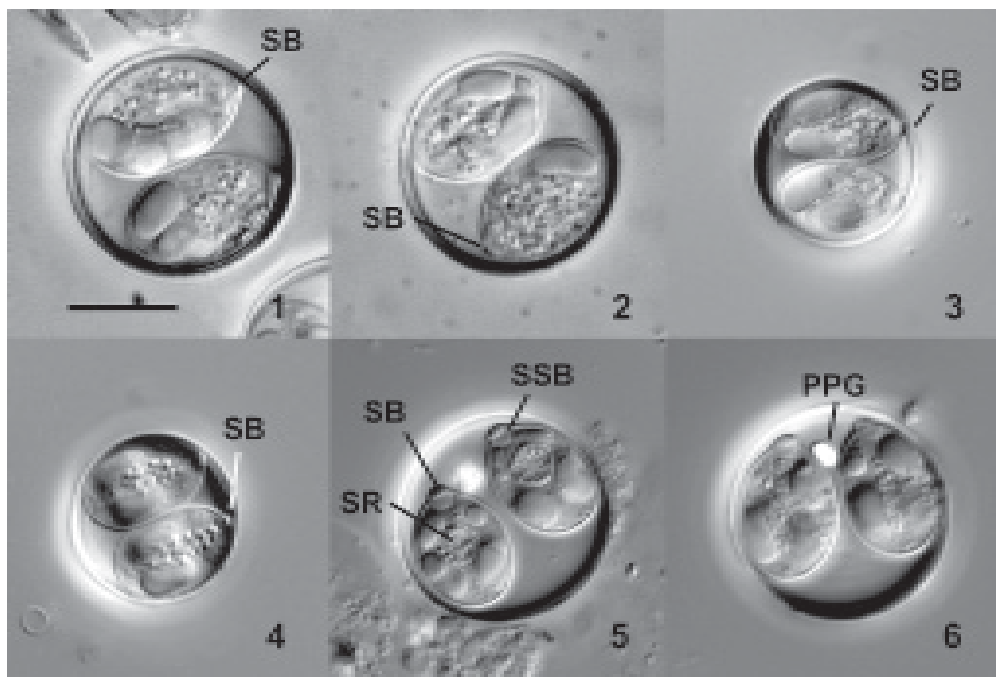
Type locality - Material was isolated from faeces of captive birds in Mato Grosso do Sul.

Prevalence - 3.55 %.

Site of infection - Unknown, oocysts recovered from faeces.

Sporulation - Exogenous. Time required unknown.

Type material - Photo-syntypes deposited under the de-



Figs 1-2: Nomarski interference contrast (NIC) photographs of oocysts of *Isospora curio* n. sp. with delicate Stieda body (SB). Figs 3-4: NIC photographs of oocysts of *I. braziliensis* n. sp. with a barely visible Stieda body (SB). Figs 5-6: NIC photographs of oocysts of *I. paranaensis* n. sp. with SB, substieda (SSB) bodies and distinct polar granule (PG). Scale bar = 10 μ m.

pository number B 231 in the Department of Parasitology, University of Veterinary and Pharmaceutical Sciences Brno, Czech Republic.

Etymology - The specific name is derived from the country of origin of the host.

Isoospora paranaensis n. sp

Figs 5, 6, 9

Oocysts are subspherical to broadly elliptical, 24.3×19.8 ($22-26 \times 18-22$), ($n = 30$), $SI = 1.22$ ($1.15-1.38$). Micropyle and oocyst residuum absent, but one distinct ellipsoid polar granule ($2.5-3.5 \times 1.5-2.5$) is present. Oocyst wall is single-layered, ~ 1.5 thick, smooth and colourless. Sporocyst are ovoid, 15.7×10.1 ($14-18 \times 8-12$) ($n = 30$), with a smooth single-layered wall ~ 0.5 thick, $SI = 1.46$ ($1.31-1.72$). A Stieda body is present, ~ 1.5 high $\times 2.0-2.5$ wide, sub-Stieda body also present, $\sim 2.0-2.5$ high $\times 2.0-2.5$ wide. A spherical sporocyst residuum is present, ~ 4 in diameter, and usually as a cluster of numerous granules. Sporozoites are elongated, 14.9×3.0 ($13-16 \times 2-4$) ($n = 10$), possessing a smooth surface and two distinct refractile bodies. Anterior refractile body is spherical, $2-3$ in diameter, the posterior one is elongate, 4.8×3.2 ($4-6 \times 2.5-3.5$) ($n = 10$). The spherical nucleus is situated between the refractile bodies.

Type host - *Oryzoborus angolensis* (Linnaeus 1766) (Aves: Passeriformes: Emberizidae), the lesser seed-finch.

Type locality - Material was isolated from faeces of captive birds in Mato Grosso do Sul.

Prevalence - 1.77 %.

Site of infection - Unknown, oocysts recovered from faeces.

Sporulation - Exogenous. Time required unknown.

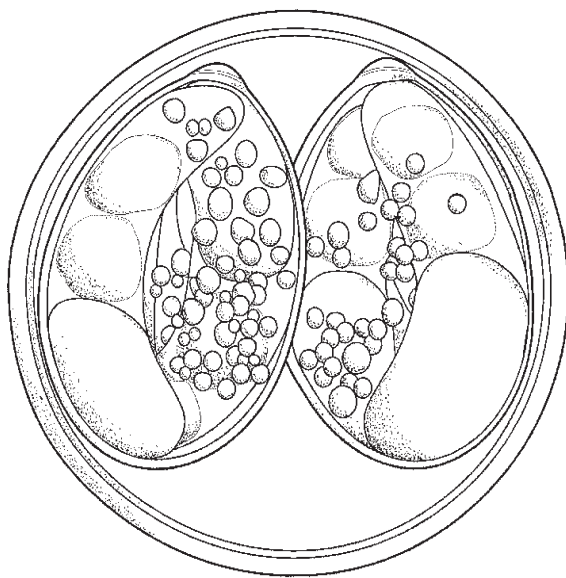


Fig. 7: composite line drawing of sporulated oocyst of *Isoospora curio* n. sp. Scale bar = 10 μ m.

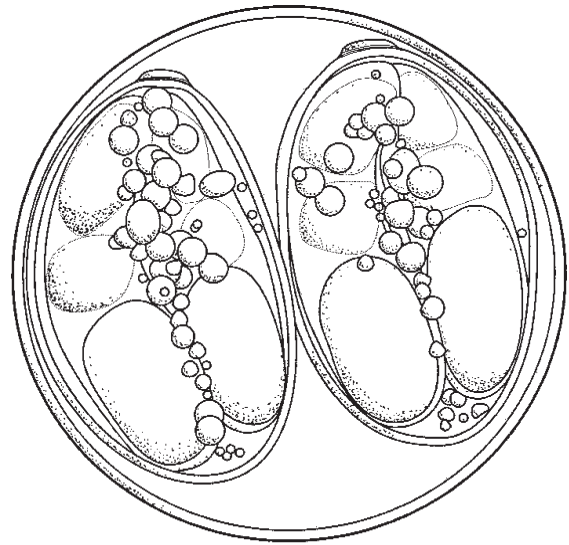


Fig. 8: composite line drawing of sporulated oocyst of *Isoospora braziliensis* n. sp. Scale bar = 10 μ m.



Fig. 9: composite line drawing of sporulated oocyst of *Isoospora paranaensis* n. sp. Scale bar = 10 μ m.

Type material - Photo-syntypes deposited under the depository number B 231 in the Department of Parasitology, University of Veterinary and Pharmaceutical Sciences Brno, Czech Republic.

Etymology - The specific name is derived from the State of Parana, Brazil.

DISCUSSION

To date, there are around 50 species of *Isoospora* described from emberizids birds (<http://www.ksu.edu/para->

sitology/worldcoccidia/PASSER01?, de Carvalho Filho et al. 2005). However, no named species have been reported from the genus *Oryzoborus*. We compared the three observed species of *Isospora* from *O. angolensis* with valid isosporans described from emberizids birds resident in South America or immigrating through this subcontinent and all were found to be different from all the presently described species. These are therefore, considered as new species.

The species most closely resembling *I. curio* n. sp. are *I. formarum* McQuistion and Capparella 1992, *I. tiaris* Ball and Daszak 1997, and *I. sporophilae* de Carvalho Filho, de Meireles, Ribeiro and Lopes 2005. The sporocysts *I. formarum*, from *Pitylus grossus* show morphological differences in the presence of a prominent substieda body which is absent in *I. curio* n. sp. Unlike *I. curio* n. sp., oocysts of *I. tiaris* from *Tiaris fuliginosa* are larger, and the sporocysts possess a prominent sub-Stieda bodies. In contrast, oocysts of *I. curio* n. sp. differ from *I. sporophilae* of *Sporophila caerulea* being larger and having no polar granule.

Oocysts of *I. braziliensis* n. sp. are similar in size to *I. flausinoides* de Carvalho Filho, de Meireles, Ribeiro and Lopes 2005 and *I. teixeirafilhoi* de Carvalho Filho, de Meireles, Ribeiro and Lopes 2005, both from *S. caerulea*. However, oocysts of *I. braziliensis* n. sp. apparently lack a polar granule and the sporocysts are slightly more elongated than those of *I. flausinoides* and *I. teixeirafilhoi*. In addition, oocysts of *I. teixeirafilhoi* possess bi-layered oocyst wall, unlike single-layered oocyst wall in *I. braziliensis* n. sp.

Oocysts of *I. paranaensis* n. sp. are similar in size to *I. formarum* McQuistion and Capparella 1992, *I. tiaris* Ball and Daszak 1997 and *I. sporophilae* de Carvalho Filho et al. 2005. However, *I. paranaensis* n. sp. tends to be broadly ellipsoid, compared with the subspherical oocysts of above mentioned species. In addition, *I. paranaensis* n. sp. differs from *I. formarum* in having a distinct polar granule.

The oocyst of *I. tiaris* differs from that of *I. paranaensis* n. sp. in its morphology, being larger and having morphological differences in the sub-Stieda body and sporocyst residuum. The sporocysts of *I. paranaensis* n. sp. show morphological differences in the presence of a sub-Stieda body, which is absent in those of *I. sporophilae*.

Based on all above mentioned morphological characteristics, together with host's distribution in nature we consider the species described here to be new coccidian species from captive *O. angolensis* kept in Brazil.

In the present study, coprological examinations indicate a widespread exposure to coccidian parasites among *O. angolensis* in this country. Coccidian infection may be responsible for loss of weight, low reproduction and death in breeding groups of this songbird. Further research should determine the pathology of these coccidian infections in these birds.

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