# Isospora borbai *n. sp. (Chromista: Apicomplexa: Eimeriidae) from gnateaters* Conopophaga *spp. (Passeriformes: Tyranni: Conopophagidae) in South America*

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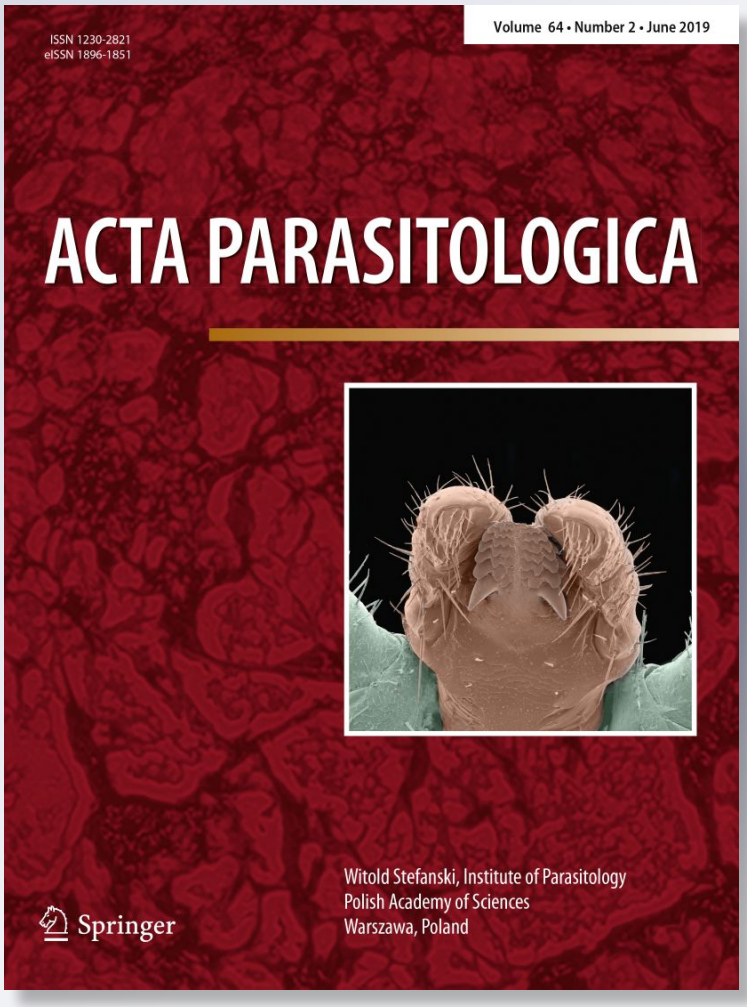


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ORIGINAL PAPER

# *Isospora borbai* n. sp. (Chromista: Apicomplexa: Eimeriidae) from gnateaters *Conopophaga* spp. (Passeriformes: Tyranni: Conopophagidae) in South America

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## Abstract

**Background** The gnateaters *Conopophaga* spp. are insectivorous passerines commonly observed in high and humid forests, where they remain lodged in thin branches and, sometimes, they fly to the ground to catch insects. The insectivorous feeding habit is related to low prevalence and density of coccidians in passerines; however, several coccidian species are recorded for families of insectivorous passerines.

**Purpose** This study aimed to examine the feces from gnateaters *Conopophaga* spp. captured in the municipality of Barra Mansa and in the Itatiaia National Park, State of Rio de Janeiro, Southeastern Brazil, to determine what coccidian parasites were present.

**Methods** Nine gnateaters were captured with mist nets. Coccidian oocysts were recovered from the fecal samples by flotation in Sheather's saturated solution. Morphological observations, line drawings, photomicrographs and measurements were made in optical microscopy and digitally edited. The molecular analysis included the study of the sequence of the mitochondrial cytochrome *c* oxidase subunit 1 (*cox1*) gene, with phylogenetic reconstructions based on the neighbor-joining and maximum likelihood analysis.

**Results** Four *Conopophaga* spp. were positive for oocysts. An *Isospora* sp. considered as new to science is described and identified from *Conopophaga melanops* (Vieillot, 1818) and *Conopophaga lineata* (Wied, 1831). *Isospora borbai* n. sp. has oocysts that are subspheroidal, 17–22 × 15–22 (20.2 × 19.1) μm, with rough, bilayered wall, *c.*1.7 μm thick. Micropyle present, but without micropyle cap. Oocyst residuum absent, but one or two polar granules are present. Sporocysts are ellipsoidal, 12–15 × 8–11 (14.1 × 9.1) μm. The Stieda body is knob-like to half-moon-shaped and sub-Stieda body is rounded. Sporocyst residuum is present, composed of scattered spherules of different sizes. Sporozoites are vermiform with refractile body and nucleus. Molecular analysis at the *cox1* gene exhibited similarity greater than 99% with *Isospora* spp. isolates from other Neotropical passerine birds.

**Conclusion** Based on the morphological and molecular features, *I. borbai* is considered as new to science and the first coccidian species recorded from Conopophagidae.

**Keywords** Morphology · Molecular biology · Taxonomy · Phylogeny · Coccidia · Oocysts · Neotropical birds · Thamnophilida · Médio Paraíba Region · Parque Nacional do Itatiaia

## Introduction

The parvorder Thamnophilida (Passeriformes: Tyranni) is divided into three families: Thamnophilidae, Melanopareiidae and Conopophagidae. The family Conopophagidae brings together two genera: *Conopophaga* Vieillot, 1816 and *Pittasoma* Cassin, 1860. In Brazil, seven *Conopophaga* spp.

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are reported; however, the two *Pittasoma* spp. are restricted to Colombia, Costa Rica, Panama and Ecuador [4, 12].

The gnateaters *Conopophaga* spp. are small, with a long tarsus, short tail and rounded wing. Most species have an elongated, usually white, post-ocular stripe. They are terrestrial insectivores commonly observed in high and humid forests, where they remain lodged in thin branches and, sometimes, they fly to the ground to catch insects [13].

The insectivorous feeding habit was previously related to the low prevalence and density of *Isospora* spp. in passerines [7]; however, several species of coccidia are recorded for families of insectivorous passerines [1]. In this context, the aim of this study was to examine the feces from gnateaters *Conopophaga* spp. captured in different localities in the Médio Paraíba region of the State of Rio de Janeiro, Southeastern Brazil, to determine what coccidian parasites were present.

## Materials and Methods

### Sample Collection

A total of five expeditions were conducted in two different localities in the Médio Paraíba Region in the State of the Rio de Janeiro, Southeastern Brazil: (1) Itatiaia National Park (Parque Nacional do Itatiaia), a protected area with a high degree of vulnerability, located in the Serra da Mantiqueira [9]; and (2) an Atlantic forest fragment area at the Municipality of Barra Mansa. A total of five black-cheeked gnateater *Conopophaga melanops* (Vieillot, 1818) (all from Itatiaia National Park) and four rufous gnateater *Conopophaga lineata* (Wied, 1831) (two from Itatiaia National Park and two from Barra Mansa) were captured with mist nets. The birds were kept in individual boxes and feces collected immediately after defecation. After identification of the species, the birds were photographed and released and stool samples were placed in centrifuge tubes containing a potassium dichromate 2.5% ($K_2Cr_2O_7$) solution at 1:6 (v/v).

### Morphological Analyses

Samples were taken to the Laboratório de Biologia de Coccídios, Universidade Federal Rural do Rio de Janeiro (UFRRJ). Samples were incubated at room temperature (25 °C) for 10 days or until ~ 70% of the oocysts were sporulated. Oocysts were isolated by flotation in Sheather's sugar saturated solution (specific gravity: 1.20) and examined microscopically using the technique described by Duszynski and Wilber [8] and Berto et al. [2]. Morphological observations, line drawings, photomicrographs and measurements were made using an Olympus BX binocular microscope (Olympus Optical, Tokyo, Japan) coupled to a digital camera Eurekam 5.0 (BEL Photonics, Monza, Italy). Line drawings were edited using two software applications from CorelDRAW® (Corel Draw Graphics Suite, Version 11.0, Corel Corporation, Canada), i.e., Corel DRAW and Corel PHOTO-PAINT. All measurements are in micrometres and are given as the range followed by the mean in parentheses.

### Molecular Analyses

An individual oocyst identified with the characteristic features of the new species under light microscopy was isolated and resuspended in PBS [5]. DNA was extracted from the oocyst using the Qiagen DNeasy Blood and Tissue Kit (Qiagen, São Paulo, Brazil) according to the manufacturer's instructions. To fully lyse the oocyst, four freeze–thaw cycles were applied prior to the DNA extraction. The PCR amplification for the mitochondrial cytochrome *c* oxidase subunit 1 (*cox*1) gene was carried out using a nested PCR, as previously described by Dolnik et al. [5] and Yang et al. [19]. The external primers COIbF1 (5′-GWT CAT TAG TAT GGG CAC ATC A-3′) and COIbR1 (5′-CCA AGA GAT AAT ACR AAR TGG AA-3′) produced a PCR product of *c*.302 bp in size. The internal primes COIbF2 (5′-GGG CAC ATC ATA TGA TGA C-3′) and COIbR2 (5′-ATA GTA TGT ATC ATG TAR WGC AA-3′) produced an amplicon of 257 bp in size. The PCR contained 10 µl of 5 × Green GoTaq® Flexi Buffer, 3 µl of 25 mM $MgCl_2$, 1 µl of 10 mM dNTPs, 0.4 µM of each primer, 1.25 units of GoTaq® DNA polymerase, 3 µl of DNA (for primary reaction) or 3 µl primary PCR product (for the secondary reaction). Both primary and secondary PCR were conducted using the same cycling conditions: one cycle of 94 °C for 5 min, followed by 35 cycles of 94 °C for 30 s, 47 °C for 45 s, and 72 °C for 1 min and a final extension of 72 °C for 5 min. The amplicons from the second round of PCR were purified using the Qiagen MinElute PCR Purification (Qiagen, São Paulo, Brazil). All PCR products were sequenced using the PCR forward and reverse primers by Ludwig Biotechnology, were an ABI-Prism 3500 Genetic Analyzer (Applied Biosystems, Foster City, California) was used for Sanger sequencing. The results of the sequencing reactions were analysed and edited using the program Chromas 2.6.

### DNA Sequence Analyses

The newly generated sequences were compared to those for *Isospora* spp. and other coccidian parasites available on the GenBank database using the Basic Local Alignment Search Tool (BLAST). Phylogenetic trees were constructed for

*Isospora* spp. at the *cox*1 sequences for additional isolates from GenBank. Alignment and parsimony analyses were conducted using MEGA version 7 [18]. The evolutionary history was inferred using the neighbor-joining (NJ) and maximum likelihood (ML) methods and the distances were computed using the Tamura-Nei method based on model selection using ModelTest in MEGA 7. Bootstrap analyses were conducted using 1000 replicates to assess the reliability of inferred tree topologies.

## Results

Nine *Conopophaga* spp. were examined and four were positive for coccidia (two *C. melanops* and one *C. lineata* from Itatiaia National Park; and one *C. lineata* from Barra Mansa). All observed oocysts were characteristic of *Isospora*. This material is described below.

Family Eimeriidae Minchin, 1903.

Genus *Isospora* Schneider, 1881.

***Isospora borbai* Silva-Carvalho et Berto n. sp.** (Figures 1, 2).

Oocysts ($n = 32$) subspheroidal, 17–22 × 15–22 (20.2 × 19.1); length/width (*L*/*W*) ratio 1.0–1.1 (1.06). Wall bi-layered, 1.5–2.1 (1.7) thick, outer layer rough, *c.*2/3 of total thickness. Micropyle present, without micropyle cap or wrinkles; however, generally with slight invagination of the inner layer. Oocyst residuum absent, but one or two

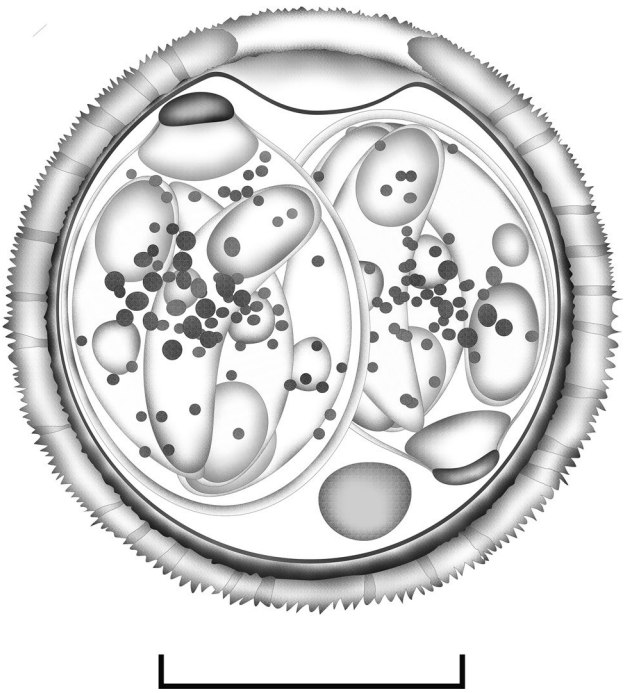

**Fig. 1** Composite line drawing of the sporulated oocyst of *Isospora borbai* n. sp. from *Conopophaga* spp. Scale-bar: 10 µm

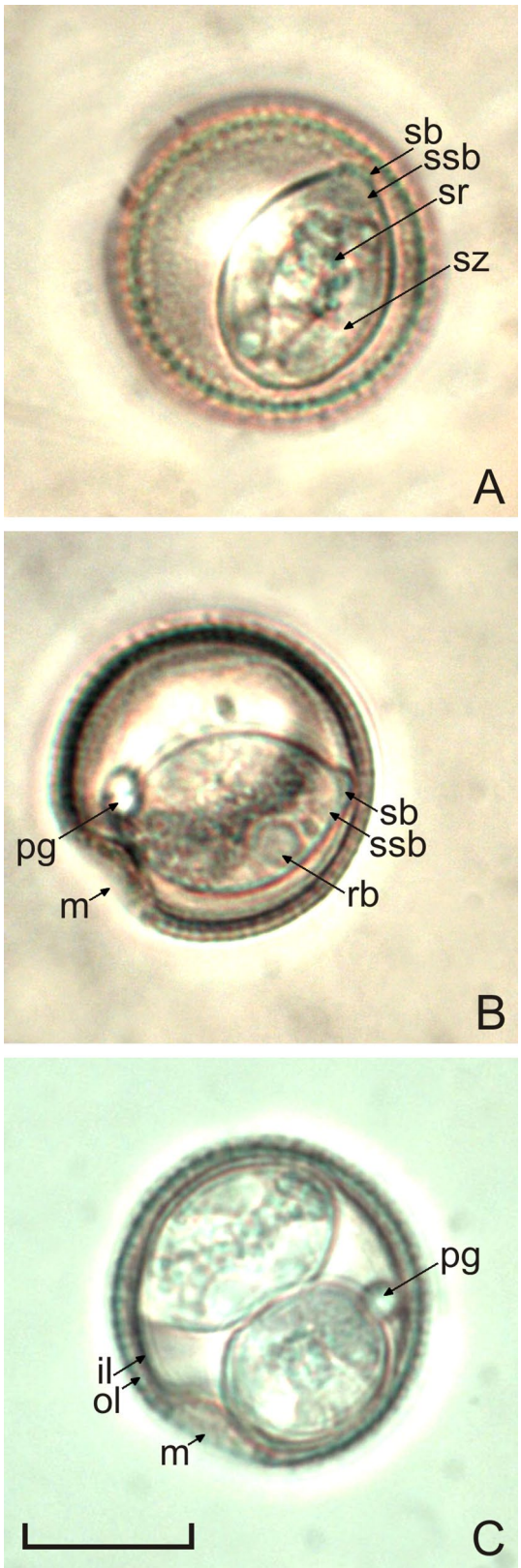


**Fig. 2** Photomicrographs of sporulated oocysts of *Isospora borbai* n. sp. from *Conopophaga* spp. Inner (il) and outer (ol) layers of the oocyst wall, *m* micropyle, *pg* polar granule, *sb* Stieda, *ssb* sub-Stieda bodies, *sr* sporocyst residuum, *sz* sporozoite, *rb* refractile body. All to same scale. Scale-bar: 10 µm

**Table 1** Comparative morphology of *Isospora* spp. recorded from the parvorder Thamnophilida

| Coccidia | Hosts | | References | Oocysts | | | | | Sporocysts | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Species | Family | | Shape | Measurements (µm) | Shape index | Wall | Polar granule | Shape | Measurements (µm) | Shape index | Stieda body | Substieda body | Residuum |
| *Isospora sagittulae* McQuistion, Capparella, [11] | *Hylophylax naevioides* (Lafresnaye, 1847) | Thamnophilidae | McQuistion and Capparella [11] | Ovoidal to ellipsoidal | 25-30×21- 24 (27.5×21.8) | 1.27 | Bilayered, smooth | Present, 1–3 | Subspheroidal to ovoidal | 13-16×12-13 (14.8×12.4) | 1.19 | Present, thin and dense | Present, triangular | Diffuse |
| | *Gymnopithys salvini* (Berlepsch, 1901); *Willisornis poecilinotus* (Cabanis, 1847) | Thamnophilidae | Berto et al. [3] | Ovoidal to ellipsoidal | 27-31×20-24 (28.4×22.4) | 1.27 | Bilayered, smooth | Present, 1–3 | Subspheroidal to ovoidal | 13-17×12-14 (15.0×12.6) | 1.2 | Thin and flattened, 0.5×2.0 | Triangular to rounded, 2.5×5.0 | Diffuse |
| | *Pyriglena leucoptera* (Vieillot, 1818) | Thamnophilidae | Silva-Carvalho et al. [16] | Ovoidal to ellipsoidal | 29-33×22-26 (30.8×24.4) | 1.27 | Bilayered, smooth | Present, 1–3 | Subspheroidal to ovoidal | 14-17×12-15 (15.9×13.4) | 1.19 | Thin and flattened, 0.5×2.0 | Triangular to rounded, 2.5×5.0 | Diffuse |
| *Isospora parnaitatiaiensis* Silva, Rodrigues, Lopes, Berto, Luz, Ferreira, Lopes, [14] | *Pyriglena leucoptera* (Vieillot, 1818) | Thamnophilidae | Silva et al. [14] | Ellipsoidal | 22-27 × 18-21 (23.8×19.4) | 1.23 | Bilayered, smooth | Present, 1–2 | Ellipsoidal | 13-16×8-10 (14.6 × 9.3) | 1.6 | Nipplelike to knoblike, 1.0×2.0 | Rounded to rectangular, 1.5×2.0 | Compact |

**Table 1** (continued)

| Coccidia | Hosts | | References | Oocysts | | | | | Sporocysts | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Species | Family | | Shape | Measurements (µm) | Shape index | Wall | Polar granule | Shape | Measurements (µm) | Shape index | Stieda body | Substieda body | Residuum |
| *Isospora borbai* Silva-Carvalho and Berto n. sp. | *Conopophaga melanops* (Vieillot, 1818); *Conopophaga lineata* (Wied, 1831) | Conopophagidae | Current work | Subspheroidal | 17–22×15–22 (20.2×19.1) | 1.06 | Bi-layered, rough | Present, 1–2 | Ellipsoidal | 12–15×8–11 (14.1×9.1) | 1.56 | Knob-like to half-moon-shaped, 1.0×2.5 | Rounded, 2.0×3.5 | Diffuse |

(frequently one subspheroidal) polar granules are present. Sporocysts ($n=25$) ellipsoidal, 12–15×8–11 (14.1×9.1); *L*/*W* ratio 1.4–1.7 (1.56). Stieda body present, knob-like to half-moon-shaped, 1.0×2.5; sub-Stieda present, rounded, 2.0×3.5; para-Stieda body absent; sporocyst residuum present, composed of scattered spherules of different sizes. Sporozoites vermiform, with posterior refractile body and centrally located nucleus.

*Type-host Conopophaga lineata* (Wied, 1831) (Aves: Passeriformes: Tyranni: Conopophagidae), rufous gnateater.

*Other host Conopophaga melanops* (Vieillot, 1818) (Aves: Passeriformes: Tyranni: Conopophagidae), black-cheeked gnateater.

*Type locality* Parque Nacional do Itatiaia (22°27′S, 44°35′W), southeastern Brazil.

*Other locality* Barra Mansa (22°29′S, 44°09′W), southeastern Brazil.

*Type specimens* Photosyntypes, line drawing, and oocysts in 70% ethanol are deposited at the Museu de Zoologia at the Universidade Federal Rural do Rio de Janeiro, Brazil, under the accession number MZURPTZ2018008. Phototypes and line drawings are also deposited and available (http://r1.ufrrj.br/labicoc/colecao.html) in the Parasitology Collection of the Laboratório de Biologia de Coccídios, at UFRRJ, under the repository number P-91/2018. Photographs of the type-host specimen (symbiotype) are deposited in the same collection.

*Site in host* Unknown.

*Prevalence* 44% (four out of nine birds infected).

*Representative DNA sequence* One representative *cox*1 sequence was deposited in the GenBank database under the accession number MK057528.

*ZooBank registration* To comply with the regulations set out in article 8.5 of the amended 2012 version of the International Code of Zoological Nomenclature [10], details of the new species have been submitted to ZooBank. The Life Science Identifier (LSID) for *Isospora borbai* is urn:lsid:zoobank.org:act:D3BE104D-300F-4251-981C-393CD86800F7.

*Etymology* The specific name is derived from the family name of the Brazilian parasitologist Dr Hélcio Resende Borba, given in his honor for his contribution to the study of antiparasitic activity of plants.

*Remarks* To date, only two *Isospora* spp. are recorded from hosts of the parvorder Thamnophilida (Table 1). *Isospora sagittulae* McQuistion, Capparella, [11] and *Isospora parnaitatiaiensis* Silva, Rodrigues, Lopes, Berto, Luz, Ferreira, Lopes, 2015 were recorded from antbirds of the family Thamnophilidae; therefore, no *Isospora* sp. is recorded from the families Conopophagidae and Melanopareiidae until now. As shown in Table 1, *I. borbai* is easily differentiated from these two *Isospora* spp. from Thamnophilidae, due to

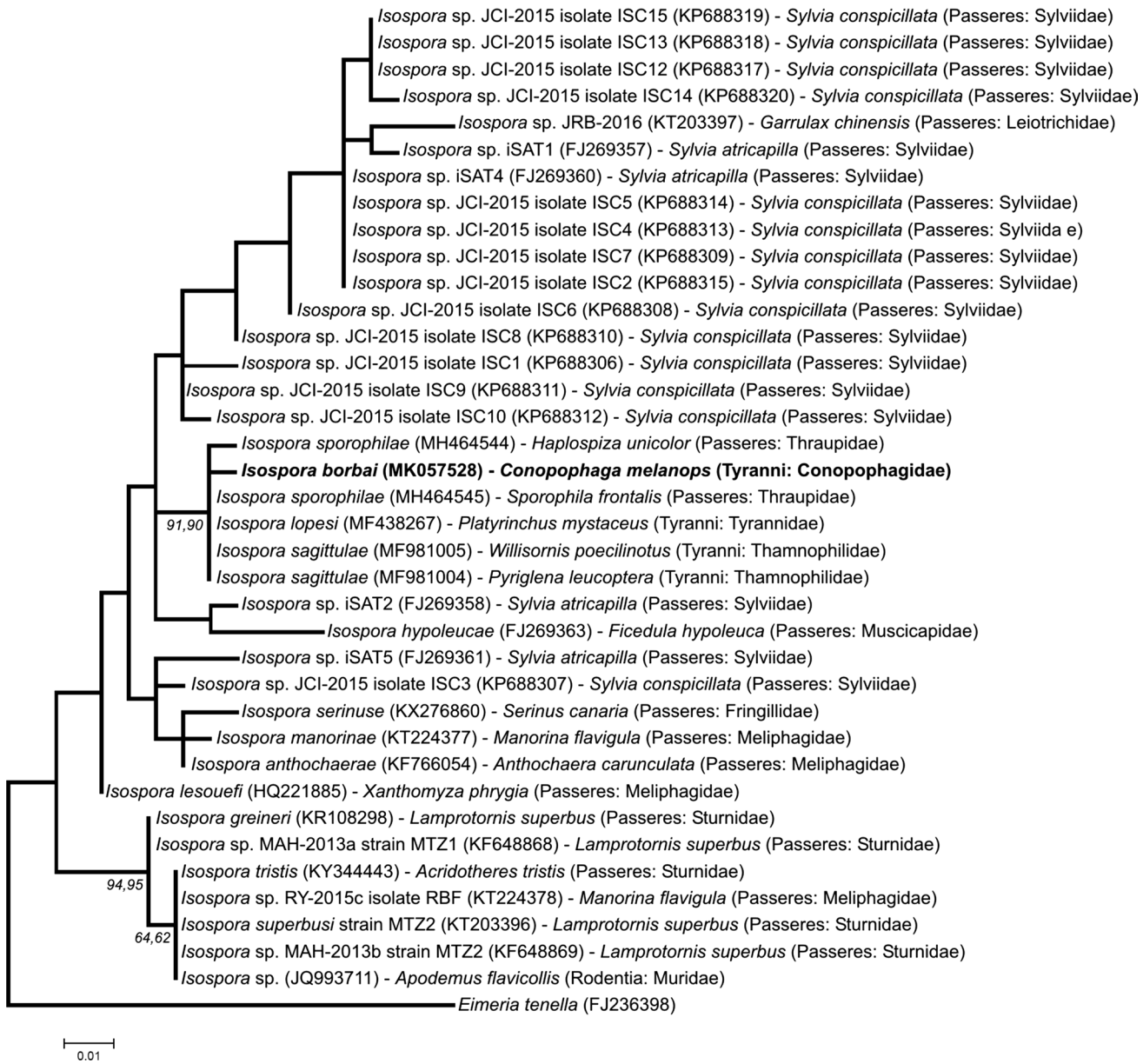


**Fig. 3** Maximum likelihood tree estimated from the *cox*1 sequences. Numbers at nodes represent bootstrap support (1000 replicates; only values > 50% shown) for Neighbor-Joining and Maximum Likelihood, respectively. The scale-bar represents the number of nucleotide substitutions per site

their smaller size, subspheroidal shape, micropyle and rough outer layer of the oocyst wall.

*Phylogenetic analysis* DNA amplification of an individual oocyst of *I. borbai* n. sp. recovered from a *C. melanops* from Itatiaia National Park showed a clear band of *c.*250 bp. Phylogenetic analysis included 36 sequences for avian *Isospora* spp. available on GenBank (Fig. 4). *Eimeria tenella* (Railliet, Lucet, 1891) was used as the outgroup. *Isospora borbai* sat in a clade with the highest similarity of 99.0–99.5% with *Isospora lopesi* Silva-Carvalho et Berto, 2018 [15], *Isospora sagittulae* McQuistion et Capparella, 1992 [16] and *Isospora sporophilae* Carvalho-Filho, Meireles, Ribeiro et Lopes, 2005 [17] (Fig. 3). In a second analysis, a subset of 215 bp long *cox*1 gene sequences for 14 *Isospora* spp. was used (Fig. 4). In this analysis, *I. borbai* was again grouped with *I. lopesi*, *I. sagittulae* and *I. sporophilae*, next to the other clade with *Isospora hypoleucae* Dolnik, Rönn et Bensch, [6] (Dolnik et al. [5]) and *Isospora* isolates from Eurasian blackcaps *Sylvia atricapilla* (Linnaeus, 1758) (Dolnik et al. [5]) with similarities of 95.7% and 94.8–97.1%, respectively.

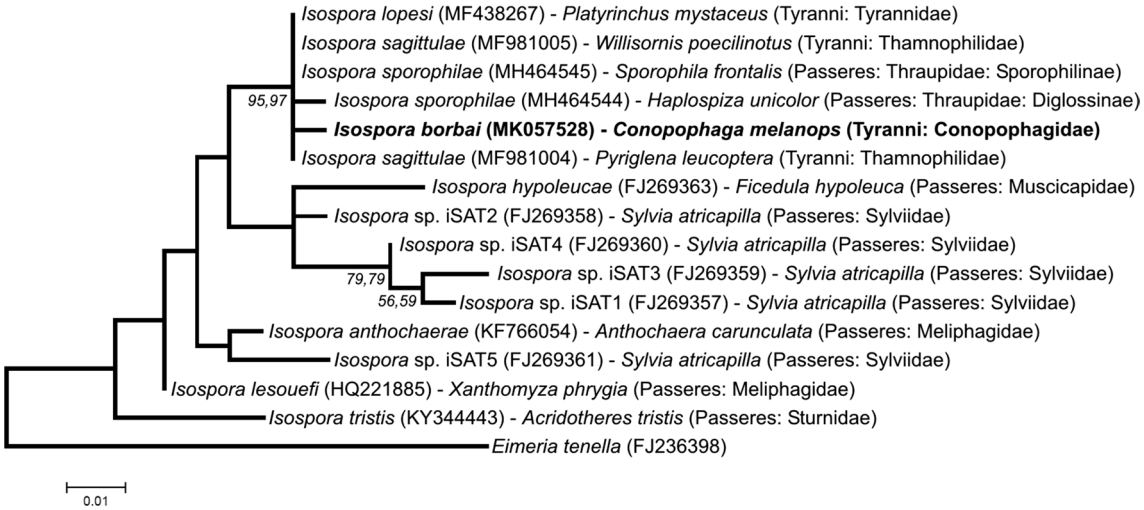


**Fig. 4** Maximum likelihood tree estimated from the 215 bp long *cox*1 sequence dataset for *Isospora* spp. Numbers at nodes represent bootstrap support (1000 replicates; only values > 50% shown) for Neighbor-Joining and Maximum Likelihood, respectively. The scale-bar represents the number of nucleotide substitutions per site

## Discussion

*Isospora borbai* is the first coccidian species to be described from the family Conopophagidae. Duszynski and Wilber [8] advise that new coccidian species should be compared morphologically with all species recorded in the family of the host; therefore, due to the lack of descriptions of coccidians from conopophagids, *I. borbai* was compared with the coccidians from the parvorder Thamnophilida. In this sense, *I. borbai* was compared to *I. sagittulae* and *I. parnaitatiaiensis*, which are the only coccidian species recorded from the parvorder Thamnophilida, specifically from the family Thamnophilidae (Table 1). In any case, the oocysts of *I. borbai* are quite distinctive because they have a rough wall with a micropyle, which are unusual characteristic features in *Isospora* spp.

The phylogenetic analysis (Figs. 3, 4) brings together *I. borbai* with *I. sagittulae*, which are also parasites from the parvorder Thamnophilida, and *I. lopesi*, parasite of eastern white-throated spadebills *Platyrinchus mystaceus* Vieillot, 1818 that also belong to the suboscines (suborder Tyranni). In contrast, this standard approach related to taxonomic groups of hosts is incompatible with the presence of the genotypes of *I. sporophilae* in this monophyletic group; since this coccidian is a parasite of buffy-fronted seedeaters *Sporophila frontalis* (Verreaux, 1869) and uniform finches *Haplospiza unicolor* Cabanis, 1851, which are passerines of the family Thraupidae and suborder Passeri. Thus, this phylogenetic analysis maintains the assumption raised in Rodrigues et al. [17] that this monophyletic group is related with coccidia of neotropical birds, and not necessarily related to taxonomic groups of hosts.

Finally, based on the morphological and molecular features described above, *I. borbai* is considered as new to science and the first coccidian species recorded from a gnateater (Conopophagidae).

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## References

1. Berto BP, Lopes CWG (2013) Distribution and dispersion of Coccidia in Wild Passerines of the Americas. In: Ruiz L, Iglesias L (eds) Birds: evolution and behavior, breeding strategies, migration and spread of disease. Nova Science Publishers, New York, pp 47–66
2. Berto BP, McIntosh D, Lopes CWG (2014) Studies on coccidian oocysts (Apicomplexa: Eucoccidiorida). Revista Brasileira de Parasitologia Veterinária 23:1–1. https://doi.org/10.1590/S1984-29612014001
3. Berto BP, Lopes BD, Melinski RD, Souza A, Ribas C, Abreu F, Ferreira I, Lopes CWG (2014) Coccidial dispersion across *trans*- and *cis*-Andean antbirds (Passeriformes: Thamnophilidae): *Isospora sagittulae* McQuistion and Capparella, 1992 (Apicomplexa: Eimeriidae) from non-sympatric hosts. Can J Zool 92:383–388. https://doi.org/10.1139/cjz-2013-0277

4. BirdLife International (2016) The IUCN red list of threatened species. http://www.iucnredlist.org. Accessed 23 Sep 2018
5. Dolnik OV, Palinauskas V, Bensch S (2009) Individual oocysts of *Isospora* (Apicomplexa: Coccidia) parasites from avian feces: from photo to sequence. J Parasitol 95:169–174. https://doi.org/10.1645/GE-1873.1
6. Dolnik OV, Bensch S (2009) *Isospora hypoleucae* sp. n. (Apicomplexa: Eimeriidae), a new coccidian parasite found in the Pied Flycatcher (*Ficedula hypoleuca*). Parasitology 136(8):841–845. https://doi.org/10.1017/s0031182009006131
7. Dolnik OV, Dolnik VR, Bairlein F (2010) The effect of host foraging ecology on the prevalence and intensity of coccidian infection in wild passerine birds. Ardea 98:97–104. https://doi.org/10.5253/078.098.0112
8. Duszynski D, Wilber P (1997) A guideline for the preparation of species descriptions in the Eimeriidae. J Parasitol 83:333–336. https://doi.org/10.2307/3284470
9. ICMBIO (2018) Parque Nacional do Itatiaia. http://www.icmbio.gov.br/parnaitatiaia. Accessed 23 Sep 2018
10. ICZN (2012) International Commission on Zoological Nomenclature: amendment of articles 8, 9, 10, 21 and 78 of the International Code of Zoological Nomenclature to expand and refine methods of publication. Zookeys 219:1–10. https://doi.org/10.3897/zookeys.219.3994
11. McQuiston TE, Capparella A (1992) *Isospora sagittulae*, a new coccidian parasite (Apicomplexa: Eimeriidae) from the spotted antbird (*Hylophylax naevioides*). Trans Am Microscop Soc 111:365–368. https://doi.org/10.2307/3226711
12. Piacentini VQ, Aleixo A, Agne CE, Maurício GN, Pacheco JF, Bravo GA, Brito GRR, Naka LN, Olmos F, Posso S, Silveira LF, Betini GS, Carrano E, Franz I, Lees AC, Lima LM, Pioli D, Schunck F, Amaral FR, Bencke GA, Cohn-Haft M, Figueiredo LFA, Straube FC, Cestari E (2015) Annotated checklist of the birds of Brazil by the Brazilian Ornithological Records Committee/Lista comentada das aves do Brasil pelo Comitê Brasileiro de Registros Ornitológicos. Revista Brasileira de Ornitologia 23:90–298
13. Sick H (1997) Ornitologia Brasileira. Nova Fronteira, Rio de Janeiro, 862 p
14. Silva LM, Rodrigues MB, do Lopes BB, Berto BP, Luz HR, Ferreira I, Lopes CWG (2016) A new coccidian, *Isospora parnaitatiaiensis* n. sp. (Apicomplexa, Eimeriidae), from the white-shouldered fire-eye *Pyriglena leucoptera* (Passeriformes, Thamnophilidae) from South America. Parasitol Res 115:745–749. https://doi.org/10.1007/s00436-015-4798-z
15. Silva-Carvalho LM, Pastura DGN, Gomes JV, Siqueira PB, Rodrigues MB, Lima VM, Berto BP (2018) *Isospora lopesi* n. sp. (Protozoa: Apicomplexa: Eimeriidae) from the eastern white-throated spadebill *Platyrinchus mystaceus* Vieillot (Passeriformes: Tyranni: Tyrannidae) in South America. Syst Parasitol 95:455–463. https://doi.org/10.1007/s11230-018-9795-z
16. Silva-Carvalho LM, Pastura DGN, Rodrigues MB, Gomes JV, Oliveira MS, Siqueira PB, Oliveira JLG, Soares SS, Oliveira AA, Lima VM, Ferreira I, Berto BP (2018) *Isospora sagittulae* McQuistion & Capparella (1992 (Apicomplexa: Eimeriidae) from antbirds (Passeriformes: Thamnophilidae) in the Amazon and Atlantic Forest of Brazil: with notes on its distribution and dispersion in the Neotropical region. Parasitol Res 117:2635–2641. https://doi.org/10.1007/s00436-018-5955-y
17. Rodrigues MB, Oliveira JLG, Silva-Carvalho LM, Pastura DGN, Gomes JV, Oliveira MS, Siqueira PB, Oliveira AA, Lima VM, Ferreira I, Berto BP (2019) The vulnerable *Sporophila frontalis* (Verreaux) and *Haplospiza unicolor* Cabanis as new hosts for *Isospora sporophilae* Carvalho-Filho, Meireles, Ribeiro & Lopes, 2005 (Eimeriidae) in Brazil. Syst Parasitol 96:423–431. https://doi.org/10.1007/s11230-019-09859-7
18. Tamura K, Dudley J, Nei M, Kumar S (2007) MEGA4: molecular evolutionary genetics analysis (MEGA) software version 4.0. Mol Biol Evol 24:1596–1599. https://doi.org/10.1093/molbev/msm092
19. Yang R, Brice B, Elliot A, Ryan U (2015) *Isospora serinuse* n. sp. (Apicomplexa: Eimeriidae) from a domestic canary (*Serinus canaria* forma domestica) (Passeriformes: Fringillidae) in Western Australia. Exp Parasitol 159:59–66. https://doi.org/10.1016/j.exppara.2015.08.020




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