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Occurrence of eggs and oocysts of gastrointestinal parasites in passerine birds kept in captivity in Para State, Brazil

Ocorrência de ovos e oocistos de parasitos gastrointestinais em aves Passeriformes mantidas em cativeiro no estado do Pará, Brasil

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ABSTRACT

The objective of this study was to detect helminth eggs and protozoan oocysts in samples of feces from birds of the order Passeriformes in Para State, Brazil. Fecal samples were collected individually from 403 passerine birds seized and kept in captivity in Para State. Samples were processed by the double centrifugation technique in saturated sucrose solution and the coccidial oocyst-positive samples were submitted to sporulation in potassium dichromate 2.0%. Helminth eggs and/or protozoan oocysts were observed in 43.18% (174/403) of the fecal samples examined. Coccidial oocysts were detected in 93.68% (163/174) of the positive samples, whereas helminth eggs were observed in 10.34% (18/174) of the positive samples. Oocyst sporulation occurred in 43.56% (71/163) of the samples, and only *Isospora* spp. oocysts were detected. Nematode eggs of the superfamilies Trichostrongyloidea (4.60%; 8/174), Ascaridoidea (0.57%; 1/174), and Trichuroidea (0.57%; 1/174) were diagnosed in the positive samples. Cestoda eggs were diagnosed in 2.87% (5/174), whereas Trematoda eggs were detected in 2.30% (4/174) of positive samples. Passerine birds seized and kept in captivity in the visited local presented parasitism by intestinal helminths and protozoan, with a predominance of infection with coccidia of the gender *Isospora*.

Key words: passerine birds, endoparasites, coccidia, helminths, Brazilian Amazon.

RESUMO

O objetivo do presente estudo foi detectar ovos de helmintos e oocistos de protozoários em amostras de fezes de aves da ordem Passeriformes no estado do Pará, Brasil. Amostras de fezes foram coletadas individualmente de 403 aves Passeriformes oriundas de apreensão e mantidas em cativeiro no estado do Pará. As amostras foram processadas usando a técnica

de dupla centrifugação em solução saturada de sacarose e as amostras positivas para oocistos de coccídios foram submetidas à esporulação em dicromato de potássio 2,0%. Ovos de helmintos e/ou oocistos de protozoários foram observados em 43,18% (174/403) das amostras fecais examinadas. Oocistos de coccídios foram detectados em 93,68% (163/174) das amostras positivas, enquanto que ovos de helmintos foram observados em 10,34% (18/174). A esporulação de oocistos ocorreu em 43,56% (71/163) das amostras, e somente oocistos de *Isospora* spp. foram detectados. Ovos de nematódeos das Superfamílias Trichostrongyloidea (4,60%; 8/174), Ascaridoidea (0,57%; 1/174) e Trichuroidea (0,57%; 1/174) foram diagnosticados nas amostras positivas. Ovos de Cestoda foram diagnosticados em 2,87% (5/174), enquanto que ovos de Trematoda foram detectados em 2,30% (4/174) das amostras positivas. Aves Passeriformes oriundas de apreensão e mantidas em cativeiro nas áreas visitadas estavam parasitadas por helmintos e protozoários, predominando a infecção por coccídios do gênero *Isospora*.

Palavras-chave: pássaros, endoparasitos, coccídios, helmintos, Amazônia Brasileira.

INTRODUCTION

Brazil, Colombia, and Peru are considered to have the largest avian fauna diversity in the world, and South America was thus entitled the “Continent of Birds” (PIACENTINI et al., 2015; REMSEN et al., 2016). The most recent list of Brazilian ornithological fauna included 1,919 species (PIACENTINI et al., 2015), and 993 species are recorded in the Brazilian Amazon Biodiversity Census (MPEG, 2016). Among

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birds, the order Passeriformes has a great importance due to its diversity, which corresponds to about 56% of total species in Brazil (PIACENTINI et al., 2015).

Parasitic diseases are among the major health problems affecting passerine birds. Such diseases can cause subclinical alterations or even death of birds subjected to stress, recently introduced or kept in high-population density precincts. Although little is known about parasitic infection rates of passerines in different regions of Brazil, it is known that helminthiasis and coccidiosis often affect birds in captivity (FIGUEROA-LYRA et al., 2002; BERTO et al., 2011). Therefore, diagnostic studies on endoparasites both extend knowledge about the epidemiology of gastrointestinal diseases and serve as a basis to implement an adequate health management for wild birds kept in captivity (GUIMARÃES, 2007). Therefore, the objective of this study was to detect helminth eggs and protozoan oocysts in samples of feces from birds of the order Passeriformes seized and kept in captivity in the Para State, Brazil.

MATERIALS AND METHODS

Animals and study area

Four hundred and three fecal samples were collected from birds of the order Passeriformes of the families Cardinalidae (n=3), Corvidae (n=1), Cotingidae (n=3), Estrildidae (n=1), Icteridae (n=19), Pipridae (n=1), Thraupidae (n=352), and Turdidae (n=23). Birds were identified based on the Field Guide of Birds in Brazil (SIGRIST, 2009) and List of Birds in Brazil (PIACENTINI et al., 2015). Passerine birds were seized by the Brazilian Institute of the Environment and Natural Renewable Resources in the interior and capital of Para State, Brazil. These birds were transported to the Mangal das Garças Environmental Park (S 01°27'56.0", W 48°30'18"), in the city of Belém, and Adhemar Monteiro Zoo and Botanical Park (S 1°44'45.5", W 47°03'27.1"), Capitão Poço city, Para State. Fecal samples were also obtained from the Albatroz commercial breeding (S 01°27'19.9", W 48°27'53.6") in Belém city.

Collection and processing of biological samples

From March 2009 to January 2012, 403 fecal samples from birds were individually collected from the cages floor early in the morning, placed in labeled plastic containers, and processed by the double centrifugation technique in saturated sucrose solution (MONTEIRO, 2010). After centrifugation (Excelsa® II 206 BL centrifuge), the supernatant was analyzed between slide and cover slip in an

optical microscope (Nikon® 50i; objectives: 10x, 40x, and 100x). Coccidial oocyst-positive samples were immersed in potassium dichromate (K₂Cr₂O₇) 2% solution and maintained at room temperature for seven days or until oocyst sporulation (DUSZYNSKI & WILBER, 1997). After analysis, all results were sent to the respective sample collection locations for treatment and adequacy of sanitary management.

RESULTS AND DISCUSSION

Occurrence of helminth and oocyst eggs of gastrointestinal protozoans was observed in passerine birds kept in captivity in the Brazilian Amazon (Table 1; Figure 1A-H), including in species such as *Cyanoloxia rothschildii*, *Paroaria gularis*, and *Tangara episcopus*, whose distribution is restricted to the Amazon biome (SIGRIST, 2009). Infections caused by gastrointestinal parasites are considered a risk to avian health, especially those under stress conditions, inadequate management, and poor nutrition in captivity (FIGUEIROA-LYRA et al., 2002; GUIMARÃES, 2007). Therefore, recently seized or imported birds deserve a closer monitoring regarding anti-parasitic diagnosis and treatment, as these birds are more susceptible to these infections (OGLESBEE, 2008).

Helminth eggs and/or protozoan oocysts were observed in 43.18% (174/403) of the fecal samples (Table 1). Coccidial oocysts (Figure 1A-C) were detected in 93.68% (163/174) of the positive samples, whereas helminth eggs (Figure 1D-H) were observed in 10.34% (18/174) of these samples. Oocyst sporulation occurred in 43.56% (71/163) of samples and only *Isospora* spp. oocysts were detected (Table 1; Figure 1A-C). Nematode eggs were diagnosed in positive samples of the superfamilies Trichostrongyloidea (4.60%; 8/174; Figure 1D), Ascaridoidea (0.57%; 1/174; Figure 1E), and Trichuroidea (0.57%; 1/174; Figure 1F). Cestoda eggs (Figure 1G) were diagnosed in 2.87% (5/174), whereas Trematoda eggs (Figure 1H) were detected in 2.30% (4/174) of positive samples.

Regarding parasitism relative to the passerine species, nematode eggs of the superfamily Trichostrongyloidea were observed in fecal samples from *S. angolensis* (1.06%; 3/282), *Sporophila maximiliani* (5.26%; 1/19), *Sporophila americana* (28.57%; 2/7), *T. episcopus* (33.33%; 1/3), and the only sample from *Sporophila plumbea*. Nematode eggs of the superfamily Ascaridoidea (4.35%; 1/23) were observed and helminth eggs of the superfamily Trichuroidea (4.35%; 1/23) were also diagnosed in *Turdus* sp. fecal samples. Typical Cestoda eggs were observed in fecal samples from *Paroaria gularis*

Table 1 - Results of coproparasitological analysis in species of passerine birds.

Families	Taxon	Number of samples	-----Samples positive for oocysts and/or helminth eggs (%)-----			
			Coccidial oocysts (sporulated and not sporulated)	Sporulated <i>Isospora</i> spp. oocysts	Helminth eggs	Total of positive samples
<i>Thraupidae</i>	<i>Sporophila angolensis</i>	282	37.23% (105/282)	15.6% (44/282)	1.06% (3/282)	37.59% (106/282)
	<i>Sporophila maximiliani</i>	19	63.15% (12/19)	42.1% (8/19)	5.26% (1/19)	68.42% (13/19)
	<i>Sporophila lineola</i>	8	62.5% (5/8)	0% (0/8)	0% (0/8)	62.5% (5/8)
	<i>Sporophila americana</i>	7	28.57% (2/7)	28.57% (2/7)	28.57% (2/7)	57.14% (4/7)
	<i>Paroaria gularis</i>	7	57.14% (4/7)	28.57% (2/7)	14.29% (1/7)	71.43% (5/7)
	<i>Tangara episcopus</i>	5	60% (3/5)	60% (3/5)	0% (0/5)	60% (3/5)
	<i>Tangara cayana</i>	4	50% (2/4)	50% (2/4)	25% (1/4)	75% (3/4)
	<i>Sporophila minuta</i>	4	25% (1/4)	0% (0/4)	0% (0/4)	25% (1/4)
	<i>Dolospingus fringilloides</i>	4	100% (4/4)	0% (0/4)	0% (0/4)	100% (4/4)
	<i>Saltator similis</i>	3	33.33% (1/3)	0% (0/3)	0% (0/3)	33.33% (1/3)
	<i>Lanio surinamus</i>	2	100% (2/2)	50% (1/2)	0% (0/2)	100% (2/2)
	<i>Sicalis flaveola</i>	2	100% (2/2)	100% (2/2)	50% (1/2)	100% (2/2)
	<i>Sporophila schistacea</i>	1	100% (1/1)	0% (0/1)	0% (0/1)	100% (1/1)
	<i>Sporophila plumbea</i>	1	100% (1/1)	0% (0/1)	100% (1/1)	100% (1/1)
	<i>Cyanicterus cyanicterus</i>	1	100% (1/1)	0% (0/1)	0% (0/1)	100% (1/1)
	<i>Cissopis leverianus</i>	1	0% (0/1)	0% (0/1)	0% (0/1)	0% (0/1)
	<i>Saltator maximus</i>	1	0% (0/1)	0% (0/1)	0% (0/1)	0% (0/1)
<i>Turdidae</i>	<i>Turdus</i> spp.	23	30.43% (7/23)	8.7% (2/23)	26.09% (6/23)	47.83% (11/23)
<i>Icteridae</i>	<i>Icterus jamacaii</i>	5	20% (1/5)	20% (1/5)	0% (0/5)	20% (1/5)
	<i>Gymnomystax mexicanus</i>	4	50% (2/4)	25% (1/4)	0% (0/4)	50% (2/4)
	<i>Gnorimopsar chopi</i>	3	66.66% (2/3)	33.33% (1/3)	33.33% (1/3)	66.66% (2/3)
	<i>Icterus chrysiocephalus</i>	3	66.66% (2/3)	33.33% (1/3)	0% (0/3)	66.66% (2/3)
	<i>Cacicus cela</i>	2	50% (1/2)	0% (0/2)	0% (0/2)	50% (1/2)
	<i>Molothrus oryzivorus</i>	1	0% (0/1)	0% (0/1)	0% (0/1)	0% (0/1)
	<i>Cacicus haemorrhous</i>	1	0% (0/1)	0% (0/1)	0% (0/1)	0% (0/1)
<i>Cardinalidae</i>	<i>Cyanoloxia rothschildii</i>	3	33.33% (1/3)	0% (0/3)	33.33% (1/3)	66.66% (2/3)
<i>Estrildidae</i>	<i>Estrilda astrild</i>	1	0% (0/1)	0% (0/1)	0% (0/1)	0% (0/1)
<i>Pipridae</i>	<i>Pipra fasciicauda</i>	1	0% (0/1)	0% (0/1)	0% (0/1)	0% (0/1)
<i>Cotingidae</i>	<i>Procnias averano</i>	3	33.33% (1/3)	33.33% (1/3)	0%	33.33% (1/3)
<i>Corvidae</i>	<i>Cyanocorax cyanopogon</i>	1	0% (0/1)	0% (0/1)	0% (0/1)	0% (0/1)
TOTAL		403	40.45% (163/403)	17.62% (71/403)	4.47% (18/403)	43.18% (174/403)

(14.29%; 1/7), *Turdus* spp. (13.04%; 3/23), and *Tangara cayana* (25%; 1/4), and trematode eggs were diagnosed in samples from *Turdus* sp. (8.7%; 2/23), *Gnorimopsar chopi* (33.33%; 1/3), and *Sicalis flaveola* (50%; 1/2).

Regarding the type of parasitism, simple parasitism was observed in 95.98% (167/174) of the positive fecal samples. Coparasitism by coccidial oocysts and helminth eggs was observed in 4.02% (7/174) of the parasitized passerine birds. This coparasitism was diagnosed in fecal samples from *Turdus* spp. (n=2), *S. angolensis* (n=2), *S. flaveola* (n=1), *G. chopi* (n=1), and *S. plumbea* (n=1). Parasitism by more than one class of helminths was

diagnosed in one fecal sample from *Turdus* sp., and nematode (superfamily Trichuroidea) and trematode eggs were observed.

Most infected birds presented simple parasitism by coccidial oocysts, with a predominance of *Isospora* spp. oocysts, similar to the data obtained by BERTO et al. (2011), who showed that most coccidia infecting passerine birds are species of the genus *Isospora*, but *Eimeria* species also occurred although with a lower frequency. Our results are also consistent with observations in *Saltator similis* in captivity, in Valença, Rio de Janeiro State, Brazil, and according to VASCONCELOS et al. (2013) *Isospora*

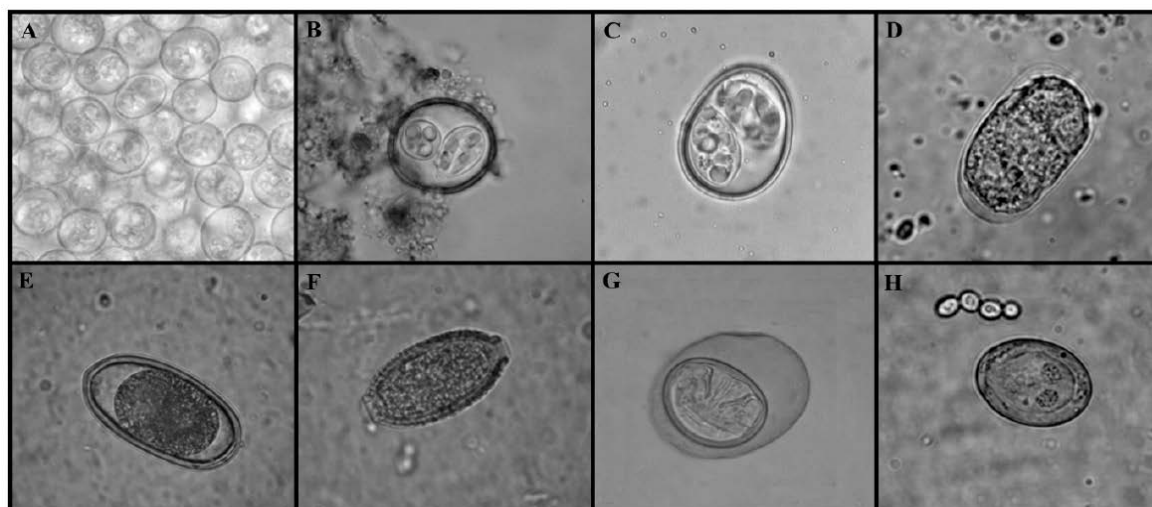


Figure 1 - Coccidial oocysts and helminth eggs diagnosed in fecal samples from passerine birds: A) *Isospora* spp. oocysts in feces from *Sporophila angolensis* (40x); B) *Isospora* sp. oocyst in feces from *Icterus jamacaii* (100x); C) *Isospora* sp. oocyst in feces from *Tangara episcopus* (100x); D) Nematode egg of the superfamily Trichostrongyloidea in feces from *Sporophila maximiliani* (40x); E) Nematode egg of the superfamily Ascaridoidea in feces from *Turdus* spp. (40x); F) Nematode egg of the superfamily Trichuridoidea in feces from *Turdus* spp. (40x); G) Cestoda egg in feces from *Turdus* spp. (40x); H) Trematoda egg in feces from *Sicalis flaveola* (40x).

spp. oocysts (90%) were detected in animals with moderate to high infection rate.

Intestinal coccidial infections can also be considered frequent in free-living passerine birds. COSTA et al. (2010) detected coccidial oocyst-positive fecal samples (58.66%) in birds captured in Seropédica city, Rio de Janeiro State, and most birds were of the order Passeriformes. These data are similar to those obtained in present study with captive animals, in which high occurrence of infection by coccidial oocysts was observed.

In the present study, a large number of *Isospora* spp. oocysts (Figure 1A) was detected in some fecal samples although helminth eggs and protozoan oocysts quantification was not performed. The high parasitism as observed in these animals may have occurred due to stress during seizure and captivity, as birds are usually kept in small cages. According to GILL & PAPERNA (2008), free-living birds are able to control coccidial infection, not manifesting clinical signs suggestive of the disease. However, these birds succumb to infection and present changes in the social and reproductive behavior, diarrhea, weight loss, and even death when subjected to capture stress or captivity.

Although helminth eggs have occurred in few fecal samples, a diversity these eggs was observed. In *Turdus* spp., e.g., trematode, cestode, and nematode eggs of the superfamilies Ascaridoidea and Trichuroidea were found. In Pernambuco State, Brazil, FIGUEIROA-LYRA et al. (2002) conducted a survey of endoparasites

in captive wild birds using sedimentation and flotation techniques and reported positive samples (46.7%; 320/685). In passerine birds of the family *Icteridae*, the authors diagnosed Cestoda eggs, whereas in birds of the family *Cotingidae* parasitism was not observed. In Para State, Cestoda eggs were also observed in fecal samples from the icteride *G. chopi*, and presence of *Isospora* sp. oocysts was also detected in samples from the cotingide *Procnias averano*.

It was also observed that the species *S. maximiliani* and *P. averano*, which are threatened with extinction in the Brazilian wildlife (MMA, 2003), were reported to be infected with *Isospora* spp. oocysts, whereas helminth eggs from the superfamily Trichostrongyloidea were found only in *S. maximiliani*. The results of the present study reinforce the importance of diagnosis for the control of coccidiosis and helminthiasis in birds seized and/or kept in captivity. Based on knowledge about these infections, is possible to establish appropriate treatment and management, in order to improve the sanitary conditions and survival of captive birds.

CONCLUSION

Passerine birds seized and kept in captivity in the visited cities presented parasitism by intestinal helminths and protozoan, with a predominance of infection with coccidia of the gender *Isospora*.

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BIOETHICS AND BIOSSECURITY COMMITTEE APPROVAL

Authors' declaration

We, authors of the article entitled "Occurrence of eggs and oocysts of gastrointestinal parasites in passerine birds kept in captivity in Para State, Brazil", declare for due purpose that the project that originated the study data was not submitted to evaluation by the Ethics Committee, Federal University of Pará. However, we are aware of the content of the National Council for Control of Animal Experimentation (CONCEA; "http://www.mct.gov.br/index.php/content/view/310553.html") resolutions on animal involvement.

Thus, the authors are fully responsible for the data presented here and are available to answer any questions by the relevant authorities.

Authorization for sample collection

This study was approved by the System for Authorization and Information on Biodiversity from the Chico Mendes Institute for Biodiversity (SISBIO/ICMBIO, protocol 30710-1).

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