



Occurrence of Endoparasites in Wild Songbirds from Illegal Wildlife Trade

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ABSTRACT

Trafficking in wild animals represents one of the main illicit activities in the world. In Brazil, among the main trafficked species the songbirds of the Passeriform Order stand out. The distress to which these birds are exposed, especially the newly caught ones, and the unhealthy conditions to which they are subjected, cause, in addition to the damage to well-being, an important drop in resistance, which exacerbates infectious and parasitic conditions. Clinical assessments and coproparasitological examinations were carried out using the Willis-Mollay method on 102 passerines from apprehension in western Paraná. Of the total number of animals evaluated, 76% (78/102) had hyperkeratosis in the pelvic limbs, 55% (56/102) were infested with mites on remiges and 54% (52/102) had changes in warping. The results indicated 94% (96/102) of positivity for endoparasites, of which 96% (92/96) were for coccidian oocysts, 1% (1/96) for tenia eggs and 3% (3/96) for mixed infection (coccid oocysts and tenia eggs). These findings corroborate that the poor conditions of hygiene, nutrition and the high stocking density observed in illegal housing can predispose birds to more severe clinical manifestations.

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Introduction

Trafficking in wild animals represents one of the main illicit activities in the world, along with trafficking in arms and drugs. It is estimated that about 38 million animal specimens are removed annually from Brazilian ecosystems (RENCTAS, 2017). Birds represent 56.24% of the total number of wild animals apprehended in Brazil, and among the different orders of this class, passerine species stand out, which have a wide geographical distribution, diversity and are especially trafficked due to their beauty and song (CBRO, 2015; RENCTAS, 2017; Ribeiro and Silva, 2007).

Inadequate management such as high density, poor hygiene and nutrition, poor transport conditions and joint accommodation of naturally antagonistic species are often observed in illegal poultry breeding and generate a state of distress for the animals, with consequent influence on the immune system, which favors the emergence of infections (Kuhlman and Martin, 2010).

Often the laboratory diagnosis of these infections can bring obstacles, in view of the enormous parasitological di-

versity and the scarcity of data on parasitic fauna in wild birds. This diversity found in wild bird species is usually attributed to the adaptations that the parasite needs to complete its life cycle, in which it can present successive transmissions through two or more hosts (Littlewood *et al.*, 2015).

Pathogenic parasites can pose a threat to programs for the management and recovery of animal populations, so the identification of parasitic fauna is necessary in protocols for reintroduction or release into the wild (Catão-Dias, 2003). The objective of this work was to present the frequency of endoparasites in fecal samples from wild birds from a large arrest operation in the western region of Paraná State (PR), Brazil.

Materials and methods

In a joint action by the Water and Earth Institute (IAT) and the Environmental Police Battalion, 262 passerines were apprehended in a residence in the municipality of Marechal Cândido Rondon / PR. The birds were transported and sent to veterinary medical care at the Veterinary Hospital, where they were individually identified. Of these, 102 passerines, distributed among the families Icteridae (07), Fringilidae (07), Thraupidae (79) and Turdidae (09) were housed in individual cages and the remaining 160 birds were kept in collective cages, since they were also housed in this way at the place of

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arrest. The data for these 160 birds were not listed in this article due to the impossibility of carrying out the individual assessment. In the cages received from apprehension, was observed a high accumulation of excreta, probably due to the difficulty of the offender to properly handle the high quantity of animals.

Upon being received, all passerines were housed in a large room with adequate air circulation, for subsequent replacement of drinking fountains and feeders, and cleaning of the cages. In order to optimize handling, the specimens were grouped according to eating habits and the cages were placed without the bottom under a plastic tarpaulin, which allowed for quick cleaning by simply replacing the dirty tarpaulin with a clean tarpaulin. During the hospitalization period, the birds were fed seeds, extruded concentrate, and fruits, in addition to water ad libitum. The water and fruit exchange took place at least twice a day.

For clinical evaluation, the birds were physically restrained and submitted to general anesthesia with isoflurane (dose-effect) in a universal vaporizer using an anesthetic mask and monitored via auscultation or with the use of vascular Doppler. Body score, condition of warping and presence of ectoparasites were evaluated. For coproparasitological assessments, individual collections were performed twice a day for three consecutive days of the excreta present under each cage. The samples were collected with the aid of a steel spatula and the collected material was placed in a collecting flask and stored under refrigeration (4°C to 8°C), until two grams of excreta were obtained, which were processed within 12 hours. The material was subjected to the simple flotation technique in saturated sodium chloride solution (density ≥ 1.18 g / cm³) (Willis-Mollay) (Willis, 1921) for semi-quantitative analysis. The degree of infection by these agents was classified according to Table 1.

This study was approved by the Ethics Committee on Animal Use of the Federal University of Paraná – Setor Palotina (n. 11/2021).

Table 1. Meaning of the degree of infection by the Willis-Mollay method (simple fluctuation in saturated solution).

Degree of infection	Meaning
A cross (+)	One oocyst / egg per slide
Two crosses (++)	Two to five oocysts / eggs per field
Three crosses (+++)	Six to 10 oocysts / eggs per field
Four crosses (++++)	More than 10 oocysts / eggs per field

Results

In the clinical evaluation of the 102 specimens, it was ob-

served that 76.47% (78/102) had hyperkeratosis in the pelvic limbs due to *Knemidocoptes scabies*, 54.90% (56/102) were infested with mites in primary and secondary remiges, 50.98% (52/102) presented alteration in the warping (gnawed, broken feathers and / or absence of some remiges or retrills) and 5.88% (6/102) presented onychogryphosis. Regarding the body score, 5.88% (6/102) of the animals had cachexia, 74.5% (76/102) had an adequate body score and 19.6% (20/102) were obese.

Only six individuals, all of the species *Sicalis flaveola*, had a negative result in the coproparasitological exam. Of the total positive samples (Table 2), 95.8% (92/96) of the birds presented positivity for coccidian oocysts, 1% (1/96) presented tenia eggs and 3.1% (3/96) presented mixed infection, both by coccid oocysts and by tenia eggs (Fig. 1). Of the birds' positive for coccidian oocysts, 14.73% (14/95), 45.26% (43/95), 14.73% (14/95) and 25.26% (24/95) presented levels of infection compatible with one, two, three and four crosses, respectively. There was no variation in results between different species.



Figure 1. Tenia egg (asterisk) and coccid oocysts (arrows) in fecal passerine sample (400x magnification). Source: Author.

Discussion

Coccids were the most frequently found parasites in the analyzes, which is in line with the results obtained by Magalhães-Matos *et al.* (2016), who observed 99% (95/96) of the positive samples in the evaluated passerines, which makes sense, since they are organisms considered ubiquitous and parasitize the vast majority of known bird species (Duszynski *et al.*, 2000) and often affect those in captivity (de Freitas *et al.*, 2002). These protozoa are mandatory intracellular parasites (Knight *et al.*, 2018) that invade host enterocytes in the form

Table 2. Percentage of positive samples in 96 individuals of the order Passeriform from trafficking, using the Willis-Mollay method.

Family	Scientific name	Number of citizens	Coccidia	Tenia	Mixed Infection
Fringillidae	<i>Euphonia chlorotica</i>	n.=7	86% (6/7)	0% (0/7)	14% (1/7)
	<i>Gnorimopsar chopi</i>	n.=2	100% (2/2)	0% (0/2)	0% (0/2)
Icteridae	<i>Icterus pyrrhopterus</i>	n.=4	100% (4/4)	0% (0/4)	0% (0/4)
	<i>Agelaioides badius</i>	n.=1	100% (1/1)	0% (0/1)	0% (0/1)
Thraupidae	<i>Sicalis flaveola</i>	n.=48	98% (47/48)	0% (0/48)	2% (1/48)
	<i>Dacnis cayana</i>	n.=1	100% (1/1)	0% (0/1)	0% (0/1)
	<i>Pipraeidea bonariensis</i>	n.=7	86% (6/7)	0% (0/7)	14% (1/7)
	<i>Saltator similis</i>	n.=7	100% (7/7)	0% (0/7)	0% (0/7)
	<i>Saltator fuliginosus</i>	n.=1	100% (1/1)	0% (0/1)	0% (0/1)
	<i>Sporophila caeruleascens</i>	n.=1	100% (1/1)	0% (0/1)	0% (0/1)
	<i>Tangara sayaca</i>	n.=8	100% (8/8)	0% (0/8)	0% (0/8)
Turdidae	<i>Turdus amaurochalinus</i>	n.=1	100% (1/1)	0% (0/1)	0% (0/1)
	<i>Turdus leucomelas</i>	n.=8	87% (7/8)	13% (1/8)	0% (0/8)
Total		n.=96	96% (92/96)	1% (1/96)	3% (3/96)

Table 3. Relationship between body score in 95 birds and degree of infection with coccidian oocysts.

Body score	Degree of infection by coccidian oocysts			
	(+)	(++)	(+++)	(++++)
1 (n=6)	0% (0/6)	67% (4/6)	0% (0/6)	33% (2/6)
2 (n=5)	20% (1/5)	60% (3/5)	0% (0/5)	20% (1/5)
3 (n=27)	15% (4/27)	41% (11/27)	18% (5/27)	26% (7/27)
4 (n=37)	16% (6/37)	46% (17/37)	11% (4/37)	27% (10/37)
5 (n=20)	15% (3/20)	40% (8/20)	25% (5/20)	20% (4/20)

of sporozoites, previously released from the oocyst in the gastrointestinal tract, characterized by oro-fecal transmission (Shirley *et al.*, 2005; Marietto-Gonçalves *et al.*, 2009; Knight *et al.*, 2018).

In this study, it was not possible to obtain evident correlations between low body score and high degree of coccid infection (Table 3), which would be expected since one of the main consequences of coccid infection is the reduction of nutrient absorption (Hörak *et al.*, 2004). However, the observed can be partially explained by the wide variation of pathogenicity in avian coccidiosis, such as the infective dose and the species of parasite involved (Knight *et al.*, 2018). In situations of immunosuppression, the asymptomatic bird may present with an aggravation of the condition with the expression of clinical feathers, which may include goosebumps, drowsiness, loss of appetite, apathy, prostration, diarrhea, weight loss, cachexia, and reproductive problems (Marietto-Gonçalves *et al.*, 2009). Thus, it is possible to observe a wide range of clinical manifestations, and high mortality is especially associated with the visceral spread of protozoa (Box, 1977). Due to the conditions described above, it is reasonable to infer that several birds died even before seizure, preventing more precise conclusions about the impact of coccidiosis in this population.

Tapeworms are Cestoda class helminths that attach to the villi of the host's digestive system through the scolex, mainly in the ventricle, intestine and, occasionally, the ureters (Urquhart *et al.*, 1998). The presence of eggs, pregnant proglottids or fragments of cestodes in the feces in the host is sufficient for the diagnosis of this infection. The frequency observed in the present study was similar to previous findings, including in relation to the gender of the host (Magalhães-Matos *et al.*, 2016). However, it was not possible to identify the *Tenia* species in terms of order, family and gender, as this requires macroscopic studies of the adult specimen such as the evaluation of the scolex and reproductive system morphology (McLaughlin, 2008). Young birds are more sensitive to these parasites than adults, however, the clinical manifestations for both are nonspecific and can include hyporexia, polydipsia and anemia and can worsen over time leading to anorexia, head spasms and even death (Marín-Gómez and Benavides-Montano, 2007; McLaughlin, 2008).

Mixed infections with the presence of adult helminths and coccids are of medical-veterinary importance, as they cause intestinal disorders such as enteritis and hemorrhage (Marín-Gómez and Benavides-Montano, 2007) and have already been reported in the literature with captive wild birds (Magalhães-Matos *et al.*, 2016). However, in this research, no specific clinical changes were observed in animals with the presence of coccid oocysts and eggs of *Tenia*. As in the work of Santos *et al.* (2015), most birds showed good body condition and no clinical manifestation of the infection. In this context, several factors can influence this result, such as resilience, parasite-host relationship, and parasitic load, which make the bird more or less susceptible to infection.

The positive passerines were treated with antiparasitic and after negativity in the coproparasitological samples, the animals were housed in a nursery for flight rehabilitation and later destined or returned to nature as assessed by the Water

and Earth Institute (IAT).

The unhealthy structural and sanitary housing conditions are directly related to the identified parasites. Mitigating measures for these parasites are based on a balanced diet, water quality, general hygiene care in the creation and maintenance of birds, in addition to training the maintainers and periodic examinations of the flock's excreta. These conditions, commonly, do not exist in illegal captivity.

Periodic cleaning of cages, drinking fountains and feeders with mild soap and submerging them in diluted sodium hypochlorite solution are recommended measures (Carneiro *et al.*, 2011). In addition, these same authors recommend the proper disposal of perches, so that they do not remain under the sources of water and food, which would prevent contamination of the latter by excreta. Newly incorporated birds are more likely to have parasitic infections due to arrival stress (Snak *et al.*, 2014). In illegal captivity, as in the case of this research, it is believed that the practice of a quarantine program, an important measure in the creation and maintenance of birds, was not carried out.

Conclusion

The obtained results inform about the parasitic fauna of these wild birds kept in this illegal captivity and demonstrate that even without evident clinical presentation there is elimination and dissemination of the parasitic agents. The maintenance of an excessive number of specimens coupled with precarious management conditions favors immunosuppression in birds and various infections, including those of parasitic origin. Given the asymptomatic and / or subclinical nature of these infections, clinical and laboratory tests are essential for an accurate assessment of the health status of individuals before referring them to a wildlife enterprise or even returning them to nature.

Conflict of interest

The authors declare that there is no conflict of interest.

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