# Natural infection by endoparasites among free-living wild animals

## Infecção natural por endoparasitas em animais silvestres de vida-livre

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## Abstract

The objective of this study was to investigate the frequency of occurrence and variety of intestinal parasites among free-living wild animals. Fecal samples from wild mammals and birds at rehabilitation centers in the states of Mato Grosso do Sul and São Paulo were analyzed by sedimentation and flotation-centrifugation methods. Parasite eggs, oocysts, cysts and/or trophozoites were found in 71% of the samples. *Cryptosporidium* sp. oocysts were detected in fecal samples from oncillas (*Leopardus tigrinus*) and scaly-headed parrots (*Pionus maximiliani*). *Giardia* cysts were identified in the feces of a gray brocket (*Mazama gouazoubira*). Among the most common parasites found, there were eggs from *Toxocara cati*, *Toxascaris leonina* and *Ancylostoma tubaeforme*, and from Cestoda. Several *Enterobius* sp. eggs were found in the feces of red howler monkeys (*Alouatta seniculus*). It can be concluded from this study that despite the small number of samples, the diversity of parasites found was noteworthy. Additional information about parasite endofauna in wild animals is needed, since their presence might suggest that there could be proximity to and interactions with domestic animals and/or humans. In addition, further studies on parasites from free-living wild animals are of prime importance for understanding the intensity of anthropic changes in wild environments.

**Keywords:** Wildlife, gastrointestinal parasites, Mato Grosso do Sul, São Paulo.

## Resumo

O objetivo deste trabalho foi pesquisar a frequência de ocorrência e a variedade de parasitas intestinais de animais silvestres de vida livre. Amostras de fezes de mamíferos e aves silvestres de centros de reabilitação dos Estados do Mato Grosso do Sul e São Paulo, foram analisadas pelos métodos de sedimentação e de centrífugo-flutuação. Foram encontrados ovos, oocistos, cistos e/ou trofozoítos de parasitas em 71% das amostras. Oocistos de *Cryptosporidium* sp. foram detectados em amostras de fezes de gato-do-mato-pequeno (*Leopardus tigrinus*) e maritacas (*Pionus maximiliani*). Cistos de *Giardia* foram identificados nas fezes de um veado catingueiro (*Mazama gouazoubira*). Entre os parasitas mais comuns, foram encontrados ovos de *Toxocara cati*, *Toxascaris leonina* e *Ancylostoma tubaeforme*, além de ovos de Cestoda. Vários ovos de *Enterobius* sp. foram encontrados nas fezes de bugio (*Alouatta seniculus*). Neste trabalho concluiu-se que, apesar de pequeno o número de amostras, a diversidade de parasitas encontrados foi relevante. Informações adicionais sobre a endofauna parasitária em animais silvestres são necessárias, pois podem sugerir a interação e proximidade com animais domésticos e/ou o homem. Além disso, maiores estudos sobre os parasitas de animais silvestres de vida livre são primordiais para a compreensão da intensidade de alterações antrópicas no meio silvestre.

**Palavras-chave:** Animais silvestres, parasitas gastrintestinais, Mato Grosso do Sul, São Paulo.

With the advance of agriculture and cattle-raising into natural areas, humans and their domestic animals have recently been coming into greater contact with populations of wild animals in their habitats. This closer contact facilitates the spread of infectious agents and parasites to new hosts and environments, thereby establishing new relationships between hosts and parasites, and new ecological niches in the disease transmission chain (CORRÊA; PASSOS, 2001).

Infection routes are directly related to causal agents, which means that knowledge of the disease transmission chain may provide an opportunity to ascertain how these agents reach new susceptible hosts. Thus, in relation to epidemiological factors, wild

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animals may have an extremely important role in the transmission of zoonoses, both in captivity and in the wild (MARVULO, 2007).

The emergence of infectious diseases with zoonotic potential has dominated research on wildlife pathogens over recent years (McCALLUM; DOBSON, 1995; HOLMES, 1996; DASZAK et al., 2000; RHYAN; SPRAKER, 2010). As a result, not only have studies on the biodiversity and ecology of parasites been neglected, but also efforts to control them have been impaired. The research focus has been directed toward humans and domestic animals. However, there is also a need to obtain greater understanding of how these emerging pathogens interact with sets of organisms living together in wild ecosystems (THOMPSON et al., 2010).

The objective of this study was to investigate the frequency of occurrence and variety of intestinal parasites among free-living wild animals in two wild animal rehabilitation centers in the states of São Paulo and Mato Grosso do Sul.

Between January 2007 and May 2008, fecal samples were collected from mammals and birds that had been newly introduced (less than five days earlier) to wild animal rehabilitation centers in Jundiai, SP, and Campo Grande, MS. The samples were collected in the mornings using sterile collecting tubes, preserved in 5% potassium dichromate, stored under refrigeration (4°C) and sent to the Parasitology and Parasitic Diseases Laboratory of the State University of Northern Paraná, Luis Meneghel Campus, in Bandeirantes, PR, not more than one week after collection. The numbers and species of the animals that participated in this study are specified in Table 1. A total of 38 samples belonging to major groups were analyzed: five primates samples, thirteen felid samples, five canid samples, two marsupial samples, one deer samples adn twelve samples (mostly in a pool) from several birds.

Prior to coproparasitological analyses, fecal samples from the carnivores were subjected to the centrifugal-sedimentation technique in water-ether. Subsequently, the feces were examined by means of simple sedimentation and centrifugal-flotation methods in sucrose solution (d = 1.203 g/cm$^3$), as described by Ferreira et al. (1962).

In the present study, several biological forms of gastroenteric parasites were found in 71% of the 38 samples analyzed (Table 1). All 15 samples of mammals and birds from São Paulo (100%) and 52% of the samples from Mato Grosso do Sul were positive for one or more parasite. Although the original locations of the animals evaluated in this study were unknown, the greater occurrence of parasites in the wild animals at the rehabilitation center in São Paulo (chi-square = 7.91; p = 0.005) may have been due to the smaller environmental conservation area in relation to Mato Grosso do Sul. There was consequently a greater possibility of contact between wild animals, humans and domestic animals in São Paulo. The animals brought into these centers were mostly animals that had been run over or, in the case of cougars, cubs that had been caught by the competent environmental agencies (such as the Environmental Police). The birds originated from wild animal trafficking and domesticated pets that had been abandoned by their previous owners.

Parasites were found in 85% (11/13) of the felid fecal samples: *Ancylostoma* sp. and *Toxocara cati* eggs were found in 46%, *Cryptosporidium* sp. and *Cystoisospora* sp. oocysts in 15% and *Giardia* cysts and *Toxascaris leonina* eggs in 8% of these animals.

*Giardia* and *Cryptosporidium* are the most prevalent intestinal parasites in humans and domestic animals, and are increasingly recognized as common parasites of several wild animals (FAYER, 2004; THOMPSON, 2004; THOMPSON; MONIS, 2004; APPELBEE et al., 2005).

*Giardia* has been reported in several wild mammal species. However, little information is available about the species and genotypes that occur naturally in these mammals (APPELBEE et al., 2005; KUTZ et al., 2009; THOMPSON et al., 2009). In most cases, when appropriate tools for characterizing this parasite were applied, the *Giardia* genotype found in free-living terrestrial and aquatic mammals was usually of human origin, i.e. of *Giardia duodenalis* genotype (THOMPSON et al., 2009). *Giardia* cysts found in one of the cougars *(Puma concolor*) in the present study were genotyped and were also of human origin (SOARES et al., 2011). In all these cases, epidemiological evidence suggests that human beings are the source of infection, through direct or indirect environmental contamination from raising domestic animals. The impact of these zoonotic species of *Giardia* in the wild is unknown. The evidence for an association of *Giardia* infection with clinical disease in primates is sparse (GRACZYK et al., 2002).

The reemergence of infection by *Cryptosporidium* in humans, particularly in immune-deficient patients, has stimulated research, monitoring and characterization of this parasite in wild species in order to identify possible reservoirs for water-borne infection of human beings (THOMPSON et al., 2005). Several varieties of *Cryptosporidium* have been identified, but with little or no significance for public health (CACCIO et al., 2005; HUNTER; THOMPSON, 2005; SLAPETA, 2009). The diversity of *Cryptosporidium* spp. found in wildlife deserves further study in terms of ecology, evolution, biology and potential impact on the health of these animals (THOMPSON et al., 2010).

*Ancylostoma tubaeforme* is a cosmopolitan nematode in cats (ANDERSON, 2000) and, in Neotropical regions, it has been reported in jaguarundi (*Herpailurus yagouaroundi*), jaguars (*Panthera onca*) and Geoffroy's cats (*Oncifelis geoffroyi*) (THATCHER, 1971; MARTINÉZ, 1987). Beldomenico et al. (2006) identified helminth eggs in seven wild felids in Argentina and found several species, including *Ancylostoma tubaeforme*, *Toxocara cati* and *Taenia* sp.

Infection of these animal species may indicate that there could be proximity to and interactions with domestic cats. However, unfortunately, information about the helminthic fauna in Neotropical felids is rare and scarce (BELDOMENICO, 2006).

Regarding the canid samples analyzed, 20% of the animals presented *Giardia* cysts, *Eimeria* sp. oocysts and *Toxocara canis* eggs. In the samples of a *Cerdocyon thous* (crab-eating fox) and *Chrysocyon brachyurus* (maned wolf) were identified Cestoda eggs and *Eimeria* oocysts. In a study carried in wild canids of Serra do Cipó National Park, Brazil, revealed the presence of Ancylostomiae, Trichuridae eggs among others parasites in feces of *Cerdocyon thous* and *Chrysocyon brachyuru*, however, only one sample (20%) of *Cerdocyon thous* was found Cestoda eggs (SANTOS et al., 2012).

In the feces of a gray brocket (*Mazama gouazoubira*), only two *Giardia* sp. cysts were identified. Since this was the only

**Table 1.** Distributions of the species studied and parasites observed in fecal samples from wild animals at rehabilitation centers in the states of São Paulo and Mato Grosso do Sul.

| Sample | Animal species (common name) | State | Parasites found |
|---|---|---|---|
| 1 | *Alouatta seniculus* (howler monkey) | SP | *Enterobius* |
| 2 | *Ara chloroptera* (red-and-green macaw) and *Ara ararauna* (blue-and-yellow macaw)* | SP | *Strongyloides*; *Eimeria* sp. |
| 3 | *Cerdocyon thous* (crab-eating fox) | SP | Cestoda (eggs); *Eimeria* spp. |
| 4 | *Chrysocyon brachyurus* (maned wolf) | SP | Cestoda (eggs) |
| 5 | *Leopardus pardalis* (ocelot) | SP | *Toxocara cati* |
| 6 | *Leopardus pardalis* (ocelot) | SP | *Toxocara cati* |
| 7 | *Leopardus pardalis* (ocelot) | SP | *Toxocara cati* |
| 8 | *Leopardus pardalis* (ocelot) | SP | *Toxocara cati*; *Ancylostoma* |
| 9 | *Leopardus pardalis* (ocelot) | SP | *Ancylostoma* |
| 10 | *Leopardus tigrinus* (leopard tiger) | SP | *Cryptosporidium* |
| 11 | *Leopardus tigrinus* (leopard tiger) | SP | *Cryptosporidium* |
| 12 | *Mazama gouazoubira* (gray brocket) | SP | *Giardia* (trophozoites) |
| 13 | *Nasua nasua* (coati) | SP | *Giardia* (trophozoites) |
| 14 | *Panthera onca* (jaguar) | SP | *Toxocara cati*; *Toxascaris leonina*; *Ancylostoma* sp. |
| 15 | *Pionus maximiliani* (scaly-headed parrot)* | SP | *Cryptosporidium* |
| 16 | *Alouatta seniculus* (howler monkey) | MS | Cestoda (eggs) |
| 17 | *Alouatta seniculus* (howler monkey) | MS | Cestoda (eggs) |
| 18 | *Amazona aestiva* (blue-fronted Amazon)* | MS | - |
| 19 | *Amazona aestiva* (blue-fronted Amazon)* | MS | - |
| 20 | *Amazona amazonica* (orange-winged Amazon) | MS | - |
| 21 | *Ara ararauna* (blue-and-yellow macaw)* | MS | - |
| 22 | *Ara chloroptera* (red-and-green macaw)* | MS | *Cryptosporidium* |
| 23 | *Brotogeris tirica* (plain parakeet)* | MS | *Cryptosporidium* |
| 24 | *Callithrix jacchus* (common marmoset) | MS | - |
| 25 | *Cebus apella* (tufted capuchin) | MS | - |
| 26 | *Cerdocyon thous* (crab-eating fox) | MS | *Toxocara canis* |
| 27 | *Cerdocyon thous* (crab-eating fox) | MS | - |
| 28 | *Didelphis marsupialis* (common opossum) | MS | Oxyuroidea (eggs) |
| 29 | *Didelphis marsupialis* (common opossum) | MS | - |
| 30 | *Diopsittaca nobilis* (noble macaw) | MS | - |
| 31 | *Panthera onca* (jaguar) | MS | *Ancylostoma* |
| 32 | *Pionus maximiliani* (scaly-headed parrot)* | MS | - |
| 33 | *Polyborus plancus* (southern caracara) | MS | - |
| 34 | *Puma concolor* (cougar) | MS | *Cystoisospora felis*; *Ancylostoma*; *Sarcocystis*; *Giardia* |
| 35 | *Puma concolor* (cougar) | MS | *Ancylostoma* |
| 36 | *Puma concolor* (cougar) | MS | *Cystoisospora felis*; *Ancylostoma*; *Sarcocystis* |
| 37 | *Puma concolor* (cougar) | MS | *Toxocara cati* |
| 38 | *Ramphastos sulfuratus* (keel-billed toucan) | MS | *Eimeria* sp. |

*pool; MS: Mato Grosso do Sul; SP: São Paulo.

animal of this species in the reserves, studies on possible parasites that could be found in this species were compromised. Several studies have been published about deer parasites in New Jersey (SAMUEL et al., 1968), southern Chile (DIAZ et al., 1977), Hawaii (McKENZIE et al., 1989) and Brazil (NASCIMENTO, 1996, 2000). However, there are few reports on protozoa in deer. In southern Chile, presence of *Moniezia* and *Sarcocystis* has been reported (DIAZ et al., 1977).

In the feces samples from coatis (*Nasua nasua*), trophozoites containing several unidentified flagella were found.

Eggs from *Enterobius* sp. (Oxyuridae) were identified in the fecal sample from a red howler monkey (*Alouatta seniculus*). This is one of the few reports of this parasite in this species of non-human primate. In a scientific excursion to the Pantanal area of the state of Mato Grosso (the banks of the rivers São Lourenço and Cuiabá) many decades ago, researchers performed necropsies on 455 birds, 62 mammals, 63 fish, 25 reptiles and two amphibians. Among these, 43% were parasitized: 33% by nematodes, 27% by trematodes, 18% by cestodes, 7% by acanthocephalans and 3% by pentastomids. In this excursion, the first report of the genus

*Enterobius* (*Enterobius minutus* species) was made in a howler monkey (*Alouatta caraya*) (TRAVASSOS et al., 1927).

*Enterobius* nematodes are Oxyuridae that are found in humans and primates and have anthropozoonotic importance (DILRUKSDHI et al., 2006). Species of *Strongyloides* and *Enterobius* in non-human primates, including the human parasites *Enterobius vermicularis*, *Strongyloides stercoralis* and *S. fuelleborni* have been described by Inglis (1961), Yamaschita (1963), Collet et al. (1986) and Monteiro et al. (2003).

Another study published with similar findings was conducted in 2001, on 125 monkeys of the species *Macaca sinica sinica*, *Trachypithecus vetulus philbricki* and *Semnopithecus priam thersites* in the Polonnaruwa Nature Sanctuary and Archaeological Reserve in Sri Lanka. The samples were analyzed by means of the fecal flotation test and the researchers identified the genus *Enterobius* in 52% of the monkeys studied, among other parasites (EKANAYABE et al., 2006).

In relation to the birds, a pool of feces was collected from the cages and examined. Several *Cryptosporidium* sp. oocysts in scaly-headed parrots (*Pionus maximiliani*), *Eimeria* sp. oocysts and *Strongyloides* sp. embryonated eggs in blue-and-yellow (*Ara ararauna*) and red-and-green macaws (*Ara chloroptera*) were identified.

Studies conducted on these birds in the state of Pernambuco showed the presence of *Ascaridia* sp., *Strongyloides* sp., *Heterakis* spp., Strongyloidea and Spiruroidea; Trematoda and Cestoda eggs; *Balantidium coli*, *Entamoeba coli* and *E. histolytica* cysts; and coccidian oocysts. Among these, higher occurrence of *Capillaria* sp., Strongyloidea, *Strongyloides* sp. and *Ascaridia* sp. was found, with prevalences of 76.5, 13.7, 5.9 and 2% respectively (FREITAS et al., 2002).

Despite the small number of samples collected, it could be concluded that the diversity of parasites found in fecal samples from the animals evaluated was noteworthy, considering the lack of information on occurrence and diversity of the vast majority of parasites in free-living wild animals under Brazilian conditions.

Studies on endoparasite fauna in wild animals and consequent detection of infection in these animals might suggest that there could be proximity to and interactions with domestic animals and/or humans. Therefore, further studies on parasites in wild animals are necessary in order to better understand the intensity of anthropic changes to wild environments.

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