SURVEY OF GASTROINTESTINAL PARASITES OF THE CENTER FOR SCREENING OF WILD ANIMALS FROM SÃO LUÍS, MARANHÃO STATE, BRAZIL

PESQUISA DE PARASITOS GASTRINTESTINAIS DO CENTRO DE TRIAGEM DE ANIMAIS SILVESTRES DE SÃO LUÍS DO MARANHÃO, BRASIL

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SUMMARY

The objective was to identify the gastrointestinal parasites of wild animals received by the Center for Screening of Wild Animals from São Luis, Maranhão State, Brazil. The fecal samples were collected from 297 animals, of which 150 (50.5%) were birds, 132 (44.5%) mammals and 15 (5%) reptiles. A total of 262 samples (227 individual and 35 pools) were analyzed by the method of sedimentation and flotation, 102 animals were parasitized by helminthes and/or protozoa, 39 of which belonged to the order Primate, 18 to the Carnivorous order and 12 to the Psittaciformes order. The mammals had the greatest diversity of eggs of gastrointestinal parasites: *Strongyloides* sp., hookworm type, *Spirometra* sp., *Ascaris* sp., *Trichuris vulpis*, *Capillaria* sp., Strongyloidea, Ancylostomidae , taenid and oocysts of coccidian. In the samples of birds were diagnosed eggs of the genera *Dispharynx*, *Ascaridia*, *Echinostoma* and oocyst of coccidian. In the reptiles, the number of gastrointestinal parasites was low, being identified oocyst of coccidian in three jiboia (*Boa constrictor*) and *Capillaria* sp. in an iguana (*Iguana iguana*). It was concluded that wild animals act as hosts for various species of parasites and to establish the dynamics and parasitic fauna of these animals at the screening center is an excellent alternative for studies *ex situ.*

KEY-WORDS: Coproparasitology; Endoparasites; Helminths; Mammals; Reptiles.

RESUMO

Objetivou-se identificar os parasitos gastrintestinais de animais silvestres recepcionados pelo Centro de Triagem de Animais Silvestres de São Luís, Maranhão. As amostras fecais foram coletadas entre agosto de 2006 a julho de 2008, totalizando 297 animais, dos quais 150 (50,5%) foram aves, 132 (44,5%) mamíferos e 15 (5%) répteis. Do total de 262 amostras (227 individuais e 35 *pools*) analisadas pelo método de sedimentação simples e flutuação, 102 animais estavam parasitados por helmintos e/ou protozoários, sendo que, 39 pertenciam à ordem Primata, 18 à Carnívora e 12 à Psittaciforme. Os mamíferos apresentaram a maior diversidade de ovos de parasitos gastrintestinais, como *Strongyloides* sp., ancilostomídeo, *Spirometra* sp., *Ascaris* sp., *Trichuris vulpis*, *Capillaria* sp., Strongyloidea, Ancilostomídeo e tenideos, além de oocistos de coccídios. Em amostras de aves identificaram-se ovos dos gêneros *Dispharynx*, *Ascaridia*, *Echinostoma*, e também de oocistos de coccídeos. Nos répteis, o número de parasitos gastrintestinais foi baixo, sendo identificados oocisto de coccídeo em três jiboias (*Boa constrictor*) e *Capillaria* sp. em uma iguana (*Iguana iguana*). Concluiu-se que animais silvestres atuam como hospedeiros para diversas espécies de parasitos, e estabelecer a riqueza e a dinâmica da fauna parasitária desses animais nos centros de triagem é uma excelente alternativa para a realização de estudos *ex situ.*

PALAVRAS-CHAVE: Coproparasitologia; Endoparasitos; Helmintos; Mamíferos; Répteis.

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Submetido: 23/01/2018 Aceito: 22/08/2018

INTRODUCTION

Brazil takes part of the sixth countries with the richest biological biodiversity (Lewinsohn; Prado, 2002). Due to its importance, surveys on wild fauna in Brazil have been conducted not only to minimize the macroscopic ecological imbalance caused by the historical withdrawal of these animals from its natural habitat but also because the microscopic imbalance that can be caused by internal parasitic fauna. The parasitic fauna have an wild cycle and could have an urban cycle too that same times can have impact on public health (Mackenstedt et al., 2015) such as leishmaniosis, Chagas' disease and hydatidosis (Alexandre, 2000). However, studies on parasitic fauna in wild animals should not be restricted to its role as reservoirs of zoonotic diseases but also as a means of conservation and maintenance of biodiversity (Thompson et al., 2010).

Studies have demonstrated that infection by parasites can have severe effects on the survival and reproduction of the host species, therefore elaborate efficient protocols to maintain health and genetic diversity must be a priority (Scott, 1988), especially regarding wild species.

Important methods to study the parasitic fauna of wild animals are the fecal exams of the hosts by searching for parasite eggs, cysts, oocysts and larvae (Vieira et al., 2006). These methods are quick, cheap and non-invasive. The relevance of the research on parasitic fauna is recognized and required in reintroduction protocols (UICN, 1998; Felasa, 1999) and also in the clinic routine of wild animals.

Though authors believe the captivity condition is a disadvantage for a great varied of parasites since the source of infection are more limited than in free life, in a general way, wild animals housed in captivity are more susceptible to infectious and parasitic diseases (Freitas et al., 2001; 2002), specially due to the inadequate hygiene of the accommodations (Marietto-Gonçalves et al., 2009). However, due to the stress of the captivity lived by these animals, they become susceptible to the actions of the parasitic fauna that until then lived in equilibrium with their host (Catão-Dias, 2003; Santos et al., 2015). Knowing that in free life the action of parasites on hosts also serve as population control of a species. Furthermore, parasite diversity can be an indicator of ecosystem health, as it reflects the stable relationships between parasites and hosts (Santana; Mesquita; Seixas Filho, 2014).

Taking into account the spread of the cities upon the wild environment and the participation of man in the parasite life cycle of wild animals (Lim et al., 2008; Li et al., 2015) it is necessary to perform studies on internal fauna of these animals to try to minimize the impact on public and animal health. Moreover, over recent decades, diseases have shown to be important causes of extinctions among wild species. Greater emphasis has been given to diseases transmitted by domestic animals, which has been increasing in numbers in natural areas, along with human populations (Santos et al., 2012).

There are few researchers on parasites of wild animals in the State of Maranhão, Brazil and the available information is restrict to external parasites (Guerra et al., 2000; Figueiredo et al., 2010). So the aim of this study was to report on the diversity of parasitic helminthes and protozoan in feces of wild animals received at Center for Screening of Wild Animals of São Luis, Maranhão (CETAS/MA).

MATERIAL E MÉTODOS

Study area

The study was performed in the Center for Screening of Wild Animals of São Luis, Maranhão (CETAS/MA) (2º56'80"S, 44º21'01"O). CETAS belongs to IBAMA, the Brazilian Institute of Environment and Renewable Natural Resources, and has as primary activities to receive, take care, rehabilitate and give a destination of wild animals that were captured, rescued or spontaneously dropped off by the population. Therefore in great majority of the cases the origin of these animals is unknown

São Luis city is located at São Luis Island, northeastern of Brazil (Figure 1). The climate is tropical humid and temperatures vary between 26º to 28ºC. There are two seasons well defined, the dry season from July to December and the rainy season from January to June. Rainfall index can reach above 2000 mm^3.

Samples collection and fecal exams

Fecal samples were collected during the entrance of the animals at CETAS, from August/2006 to July/2008 covering the dry and rainy seasons. The samples were collected early in the morning in canvas placed under the cages or enclosure of animals'. They were put on recipient per sampled animal or pools when the cage or enclosure had more than one animal. The following methods for finding eggs, cysts, oocysts and larvae of parasites in the samples: flotation in saturated chloride solution (Willis, 1927) and sedimentation (Hoffmann et al., 1934). To visualize parasitic forms a light microscopic was used and the identification was done according to Soulsby (1968), Skryanbin (1969) and Yamaguti (1961).

Figure 1 - Satellite image of Maranhão State, São Luis Island and Center for Screening of Wild Animals of São Luis, Maranhão (CETAS/MA). Source: MapInfo Professional 7.5 SCP; Google earth.

RESULTS

A total of 297 animals were sampled as follows: 15 (5%) reptiles (Table 1), 132 (44.5%) mammals and 150 (50,5%) birds (Table 2). A total of 262 fecal samples were analyzed being 227 individual samples and 35 pools. From the 297 animals, 102 (34.34%) were infected by helminthes and/or protozoan, from these, 63 (61.77%) were mammals, 35 (34.31%) were birds and 4 (3.92%) were reptiles. Same eggs identification only could be done until the taxon of superfamily.

Eggs of the following genera of gastrointestinal helminths were identified in birds: *Ascaridia*, *Dispharynx*, *Echinostoma*. Coccidian oocysts were also detected. In the sample pools of *Dendrocygna viduata, Amazona amazonica, Amazona aestiva* and *Ara maracana* we detected oocysts of cocidian and eggs of *Ascaridia* sp. (Table 3). Mammals were infected by *Strongyloides* sp., eggs of the hookworm type, *Spirometra* sp., *Ascaris* sp., *Trichuris vulpis*, *Capillaria* sp. (Table 3). Eggs of taenid, Strongyloidea and hookworm type and oocyst of coccidian were also observed. In reptiles, the prevalence of gastrointestinal parasites was low. Coccidian oocystis in three *B. constrictor* and eggs of *Capillaria* sp.in a *I. iguana* were detected. The results are summarized in Table 3.

Table 1 - List of reptiles sampled at the Center for Screening of Wild Animals of São Luis, Maranhão State (CETAS/MA), Brazil, from August/2006 to July/2008.

Order	Family	Common name in English	Number sampled/Specie
Squamata	Iguanidae	Green iguana	01 *Iguana iguana*
	Boidae	Boa constrictor	12 *Boa constrictor* 01 *Boa constrictor amarili*
Chelonia	Chelonidae	Yellow-footed Tortoise	01 *Geochelone denticulata*

Table 2 - List of mammals and birds sampled at the Center for Screening of Wild Animals of São Luis, Maranhão State (CETAS/MA), Brazil, from August/2006 to July/2008.

Order	Family	Common name in English	Number sampled/Species
Mammals			
Primata	Cebidae	Tufted capuchin	69 *Sapajus apella*
		Squirrel monkey	07 *Saimiri sciureus*
	Callithrichidae	Common marmoset	04 *Callithrix jacchus*
		Tamarin	05 *Saguinus midas niger*
	Atelidae	Black Howler	01 *Alouatta caraya*
Carnivora	Canidae	Crab-eating fox	03 *Cerdocyon thous*
	Procyonidae	South American Coati	12 *Nasua nasua*
		Raccoon	01 *Procyon cancrivorus*
	Felidae	Oncelot	04 *Leopardus pardalis*
		Jaguarundi	02 *Puma yagouaroundi*
		Margay	02 *Leopardus wiedii*
		Northern Tiger Cat	06 *Leopardus tigrinus*
	Mustelidae	Lesser grison	03 *Galictis cuja*
Didelphimorphia	Didelphidae	Opossum	02 *Didelphis marsupialis*
Artiodactyla	Cervidae	Brown brocket	01 *Mazana gouazoubira*
Rodentia	Agoutidae	Spotted paca	03 *Agouti paca*
Xenarthra	Myrmecophagidae	Southern tamandua	02 *Tamandua tetradactyla*
	Bradypodidae	Brown-throated Sloth	05 *Bradypus variegatus*
Birds			
Anseriforme	Anatidae	White-faced duck	28 *Dendrocygna viduata*
		Black-bellied Whistling-duck	18 *Dendrocygna autumnalis*
Psittaciforme	Psittacidae	White-winged Parakeet	04 *Brotogeris versicolurus*
		Golden Parakeet	03 *Guaruba guarouba*
		Peach-fronted Parakeet	02 *Eupsittula aurea*
		Orange-winged Amazon	23 *Amazona amazonica*
		Turquoise-fronted Amazon	22 *Amazona aestiva*
		White-eyed Parakeet	02 *Aratinga leucophthalmus*
		Scarlet Macaw	02 *Ara macao*
		Blue-winged Macaw	03 *Ara maracana*
		Scaly-headed Parrot	01 *Pionus menstruus*
		Scaly-headed Parrot	01 *Pionus maximiliani*
Strigiforme	Tytonidae	Common Barn-owl	07 *Tyto Alba*
	Strigidae	Tropical Screech-owl	02 *Megascops choliba*
		Striped Owl	01 *Asio clamator*
		Burrowing Owl	02 *Speotyto cunicularia*
Passeriforme	Fringilidae	Purple-throated Euphonia	03 *Euphonia chlorotica*
		Lined Seedeater	05 *Sporophila lineola*
		Campo Troupial	05 *Icterus jamacaii*
		White-lined Tanager	01 *Thachyphonus rufus*
		Chopi Blackbird	01 *Gnorimopsar chopi*
		Yellow-rumped Cacique	01 *Cacicus cela*
	Sturnidae	Tropical Mockingbird	05 *Mimus gilvus*
Piciforme	Ramphastidae	Channel-billed Toucan	06 *Ramphastos vitelinus*
		Red-billed Toucan	01 *Ramphastos tucanus*
		Spot-billed Toucanet	01 *Selenidera maculirostris*

Table 3 - Parasites (eegs and oocysts) in wild animals from the Center for Screening of Wild Animals of São Luis, Maranhão State (CETAS/MA), Brazil, from August/2006 to July/2008.

Animals	Number of positive samples	Species	Parasites (eggs/oocysts)*
Birds			
Pool	04	*Dendrocygna viduata*	oocysts of coccidia , *Ascaridia* sp. *Dispharynx* sp.
	04	*Amazona amazonica*	oocysts of5uiolkjgfghrtyuighlkjujm´kujytgrttyg coccidia e *Ascaridia* sp.
	03	*Amazona aestiva*	
	01	*Ara maracana*	
Individual	05	*Tyto Alba*	oocysts of coccidia
Pool	03	*Dendrocygna autumnalis*	*Echinostoma* sp., *Ascaridia* sp
Individual	01	*Sporophila lineola*	oocysts of coccidia
Individual	01	*Ramphastos toco*	oocysts of coccidia
Mammals			
Individual	06	*Leopardus tigrinus*	*Spirometra* sp.
Individual	04	*Leopardus pardalis*	*Trichuris vulpis*
Individual	02	*Puma yagouaroundi*	*Trichuris vulpis*
Pool	04	*Nasua nasua*	*Strongyloides* sp., eggs of the hookworm type
Pool	02	*Cerdocyon thous*	eggs of the hookworm type, oocysts of coccidia
Individual	05	*Saimiri sciureus*	eggs of the hookworm type, *Ascaris* sp., taenídeo
Individual	15	*Sapajus apella*	eggs of the hookworm type, *Ascaris* sp., Strongyloidea
Pool	05	*Sapajus apella*	eggs of the hookworm type, *Ascaris* sp., oocysts of coccidia, *Capillaria* sp., taenídeo, Strongyloidea
Individual	03	*Agouti paca*	*Strongyloides* sp., eggs of the hookworm type.
Individual	02	*Tamandua tetratactyla*	eggs of the hookworm type
Individual	01	*Mazana gouazoubira*	*Strongyloides* sp.
Reptiles			
Individual	01	*Iguana iguana*	*Capillaria* sp.
Individual	03	*Boa constrictor*	oocysts of coccidia

DISCUSION

The biodiversity can be composed by different co-evolutionary process in a variety of taxonomic levels (Ehrlich; Raven, 1964; Margulis, 1971; Hamilton et al., 1990; Thompson, 2009), including parasites. They suffer selective pressure along with their hosts (co-evolution) so when they came to extinction their parasites are extinct as well (Thompson, 2009).

The study of the parasitic fauna of domestic and wild animals have much more emphasis in the species that causes economic loss and zoonosis and parasites that have effect on wild fauna are not a priority (Thompson et al., 2010). However, importance should be given to parasitic fauna of wild animals housed in captivity such as the ones from zoo and center of conservation.

The release of animals housed in captivity for a long time decrease their immunological capacity to react to great majority of natural pathogens from animals of the same species in a natural environment (Wyatt et al., 2008). Additionally, the impact of human proximity and anthropic action upon the wild environment and consequently to their pathogens is not well stablished (Thompson et al., 2010).

In the present study the eggs of the hookworm type and *Strongyloides* were the most frequent in fecal samples of mammals. Eggs of the hookworm type were identified in samples from primates, carnivore, rodents and xenarthrans. This helminth has direct life cycle, the infective third larva stage is very active. In domestic canids and felids it is the most common gender observed (Bowman, 2014) and the most pathogenic species cause anemia and weight loose (Fortes, 1993), it is also common in wild animals (Santos et al., 2015; Kouassi et al., 2015). *Ancylostoma*, one of the most

important geohelminth, causes environmental contamination and zoonosis such as cutaneous larva migrans and eosinophilic enteritis in humans. The pathogenicity of the adult worm depends on the parasite load and host age (Bowman, 2014).

The genus *Strongyloides* was identified in samples of mammals (primates, carnivores and artiodactyls), the same reported in *Alopex lagopus* by Aguirre et al. (2000) and in carnivores and artiodactyls by Freitas et al. (2001). The greatest number of positive samples for *Strongyloides* was in primates. It could be explained by the fact that the third infective stage of this genus has the capacity of active penetration so animals can be re-infected or infected themselves by entering in contact with contaminated soil (Fortes, 1993). Infection by *Strongyloides* sp. taenid (*Hymenolepis* sp.) in samples from primates have been reported (Gonzalo et al., 1990; Arrojo, 2002), as observed in the present study since we also identified taenid eggs in four samples of Neotropical primates. The genus *Strongyloides* infect numerous vertebrate, such as snakes, felids, canids and ruminants (Dorris et al., 2002). In Brazil, species of this genus have been reported infecting a wide diversity of hosts (Vicent et al., 1997) as the first report of the occurrence of *Strongyloides* in *Leopardus trigrinus* in the Botucatu, State of São Paulo by Santos et al. (2009)

Eggs of *Spirometra* sp. were detected in samples from *L. tigrinus*, it is a common finding in carnivores, birds and amphibians. These animals generally feed on crustaceans, the intermediate host. This parasite has dogs, cats and raccoons as its definitive hosts (Bowman, 2014) and can determine a zoonosis called sparganosis (Mentz et al., 2011). The infection by *Spirometra* was also reported in *Puma concolor* and *Panthera onca* in Perú (TANTALEÁN; Michaud, 2005), in *Leopardus colocolo* in Brazil (Gresseler at al., 2016) and other felids, as observed by Khatun et al. (2014), in lions in captive condition in Bangladesh and snakes (Almeida et al., 2016).

Trichuris vulpis was identified in samples from *L. pardalis* and *P. yagouaroundi* in the present study. This parasite was reported in wild felids in captive in the State of Santa Catarina, Brazil (Muller et al., 2005). Species of the family Trichuridae were reported in *Leopardus colocolo* by Gresseler et al. (2016) in the State of São Paulo. *Trichuris vulpis* has a short period of maturation in the environment (9 to 10 days) becoming infective in short time enabling re-infection (Fortes, 1993).

Birds were infected by *Dispharynx*, *Ascaridia*, *Echinostoma*, besides coccidian as previously reported by Freitas et al. (2002) in the State of Pernambuco, Brazil. *Ascaridia* was the most frequent finding along with coccidia. *Ascaridia* is the most common gastrointestinal helminth in captive birds. It has been reported in exotic birds in the State of Sergipe, Brazil (Lima et al., 2016). This parasite causes deficient absorption, weight loose, anorexia and diaarrhea. It has also been described causing intussusception, occlusion and death (Ritchie et al., 1994). It is common in Psittaciformes birds being identified in all orders of birds sampled in the present study. In CETAS of João Pessoa, State of Paraíba, Brazil, *Ascaridia* was recovered by necropsy of Psittaciformes birds as *causa mortis* intestinal due to intestinal obstruction (Melo et al., 2013). According to Snak et al. (2012) taenid eggs were prevalent in captive birds in Paraná State, Brazil, however this eggs were identified in the present study.

Coccidian are common intestinal parasites in birds. Here we detected non sporulated oocysts all orders of birds sampled; however they were more prevalent in Anseriformes. In Brazil toucans in captivity are frequently found infected (Benez, 2004). Generally it is necessary to make intense work of erradication in places where coccidan are present since they are resistant to environmental condition, besides they have direct life cycle that enables the persistence of infected animals (Benez, 2004). They are also identified in co-infection with nematodes (Lima et al., 2016), as demonstrated in the present study in *pools* of *Dendrocygna viduata*, *Amazona amazônica*, *Amazona aestiva* e *Ara maracana* (oocysts coccidian and *Ascaridia*).

According to Junker et al. (2015) coccidiosis is a disease of intensification due to the build-up of the sporulated oocysts in accumulated feces, facilitating ingestion of large infective doses. A further fact is immunosuppression of host, due to stress. This is particularly relevant in free-ranging wild animals brought in captivity, even temporarily.

Dispharynx is a common nematode on wild birds and birds reared in extensive system (turkey, free-range chicken and guinea fowls). Their habitat in the host is the proventriculus and gizzard (Bartmann; Amato, 2009). Eggs of this parasite were detected in Anseriformes. The pathology caused by this parasite is more severe in young animals and influences their growth (Ritchie et al., 1994; Bartmann; Amato, 2009).

In reptiles, the number of gastrointestinal infection was low and oocysts of coccidian were identified in three *B. constrictor* and *Capillaria* sp. in an *I. iguana*. Infections by coccidia are very common in free reptiles and the majority of the cases are of low or none pathogenicity. The infected animals present the intestinal epithelium health or with few lesions, they recovery is fast enough so they are asymptomatic (Raś-Noryńska; Sokół, 2015). Protozoan of the *Eimeria* gender are found in the biliary ducts and gall-bladder of reptiles while *Isospora* is found mainly in the intestine (Raś-Noryńska; Sokół, 2015). According to Rataj et al. (2011) *Capillaria* is a common finding in reptiles, however it was not identified here.

It should be emphasizes that reptiles became increasingly common domestic pets. In wild, they rarely come into contact with their own waste or uneaten food, which is a common occurrence in captive (Dovc et al., 2004) favoring contact with contaminated materials.

The samples that were analyzed by the sedimentation method of Hoffmann et al. (1934) presented a more satisfactory results when compared to the results obtainded by the use of the flotation method of Willis (1927) since it detected a greater number of positive samples as observed by Cerqueira et al. (2007) in comparing the sensibility both methods in

diagnosing ancilostomid eggs. Similarly, Freitas et al. (2001; 2002) verified that the sedimentation method was more efficient to detect eggs, oocysts and cysts in the feces of wild mammals and birds under captive conditions.

It can be concluded that wild animals act as hosts of different species of parasites and the knowledge of the richness parasitic fauna in the Centers for Screening of Wild Animals is an excellent alternative to perform *ex situ* studies as well important for controlling and preventing parasitic diseases.

ACKNOWLEDGEMENTS

To Cetas' staff, specially Dr. Roberto Rodrigues Veloso Junior. To Conselho Nacional de Desenvolvimento Científico e Tecnológico (***CNPq***) for the financial support by means of the schorlarship.

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