Cryptosporidiosis in Reptiles from Brazil

Subjects: Parasitology

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The concern about the protection of wildlife has been gathering attention from researchers worldwide. Zoos and aquariums have become widely recognized sites for the conservation of wildlife. The persistence of the illegal trade of wild animals, such as reptiles, and their use as pets can endanger not only the preservation of the species, but also allow the introduction of new pathogens and zoonotic diseases. It is important to highlight that preventive exams should be carried out prior to introducing these animals into a new facility to guarantee zoological management strategies. There are several reports of parasitism in reptiles, some of them with zoonotic potential, such as the genus *Cryptosporidium* spp. In Brazil, reports that explore the prevalence of cryptosporidiosis in reptiles are scarce, and very few have used molecular methods for the detection of *Cryptosporidium* spp., or the genotyping of its species and subtypes.

Cryptosporidium spp. zoonosis snakes

1. Introduction

At present, the Reptilia class is divided into three subclasses and four orders. The subclass Anapsida includes the Order Testudines, represented by tortoises and turtles. The subclass Diapsida includes: The order Rhynchocephalia, which is represented by tuataras, the order Squamata, which encompasses the suborders Sauria (Lacertilia), and Amphisbaenia, which are represented by lizards, and the suborder Ophidia (Serpentes) is represented by snakes. The subclass Archosauria includes the order Crocodylia, which is represented by crocodiles, alligators, gharials, and caimans [1].

According to the Reptile Database, there are more than 11,050 species recognized in the world [1]. Australia leads the world in reptile species richness, and Mexico takes second place. An updated checklist of Brazilian reptile species has highlighted Brazil as being the third-richest in the world, regarding reptile fauna, with 795 species: 36 Testudines, 6 Crocodylia, and 753 Squamata (72 amphisbaenians, 276 “lizards”, and 405 snakes) with almost half (47%) of Brazilian reptiles endemic to the country [2].

In the survey conducted by Costa and Bérnils [2], the Northern region is the richest in species of reptiles (453), Squamata (423), snakes (243), lizards (152), chelonians (25), and alligators (5)—the latter group on an equal footing with the Midwest region. The Northeast region is the second-richest for these groups, except for alligators and snakes, and is the region with the most Amphisbaenia taxa (35), while the least wealth regarding the number of reptiles of all groups is found in the South.

In a fast-changing world with growing concerns about biodiversity loss, zoos and aquariums have become widely recognized sites for the ex situ conservation of wildlife. These sites are essential not only for the protection of endangered species, but also to improve research regarding conservation strategies, captive care, environmental education, and the elucidation of diseases [3][4].

In general, captivity is often associated with frequent exposure to stressors, as the animals are restricted to a smaller common area, which may favor the transmission of diseases. Parasites are likely to become a major challenge for maintaining wildlife populations of endangered species in this preservation modality. Determining the presence of gastrointestinal parasites is critical for more appropriate decision making in the management of these populations, as its requires careful control to minimize loss diversity [5][6]

Currently, the popularity of exotic pets has been increasing, and they have been drawing heightened attention. Many of them are collected from the wild at the point of their origin, or are the offspring of wild-caught animals [7]. Inadequate capture techniques, and poor and/or improper shipping, are causes of death for many reptiles before they even reach the pet stores.

Unfortunately, reptiles are among the most inhumanely treated animals in the pet trade. Rataj et al. [8] highlights that about 90% of wild-caught reptiles die in the first year of captivity, mainly because of physical trauma or because, for many species, the basic requirements for housing are unknown and their owners lack knowledge concerning their nutritional management, making them highly susceptible to metabolic diseases.

Additionally, the introduction of exotic species into the country, probably derived from the illegal pet trade which occurs freely in shopping portals and on social networks on the internet, increases the risk of establishing those animals in a new natural area. After the successful establishment of those specimens, control and management costs will become higher, and total eradication may be impossible, in most cases [9].

Nevertheless, it is important to highlight that the practice of keeping exotic or wild animals as pets may pose a risk to human health, as those animals can carry diseases, with possibly serious effects on the increase in invasive pathogens, such as viruses (e.g., West Nile virus), bacteria (e.g., *Salmonella* spp., *Leptospira* sp., *Mycobacterium* sp.), fungus (e.g., *Candida* sp., *Trichosporon* sp.), and protozoans (e.g., *Cryptosporidium* sp.) [8][10] outside of their native distributions. Moreover, captive environments can be stressful to these animals, as they usually live in high densities and limited spaces, compromising their immune systems, which favor the presence of parasitic diseases [11]. On the other hand, the anthropic influence on the environment has been constantly cited as a potential risk factor, as well as and cross-transmission of pathogens between wild animals, domestic animals, and humans [12].

All reptiles should be examined for specific pathogens (endo and ectoparasites, *Salmonella* spp., *Leptospira* spp., etc.) before introducing them into a new facility. It is essential to perform preventive exams to better understand which parasites can be found in captive animals to guarantee zoological management strategies [11].

2. *Cryptosporidium* spp.

There are several reports of parasitism in reptiles. The most common protozoa that may have public health implications belong to the genus *Cryptosporidium* [13][14][15][16][17].

Cryptosporidiosis is a disease caused by a protozoan parasite of the genus *Cryptosporidium* that infects epithelial cells in the microvillus border of the gastrointestinal tract of a broad range of vertebrates worldwide, including amphibians, reptiles, mammals, and birds [18][19][20].

Until recently, this apicomplexan parasite was grouped as a coccidian. However, Cavalier-Smith [21] reclassified *Cryptosporidium* from class Coccidiomorphea, subclass Coccidia, to class Gregarinomorphea, within a new subclass, Cryptogregaria, and a new order, Cryptogregarida, within the Family Cryptosporidiidae. According to Ryan et al. [22] similarities with gregarines rather than other coccidia include completing host-free life cycles, exhibiting sizable extracellular gamonts, syzygy, and a changing cell architecture to adapt to diverse environments, e.g., biofilms, coelom, intestines, soil, and water, and even the lack of an apicoplast, with the ability to complete its life cycle in the absence of a host cell in vitro.

Cryptosporidium spp. is an intracellular, but extra-cytoplasmic, parasite. Even though some scientific publications describe *Cryptosporidium* spp. as an epicellular parasite, this term does not best define them. The life cycle is monoxenous, requiring a single host. Cosmopolite transmission occurs by the ingestion of oocysts, which are highly resistant to environmental conditions [23].

More than 38 species of *Cryptosporidium* have been identified, and there are more than 40 additional genotypes of unknown status yet to be formally named. Nevertheless, only four species of *Cryptosporidium* are known to infect reptiles, and only two have been shown to cause disease in snakes [24][25][26].

3. *Cryptosporidium* spp. in Reptiles

Cryptosporidium infections are common in reptiles, and may affect many different species [19][20][27]. Reptilian *Cryptosporidium* species can also be distinguished by their predilection sites, e.g., gastric or intestinal. Most of them affect the gastrointestinal tract. *Cryptosporidium serpentis* and *Cryptosporidium testudines* are gastrointestinal parasites, whereas *Cryptosporidium varanii* (*Cryptosporidium saurophilum*) and *Cryptosporidium ducismarci* are intestinal parasites species. Additionally, some species differ in morphology, as *C. serpentis* oocysts are bigger than those of *C. varanii* [24][27][28].

Other species of *Cryptosporidium* that have been isolated from reptilian feces include *Cryptosporidium baileyi*, *Cryptosporidium muris*, *Cryptosporidium parvum* mouse genotype, and *C. parvum* bovine genotype. Although animals can present oocysts that are detected in the feces, it is important to recognize that an infected prey can be a source of oocysts that are ingested by the reptilian, undergo a passive oocyst transfer through the gastrointestinal tract, and do not cause subsequent infection [29]. Prior to recent studies, no infections in humans had been linked with reptilian *Cryptosporidium* species [30][31].

In wild animals, infection occurs predominantly asymptomatically, but apparently, there are some animal groups that are more sensitive than others. Cryptosporidiosis is a disease with a generally chronic course, and it can manifest in two ways: clinical, causing gastritis, enteritis, and gastroenteritis, or subclinical, in which the infected animal plays an important role as a carrier of oocysts to the environment [32]. Unlike in other animals, in which *Cryptosporidium* infection is usually self-limiting in immunocompetent individuals, in reptiles, it is frequently chronic and sometimes lethal, especially in snakes [19][27][33].

In lizards, protozoan infections have been associated with acute enteritis and bacterial gastritis, with clinical signs including diarrhea, anorexia, lethargy, and weight loss which may even be a reason for euthanasia in these animals [32][34]. There are reports of polyps forming in an iguana (*Iguana iguana*) ear canal, and the parasite has also been described as causing prolapse and cystitis, associated with severe lesions, in the gastrointestinal tract of these animals, in hosts of the same species, which demonstrates the versatility of the breeding site [35][36].

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