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Prevalence of cryptosporidiosis among captive wild animals and birds in the arid region of north-eastern Nigeria

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ABSTRACT

The prevalence of cryptosporidial infection among captive wild animals and birds in Sanda Kyarimi Park, Maiduguri, north-eastern Nigeria, was evaluated by faecal examination for oocysts using three different staining techniques: the modified Ziehl-Neelsen, Giemsa, and Safranin-methylene blue. A total of 66 captive wild animals and birds were examined based on age and sex differences. Fifteen (22.7%) of the animals examined were young and 51 (77.3%) were adults; 36 (54.5%) were males and 30 (45.5%) were females. Age and sex were found to have no significant effect on the prevalence of infection among the different age and sex groups. A total of 15 (22.7%) positive samples were recorded using the modified Ziehl-Neelsen stain, while Giemsa and Safranin-methylene blue stains recorded 12 (18.2%) and 8 (12.1%) positive cases, respectively. Captive wild animals and birds could play an important role in the epidemiology of cryptosporidiosis as a zoonotic infection.

Key words: cryptosporidiosis, captive wild animals, zoonotic infection, Nigeria

Introduction

Cryptosporidial species are small coccidian parasites of the digestive and respiratory organs of vertebrates that have long been recognized as potential pathogens of man, animals and birds (SLAVIN, 1955; POHLENZ et al., 1978; TZIPORI et al., 1980a; LINKS, 1982; FAYER and UNGER, 1986). However, the parasite lacks a specific host, with cross-infections frequently occurring between different animal species and in man (TZIPORI et al. 1980b; CURRENT et al., 1983). Studies have shown that bovine cryptosporidia from

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calves can cause infection in newborn lambs, pigs, rats, mice, guinea pigs and chicks (ANDERSON, 1982), and the apparent role of cryptosporidium species as a pathogen has been established through transmission studies (MOON and BEMRICK, 1981; REUSE et al., 1982). This evidence suggests the possibility of cryptosporidiosis as a potential zoonosis and that species of *Cryptosporidium* may not be absolutely host-specific. The parasite has been incriminated as a cause of neonatal diarrhoea in calves, lambs, rhesus monkeys and turkey poults (ANDERSON, 1982), and is associated with diarrhoea in immunosuppressed or immunodeficient human beings (BIRD and SMITH, 1980) and foals (SNYDER et al., 1978; POONACHA and TUTTLE, 1989; MAIR et al,. 1990).

The veterinary importance of the parasite lies with young animals for which diarrhoea can be debilitating. Infection may occur sporadically or as outbreaks following zoonotic transmission from animals, person-to-person spread or the contamination of water supplies (CASEMORE, 1991) and feed (MILLARD and GENSHEIMER, 1994). Zoonotic transmission may account for most human infections especially for persons living or working in close association with potential animal reservoirs (CURRENT, 1985). Wild animals such as rodents have been identified as reservoirs of *Cryptosporidium* by several authors (KLESIUS et al., 1986; ANGUS, 1991; WEBSTER and MacDONALD, 1995; CHALMERS et al. 1997) and some of them serve as a source of diet for many domestic carnivores and man in many parts of the world. Therefore, the presence of *Cryptosporidium* in wildlife species could serve as a potential reservoir for transmission of infection to domestic animals and man (BRENT et al., 1982).

Although some researchers have worked on cryptosporidiosis of domestic animals in Nigeria (AYENI et al., 1985; OJEH, 1986; JACOB et al., 1988; BIU and NWOSU, 2000), with reports on the organism in different parts of the country, there is, however, a dearth of information on the prevalence of the parasite among the captive wildlife population in Nigeria, despite our extensive search of the literature. Therefore, in this paper we present the prevalence of cryptosporidiosis for the first time among captive wild animals and birds in the arid region of north-eastern Nigeria.

Materials and methods

Study area. Animals used for this study were made available for sample collection by the management of Sanda Kyarimi Park, Maiduguri, Borno State. A total of 36 different species of wild animals and birds were examined during the study.

Sample collection. Faecal samples were collected from 66 animals (out of 36 species) either per rectum or adventitiously from the ground as fresh as possible, and immediately taken to the laboratory. Each sample was properly labelled with information on the age and sex of the animal from which the sample was taken.

Faecal smear. Smear from the faecal sample of each animal was prepared on 3 different glass slides. The fresh faecal sample was mixed with a fresh physiological saline solution using mortar and pestle, and then sieved. Thin smears were made from the faecal solution on 3 different glass slides, and dried. Smears were fixed using methanol for 3-5 min.

Staining procedure. Three different staining methods were employed in this study: Safranin-methylene blue staining technique, Giemsa staining technique, and modified Ziehl-Neelsen staining technique as described by GARCIA et al. (1983), ARROWOOD and STERLING (1989) and CASEMORE et al. (1989), respectively.

Results

The prevalence of *Cryptosporidium* oocyst in different age groups of captive wild animals and birds is presented in Table 1. In all, a total of 66 wild animals and birds were examined. On an age basis, 15 (22.7%) of these animals and birds were classified as young, while the remaining 51 (77.3%) were classified as adults. Faecal samples from four different orders - *Artiodactyl*a/*Proboscidae*, carnivores, primates and avian - were examined. Five (16.7%) samples tested positive of the 30 samples examined under the order *Artiodactyla/Proboscidae*. The positive samples comprised 4 (18.2%) adults and 1 (12.5%) younger animal. A total of 5 (50.0%) samples out of 10 were positive for *Cryptosporidium* oocyst in carnivores. The positive samples comprised 3 (42.9%) from adult animals and 2 (66.7%) from younger animals. Of the 5 primates examined, 2 (40.0%) were found positive. These comprised the only 1 (100.0%) younger animal and an further 1 (25.0%) from the older animals. Three (14.3%) samples from the avian group were found positive out of the 21 samples tested and 1 (33.3%) was from the younger age group, while 2 (11.1%) were from adults.

The effect of sex on the prevalence of *Cryptosporidium* oocyst in captive wild animals and birds is presented in Table 2. Of the 66 animals and birds examined, 36 (54.5%) were males while the remaining 30 (45.5%) were females. Of the 30 *Artiodactyla/proboscidae* tested, 5 (16.7%) positive samples were detected, which include 4 (28.6%) males and 1 (5.6%) female. Of the 10 carnivores examined, 3 (50.0%) samples from males and 2 (50.0%) from females were found positive for *Cryptosporidium* oocyst of the total number of 5 (50.0%) positive cases. The 5 primates sampled comprised 4 males and 1 female. Two (50.0%) of the 4 males were found positive, while the only female tested negative for *Cryptosporidium* oocyst. Three (14.3%) samples of the 21 birds examined tested positive and 1 (8.3%) of the positive samples was from a male bird, while the other 2 (22.2%) were from female birds.

The sensitivity of different staining methods used in detecting *Cryptosporidium* oocyst from faecal samples of captive wild animals and birds was studied (Table 3). Three

different staining methods were used in the study: Giemsa, Safranin-methylene blue, and the modified Ziehl-Neelsen. A total of 66 faecal samples obtained from different wild animals and birds in captivity were each tested using the 3 staining methods mentioned above. The modified Ziehl-Neelsen stain recorded the highest number of positive samples 15 (22.7%), while the Giemsa stain recorded 12 (18.2%) positive samples. The Safranin-methylene blue stain recorded the least number, 12 (12.1%), of positive samples detected.

Table 1. Prevalence of *Cryptosporidium* oocyst among different age groups of different species of captive wild animals and birds

Species of animals examined	Total number examined	Number (%) positive Total number	
		Young	Adult
Artiodactyla/Proboscidae	30	1/8 (12.5)	4/22 (18.2)
Carnivores	10	2/3 (66.7)	3/7 (42.9)
Primates	5	1/1 (100)	1/4 (25)
Avians	21	1/3 (33.3)	2/18 (17.7)
Total	66	5/15 (7.5)	10/51 (15.2)

Table 2. Prevalence of *Cryptosporidium* oocyst among different sexes of different species of captive wild animals and birds

Species of animals examined	Total number examined	Number (%) positive Total number	
		Males	Females
Artiodactyla/Proboscidae	30	4/14 (28.6)	1/16 (5.6)
Carnivores	10	3/6 (50)	2/4 (50)
Primates	5	2/4 (50)	0/1 (0)
Avians	21	1/12 (8.3)	2/9 (22.2)
Total	66	10/36 (15.2)	5/30 (7.6)

Table 3. Comparison of sensitivity of the different staining methods in detecting *Cryptosporidium* oocyst in faecal samples of captive wild animals and birds

Types of stain	N° of samples tested	N° of positive samples	Prevalence (%)
Safranin-methylene blue stain	66	8.0	12.1
Modified Ziehl-Neelsen stain	66	15.0	22.7
Giemsa stain	66	12.0	18.2

Discussion

The result of this study revealed that *Cryptosporidium* species occurs in almost all species of captive wild primates, carnivores, *Artiodactyla/Proboscidae* and birds in the Sanda Kyarimi Park, located in the semi-arid region of Nigeria. Although there has been no prior reports on such a study among wild animal species to evaluate their reservoir status, high prevalence rates have been recorded among domestic animals at Ile-Ife in the humid south-western part of the country (AYENI et al*.,* 1985) and in the semi-arid region of north-eastern Nigeria (BIU and NWOSU, 2000).

Despite previous reports of high prevalence of the parasite among both diarrheic and non-diarrheic domestic animals (KENNEDY et al., 1977; POHLENZ et al., 1978; TZIPORI et al., 1980a; LINKS, 1982; AYENI et al., 1985; BIU and NWOSU, 2000), this study however reports all the cases among wild animals that showed no form of diarrhoea (non-diarrheic) or immunosuppression as reported in foals (SNYDER et al., 1978). This suggests that wild animals may possibly serve as reservoirs to *Cryptosporidium* and may be a potential source of infection to both domestic animals and human attendants on the one hand, and human visitors on the other.

Although certain adverse conditions of stress, such as those which obtain in captivity, have often compromised the existing reservoir status, leading to overwhelming infection as reported in a captive gray squirrel (JOHN et al., 1982) and in raccoon (BRENT et al., 1982), in the present study, results did not concur with previous findings (SNYDER et al., 1978; AYENI et al., 1985; ARROWOOD and STERLING, 1989) in which significant differences in prevalence of infection between adult and young domestic animals was reported. The results also showed that age and sex of the animals and birds under this study have no significant effect on the prevalence of *Cryptosporidium* infection among captive wild animals and birds.

Furthermore, the results achieved from the evaluation of different staining techniques (Giemsa, Safranin-methylene blue and modified Ziehl-Neelsen) agree with findings from previous works (BIU and NWOSU, 2000), which scored the modified Ziehl-Neelsen as most sensitive. Next to the modified Ziehl-Neelsen is Giemsa and, lastly, Safranin-methylene blue was rated sensitive in descending order. Unlike in the previous study (BIU and NWOSU, 2000) where Safranin-methylene blue and Giemsa stains were rated sensitive next to modified Ziehl-Neelsen in descending order, in this study, however, Giemsa stain was found to be the most sensitive, next to modified Ziehl-Neelsen, followed by Safranin-methylene blue, in descending order. As no immediate reason for the differences in sensitivity could be offered, however, the fact that the previous study was conducted on domestic animals, while the present study was conducted on wild animals, could suggest further investigation into the effects of these stains on different samples and tissue types of wild animals. Early reports of cryptosporidial infection in Nigeria showed the occurrence

of the parasite in domestic animals. This study reports for the first time the prevalence of infection with the parasite in captive wild animals and birds in Nigeria. Therefore, captive wild animals and birds could also play a role in the epidemiology of cryptosporidiosis as a zoonotic infection.

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SAŽETAK

Kriptosporidioza u divljih sisavaca i ptica držanih u zatočeništvu u sušnom području u parku Sanda Kyarimi u Maiduguriju u sjeveroistočnoj Nigeriji određena je koprološkom pretragom dokazivanjem oocista različitim metodama bojenja. Rabljeno je preinačeno bojenje po Ziehl-Neelsenu, po Giemsi te bojenje safranin-metilenskim plavilom. Ukupno je pretraženo 66 divljih sisavaca i ptica. Za sve su životinje zabilježeni podatci o spolu i dobi. Pretraženo je 22,7% mladih te 77,3% starih životinja te 54,5% mužjaka i 45,5% ženki. Nije dokazano da dob i spol utječu na učestalost kriptosporidioze u pretraženih životinja. Preinačenim postupkom po Ziehl-Neelsenu dokazano je ukupno 15 (22,7%), bojenjem po Giemsi 12 (18,2%) te bojenjem safranin-plavilom 8 (12,1%) pozitivnih uzoraka. Istraživanje je pokazalo da različite divlje životinje i ptice mogu imati značajnu ulogu u epidemiologiji kriptosporidioze.

Ključne riječi: kriptosporidioza, divlje životinje, zoonoza, Nigerija