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OCCURRENCE OF *CRYPTOSPORIDIUM* OOCYSTS IN FAECES, MILK AND WATER SOURCES IN SEDENTARY FULANI HERDS IN SELECTED LOCAL GOVERNMENT AREAS OF KADUNA STATE, NIGERIA

Daniel O. Esonu[1], Junaidu Kabir[1], Isa D. Jatau[2], Mohammed K. Lawan[1], Mohammed S. Yusuf [1], Mohammed B. Aliyu[1], Fatima L. Yusuf [1]

[1] Department of Veterinary Public Health and Preventive Medicine, [2,3] Department of Veterinary Parasitology and Entomology, Faculty of Veterinary Medicine, Ahmadu Bello University, Zaria Kaduna State Nigeria

doesonu@abu.edu.ng

ABSTRACT

Cryptosporidiosis is a neglected tropical zoonotic disease, commonly associated with a diarrhoea, caused by a protozoan parasite of the genus *Cryptosporidium*. The aim of this study was to determine the occurrence of *Cryptosporidium* oocysts in faeces, milk, and water sources, and its associated risk factors in sedentary Fulani herds. Seven hundred and fifty faecal samples (240, 180, 240 and 90 from cattle, sheep, goats and dogs, respectively), and 120 samples each, of milk and water were collected for this study. Faecal samples were concentrated by formol ether concentration technique, then stained using Modified Ziehl Neelsen staining method, to detect *Cryptosporidium* oocysts. *Cryptosporidium* oocysts in milk and water samples were also concentrated and stained with same stain. The prevalence of *Cryptosporidium* oocysts in (cattle, dog, sheep and goat faeces were 18.3 %, 15.6 %, 13.9 % and 4.2 %, respectively.), Significantly higher prevalence was observed in calves ≤ 1 year (P = 0.002), female cattle (P = 0.007) and animals with poor body condition (P<0.001) than in the other groups. Goats with poor body condition scores had significantly higher prevalence (P = 0.008) than other groups while dogs ≤ 6 months of age also had significantly higher prevalence (P = 0.03) than older dogs. The prevalence of *Cryptosporidium* oocysts in bulk cow milk was 11.7 % while a prevalence of 13.3 % was observed in the water bodies sampled. Hence the findings of this study are of great public health significance, therefore, inhabitants of these LGAs should be informed and educated on the need for improvement of sanitary measures during the course of milking these animals, and the need for adequate pasteurization, of milk before consumption.

Key words: cattle; *Cryptosporidium*; dogs; goats; milk; oocysts; sheep; water

INTRODUCTION

Cryptosporidium is a pervasive protozoan parasite found in the small intestine and stomach of vertebrates

[13]. Infection occurs through the faecal-oral route, either by direct contact, or by consuming contaminated food or water. Human and animal cryptosporidiosis typically presents with diarrhoea, dehydration, fever, nausea, and anorexia [58]. The transmission of *Cryptosporidium* to humans involves various mechanisms, including human-to-human, animal-to-human, and environmental transmission through water and food [35]. The parasite has a life cycle involving a phase of proliferation on the mucosal surface and a reproductive phase. The infective stage, released in faeces, remains encysted, and can survive in the environment for an extended period [12].

Initially described as an intracellular parasite in the gastric glands of mice, *Cryptosporidium* gained public health significance during the AIDS pandemic as an opportunistic parasite affecting immune-compromised individuals [22]. In livestock, *Cryptosporidium* causes significant morbidity and mortality, particularly in young animals, leading to economic losses [41]. It is a major enteric pathogen associated with neonatal diarrhoea and mortality in sheep and goats [34]. The environmentally resistant oocysts shed in faeces are resistant to water disinfectants [18]. Dairy products, consumed globally on a daily basis, pose a major concern for food-borne illnesses [11]. Although oocysts are not excreted in milk, but milk can become contaminated, if faecal matter containing *Cryptosporidium* oocysts comes into contact with it during milking. Contamination of fresh milk can occur during processing due to various factors, including udder health, milking equipment, and handling practices [10]. *Cryptosporidium parvum* oocysts are commonly found on dairy farms, and can be transmitted to humans through contaminated raw milk and dairy products, presenting a risk to vulnerable individuals. Effective control strategies are crucial to managing this sanitary risk.

Compared to most human pathogens, *Cryptosporidium* is relatively stable in environmental waters, remaining infectious for weeks or months at typical surface water temperatures [38]. The fact that waterborne outbreaks occur demonstrates that human-pathogenic oocysts can survive in environmental waters, and retain their infectivity, but the amount of attenuation due to combined environmental stressors is not known. Oocysts are very sensitive to inactivation by UV-C irradiation [26] and it has been suggested that sunlight may be the most significant inactivating agent in environmental waters. Water temperature is also a critical factor in oocyst survival in the environment [39].

Studies on cryptosporidiosis in various locations have reported its prevalence in humans, piglets, cattle, birds, and raw vegetables [2, 4, 12, 15, 47]. The prevalence of *Cryptosporidium* in Fulani settlements, where fresh milk is a staple, is yet to be determined. Hence, this work aims to investigate the possibility of milk contamination during milking in Fulani settlements. For a comprehensive understanding of *Cryptosporidium* epidemiology in Fulani settlements, the prevalence of oocysts in livestock and dogs' faeces, raw cow milk, and water sources were examined. Additionally, associated risk factors contributing to the transmission of these infections in humans and animals were assessed. The study also evaluated the impact of physico-chemical parameters of water on the prevalence of *Cryptosporidium* oocysts in water sources in selected Local Government Areas of Kaduna State, Nigeria.

MATERIALS AND METHODS

Study area

The study was carried out in six (6) selected Local Government Areas (LGA) of Kaduna State, Nigeria; namely, Zaria, Sabon Gari, Giwa, Igabi, Soba and Kudan.

Study design

This was a cross-sectional study in which 240, 180, 240 and 90 faecal samples were collected from cattle, sheep, goats, and dogs, respectively, in sedentary Fulani herds in selected Local Government Areas of Kaduna State, Nigeria from May to September, 2021 (Table 1). Five (5) cattle herds were selected from each of the six LGAs based on inclusion criteria as follows: (1) Settled herds of ≥ 20 cattle capacity (2) Herds which produce and sell raw cow milk for public consumption (3) Herds that rear cattle with other ruminants (sheep or goat), or at least a dog. Based on these criteria thirty (30) cattle herds were selected for this study. Eight (8) cattle in each herd were selected using systematic random sampling. A total of 30, 40 and 15 faecal samples were collected from sheep, goats and dogs respectively, in each local government area while samples in each herd were collected based on availability. One milk sample was collected from the bulk milk in each cattle herd (30 herds) selected for the study during visit weekly for four (4) weeks, giving a total of 120 milk samples. Water sources (n = 30) within the vicinity of each

cattle herd or water bodies where cattle from different herds congregate to drink were sampled weekly for four (4) weeks, i.e. 120 water samples from the thirty (30) identified water sources.

Ethical clearance and consent

An ethical clearance was obtained from the Ahmadu Bello University Committee on Animal Use and Care (ABUCAUC). Samples were collected only with the farmers' consent and willingness to participate in the research.

Sample collection

About 5–10 g (based on the specie of animal) of faeces were collected directly from rectum of randomly selected animals using clean disposable rubber hand glove and emptied into sterile sample bottles. About 10 ml of milk was collected into clean sterile sample bottle from the bulk milk. Approximately 5 litres of water were collected into plastic containers. Primary sampling points for water bodies such as dams, rivers, streams, ponds, etc., were slightly below the surface water layer at the centre of the main flow; this was to avoid collecting floating dust, oil, etc. All samples were properly labelled and transported on an ice packed cold box to the Parasitic Zoonoses Laboratory, Department of Veterinary Public Health and Preventive Medicine, Ahmadu Bello University, Zaria.

Laboratory analysis

Microscopic analysis of faecal samples

The faecal samples were analysed using formol ether concentration technique as described by Arora and Brij [5]. The presence of *Cryptosporidium* oocysts was detected using the modified Ziehl-Neelsen staining technique as described by Clarke and McIntyre [14]. *Cryptosporidium* oocysts appear as pink to red, spherical to ovoid bodies against a green to purple background [57]. Samples were considered positive, if at least one morphologically distinct *Cryptosporidium* oocyst was observed.

Microscopic analysis of milk samples

Detection of *Cryptosporidium* oocysts in milk samples was carried out by the method described by Machado et al. [31]. Briefly, 5 ml of milk sample was placed in a 15 ml centrifuge tube and 5 ml of magnesium sulphate ($MgSO_4$) solution (density 1.30 $g.ml^{-1}$; 750 $g.1^{-1}$) was added. This was stirred vigorously, and centrifuged at 206 x g for 10 minutes. Three layers were formed in the tube; supernatant at the topmost layer, milk clot at the intermediate layer and sediment at the bottom of the tube. The supernatant was carefully decanted into a washing basin, while the resulting clot was transferred into a tube where it was washed with distilled water. The clot was re-suspended in 8 ml of distilled water, where it was dissolved with an applicator stick and sieved. The filtrate was centrifuged, at 206 x g for 10 minutes. Milk smears were made with 5 µl of both the supernatant and sediment. The modified Ziehl-Neelsen staining technique [14] was also used to detect the presence of *Cryptosporidium* oocysts in the milk samples.

Analysis of water samples

The temperature of the water samples were taken using a thermometer at the point of sample collection. Also, the hydrogen ion concentration (pH) of each sample was measured using a digital pH meter. The amount of dissolved oxygen was determined using the method described by Ologbosere et al. [46]. Turbidity in the water samples was determined using a spectrophotometer. The concentration of *Cryptosporidium* oocysts in water samples was determined using the technique described by Njoku et al. [40]. The sediments obtained were transferred to microscope slides and smears were made. Modified Ziehl-Neelsen staining technique was also used to detect the presence of *Cryptosporidium* oocysts in the water samples.

Data analysis

Data from the study were analysed using a Statistical Package for Social Science (SPSS) version 20.0 (Standard Version SPSS Inc., Chicago, IL, USA). Chi-square or Fisher's exact test was used, as appropriate to test for association between *Cryptosporidium* species and factors such as age, sex, breed, consistency of faeces and body condition score. Chi-square and Fisher's exact test was also used to determine the effect of the physico-chemical parameters of water on the occurrence of *Cryptosporidium* oocysts in water sources sampled. P values ≤ 0.05 were considered significant.

RESULTS

The overall prevalence of *Cryptosporidium* oocysts (Fig. 1) in cattle faeces was 18.3 % (44/240) (Table 2). Higher prevalence of *Cryptosporidium* oocysts (28.9 %)

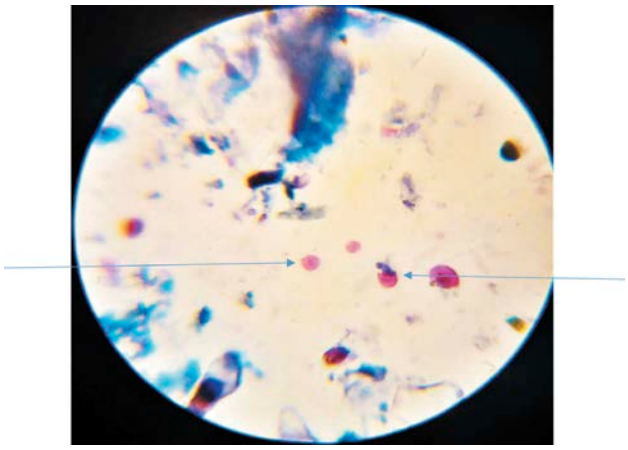

Fig. 1. Photomicrograph of *Cryptosporidium* oocysts in cattle faeces. Arrows showing *Cryptosporidium* oocysts using Modified Ziehl Neelsen staining method. Magn. x 400.

was recorded in cattle ≤ 1 year of age than in cattle > 1 year of age (12.7 %). There was a statistically significant difference (P = 0.002) between the prevalence of *Cryptosporidium* oocysts and the different age groups sampled. Significantly (P = 0.007) higher prevalence of *Cryptosporidium* oocysts (23.9 %) was observed in female cattle than in male cattle (10.2 %). The highest prevalence of 32.3 % (21/65) *Cryptosporidium* oocysts was observed, in cattle with poor body condition (score 2), followed by those with medium body condition (score 3) with a prevalence of 25.4 % (16/63). The lowest prevalence was recorded in cattle with good body condition (score 4) which had a prevalence of 6.3 % (7/112). There was a significant difference (P < 0.001) between the prevalence of *Cryptosporidium* oocysts, and the body condition scores of the animals sampled. Table two also shows that the highest prevalence of 27.5 % (11/40) of *Cryptosporidium* oocysts was observed in cattle from Sabon Gari and Giwa LGAs, followed by those sampled in Zaria and Kudan LGAs which had the same prevalence of 22.5 % (9/40) of *Cryptosporidium* oocysts. Statistical analysis showed a significant difference (P = 0.009) in the prevalence of *Cryptosporidium* oocysts in cattle faeces and LGAs.

All of the 30 herds sampled and studied belonged to nomadics who settle in a particular area for a period of time, then migrate at the end of each season. A higher prevalence of *Cryptosporidium* oocysts (45.8 %) was observed in herds with >100 cattle compared to herds with 50–100 animals with a prevalence of 20.3 %. Statistical analysis showed a significant difference (P = 0.001) between the prevalence of *Cryptosporidium* oocysts in cattle faeces and the herd size (Table 3). Furthermore, a higher prevalence was observed in herds with 21–40 lactating cattle (39.3 %), compared to those with 0–20 lactating cattle (12.0 %), which was also significantly different (P < 0.001). A higher prevalence of *Cryptosporidium* oocysts (27.1 %) was observed in herds that mostly utilized ponds than in herds that used river (12.5 %) as a major source of water. This difference in the prevalence was statistically significant (P = 0.004).

The prevalence of *Cryptosporidium* oocysts in sheep (Table 4) and goats (Table 5) were 13.9 % (25/180), and 4.2 % (10/240), respectively. Although, there was no significant difference (P = 0.210) between the prevalence of *Cryptosporidium* oocysts and body condition scores in sheep, a significant difference (P = 0.008) was observed in goats. There was a significant difference (P < 0.001) between the prevalence of *Cryptosporidium* oocysts in sheep and LGAs sampled. The prevalence of *Cryptosporidium* oocysts in dog faeces was 15.6 % (14/90) (Table 6). A higher prevalence of *Cryptosporidium* oocysts was observed in dogs ≤ 6 months (27.6 %) of age than in dogs older than 6 months (9.8 %). There was a significant association (P = 0.03) between the prevalence of *Cryptosporidium* oocysts and the age of the dogs sampled.

The prevalence of *Cryptosporidium* oocysts in bulk milk was 11.7 % (14/120) (Table 7), while the prevalence of *Cryptosporidium* oocysts observed in water samples was 13.3 % (16/120) (Table 8). Furthermore, prevalence of 28.6 % was observed in water samples with pH range of 6.0–6.9 while no *Cryptosporidium* oocysts was observed in those with pH range of 7.0–7.9. There was statistically significant difference (P < 0.001) in prevalence of *Cryptosporidium* oocysts in water samples with different pH. Higher prevalence was observed in water samples with turbidity range of 0.601–0.900 NTU (33.3 %) than in samples with range of 0.301–0.600 NTU (17.6 %). Also, there was statistically significant difference between the prevalence of *Cryptosporidium* oocysts, and the turbidity of the water sampled.

DISCUSSION

The prevalence of *Cryptosporidium* oocysts recorded in this study is comparable with that of O k o j o k w u et al. [43] and M a i k a i et al. [32] who reported 15.7 % in

cattle slaughtered in abattoir and 16.0 % in native breeds of cattle in Kaduna State, respectively. The prevalence reported in this study is lower than the prevalence of 28.0 % reported by Danladi and Ugbomoiko [15] in Kebbi State, 22.3 % by Adamu et al. [2] in Maiduguri, Borno State, 33.0 % by Faleke et al. [21] in Sokoto State, 23.4 % among cattle in Western Nigeria [10] and 28.0 % by Pam et al. [47] in Jos, Plateau State. The prevalence of *Cryptosporidium* oocysts in sheep recorded in this study is also comparable with the prevalence of 11.7 % reported by Danladi and Ugbomoiko [15] in Kebbi State but the prevalence from this study in goats is lower than 17.1 % reported by them [15] in Kebbi State. These disparities in prevalences in these livestock could be due to differences in the season of sampling, geographical locations, study design, management system, ages of animals sampled, species of animals, herd size and laboratory techniques employed. *Cryptosporidium* is considered one of the most important parasitic diseases in terms of morbidity and is one of the most common gastrointestinal pathogen in children with severe retardation of growth and development [13]. Hence the findings of this study are of great public health significance as these groups of animals sampled produce milk for consumption in both urban and rural communities, thus raising the possibility of contamination of the product in the course of milking. This also indicates that cryptosporidiosis is a problem affecting a wide range of farm animals and age groups in the study location. The occurrence of this infection in ruminants, as revealed by this study, may be attributed to factors such as poor sanitation and unhygienic management of farms in the study area, that expose these animals to the risk of infection. The unavailability of potable water for these animals as well as contamination of grazing pasture by faeces of carrier animals and humans, which is a common practice in developing countries, might also be additional factors that contribute to such prevalence.

The prevalence of *Cryptosporidium* oocysts observed in dogs in this study was higher than the 4.3 % prevalence reported in Ibadan [27], the 5.4 % reported in Abuja [44] and 5.9 % reported in Zambia [37]. This disparity may be due to breed, management system and the reason for keeping the dogs, because in this study, the dogs sampled were mostly guard/hunting dogs that most times scavenge for themselves thus increasing their chances of infection. *Cryptosporidium* infection in these guarding/hunting dogs may cause cryptosporidiosis in humans due to zoonotic transmission of the infection through close contact and may also serve as sources of transmission to ruminants.

The higher prevalence of *Cryptosporidium* infection in younger animals as compared to the adults may occur because calves are less immune to infection as immunity acquired during birth from the dams gradually wanes off making the calves vulnerable to infections [9]. Higher prevalence in younger animals is in accordance with the findings of Santin et al. [50], Fayer et al. [23], Pam and Anwuliri [48] and Maikai et al. [32]. The infection might have resulted from environment, milk and water contamination by carrier dams. Ralston et al. [49] observed that the geometric mean number of *Cryptosporidium* oocysts in the infected cow faeces increased significantly to 37.48 oocysts per g at one-week post-calving then decreased with age, thus they act as carriers of infections for calves. This outcome may also be attributed to higher susceptibility of calves to infection due to lack of previous exposure to the parasite [7]. The observation of higher infection rate in goats below 6 months corroborates the report of Pam et al. [47]. This observation may be associated with the underdeveloped immune system of these goat kids [47], and the husbandry/management system employed in Nigeria, in which goat kids are housed with adults in a pen thereby facilitating transmission of infection. Infection rate was higher in younger dogs than in older ones, which was in accordance with the findings of Lorezini et al. [30] and Titilincu et al. [55]. This might be due to the immune-incompetence of the younger dogs probably, as a result of low level of passive immunity received from their dams as suggested by Oliveira-Sequeira et al. [45].

The results of this study also revealed a regular pattern of higher prevalence among females than males. The consistency of higher prevalence in females among the cattle and goats may be explained by stress associated with hormonal imbalances during pregnancy and lactation in the adult females. The higher prevalence rates recorded in females resemble various reports on different animal species in Nigeria [7, 47].

Higher prevalence in local breed of dogs compared to crossbreeds could be due to the higher number of local dogs sampled, as exotic/cross breed were rarely found in local Fulani settlements. This could also be due to the less attention/care given to local breeds which are left to fend for themselves. They roam about and eat anything such as

human excreta in diapers, dead animals and birds exposing them to chances of infection. The infection rate in local breeds sampled in this study was higher than that reported by Johnson and Joseph [27] and Olabanji et al. [44], who observed an infection rate of 4.3 % in Ibadan and 4.0 5 % in Abuja, respectively. This difference might be due to season of study and the reason for keeping the dogs, as these studies involved mostly dogs kept for security purposes in households and as pets that received more care and their chances of exposure were reduced.

The highest prevalence of *Cryptosporidium* infection was observed in some species of animals with poor body conditions, followed by those with medium body conditions, and the lowest prevalence was recorded in most animals with good body condition scores. These findings could suggest the importance of cryptosporidiosis in causing weight loss and emaciation due to diarrhoea which is a characteristic sign of the disease. When cattle are infected with *Cryptosporidium*, they often experience diarrhoea, which, can be severe and persistent. This diarrhoea leads to fluid loss and electrolyte imbalance, ultimately resulting in weight loss and emaciation [23]. Besides, the high infection rate in animals with poor condition could be justified by the fact given by Taylor et al. [53] who indicated that animals with poor body condition are vulnerable to parasitic diseases. Sweeny et al. [52] stated that the infection can affect production, with reduced body condition score, reduced growth rate, and reduced carcass weight and dressing percentage at slaughter [52].

Higher infection rates were observed in animals (cattle, sheep, goats and dogs) with loose/watery faeces than those passing out firmly formed faeces. This observation is in agreement with the studies by Akinkuotu et al. [3, 4], Danladi and Ugbomoiko [15] and Abare et al. [1]. *Cryptosporidium* has been reported as a major enteric parasite associated with neonatal diarrhoea and mortality in lamb and goat kids [16, 19]. It has been suggested that the pathogenesis of *Cryptosporidium* infection alongside other enteric infections, such as rotavirus, *Salmonella, Escherichia coli*, *Eimeria*, *Giardia,* etc., are very likely to result in such diarrhoeic condition [6, 20, 33, 42, 54]. Characteristic diarrhoea is thought to result from maldigestion and malabsorption, due to reduction in both enzymatic action and absorptive area in the gastrointestinal tract owing to diminution of microvilli and destruction of intestinal epithelia by *Cryptosporidium*. An increase in paracellular permeability of the intestinal tract and destruction of the functional mucosal barrier system are both, as a result of the damage caused by the parasite [28]. Foster and Smith [24] had explained that *C. parvum* infection in calves have shown that jejunum and ileum is mainly affected and the diarrhoea occurs either due to hindrance in sodium absorption coupled with increased prostaglandin production in the intestinal mucosa or owing to increased permeability. It could also be due to *Cryptosporidium* oocysts inhabiting the inner lining of the small intestine, where excystation takes place releasing motile tropozoites and due to the migratory habits of these tropozoites along the villi of the small intestine, there is irritation, and sloughing off of the lining of the intestine which is seen as diarrhoea.

Higher prevalence was observed in Sabon Gari, Giwa and Kudan LGAs across all animal species except goats sampled in Kudan LGA. This may be attributed to the wetness and high humidity along the river banks, ponds or lakes observed around these areas encouraging oocysts survival and increasing the chances of infection. The present finding on the distribution of *Cryptosporidium* oocysts in cattle, sheep, goats and dog faeces across the six LGAs has significance because *Cryptosporidium* is zoonotic and has public health importance in terms of gastrointestinal protozoan disease.

Prevalence was significantly higher in herds with >100 cattle capacity and herds that have 21–40 lactating animals at the point of sampling. This could be due to the high stocking density in the herd, which prevents proper cleaning and disinfection, leading to horizontal spread of infections with protozoan parasites. This may also be due to poor management and poor sanitation sometimes associated with managing herds of large number of cattle. Moreover, the significantly higher prevalence observed in herds that utilized ponds than rivers as the primary source for drinking could be due to the fact that ponds are stagnant and not free-flowing, thus favouring the survival of the infective oocysts.

The occurrence of *Cryptosporidium* oocysts in milk sampled from all the LGAs as revealed by this study may be attributed to factors such as poor sanitation and unhygienic management of herds in the study area, thus exposing humans to the risk of infection. Although the prevalence observed in the study is lower than that reported by Hasan et al. [25] in Nineveh, Iraq, who showed that the incidence rate of *Cryptosporidium* oocysts in ovine

raw milk was 32.0 % while that in caprine milk was 46.0 % using direct immunofluorescence assay which is more sensitive. Others reported that infection rates of *Cryptosporidium* in sheep and goats' milk were 35.1 % and 26.4 % respectively [8] in Iraq using microscopy. M o s a [36] in Egypt found that incidence of *Cryptosporidium* species in sheep and goat milk samples based on microscopic examination were 31.43 % and 20 %, respectively. Variations in incidence rates of *Cryptosporidium* spp. oocysts were presented according to type of rearing management, geographical location and season of the study. The occurrence of oocysts in milk comes from the shedding of oocysts in the faeces of these animals leading to contamination of udder and subsequently the milk in the course of milking process [17].

This study revealed occurrence of *Cryptosporidium* oocysts in water bodies around sedentary herds in the selected Local Government Areas (LGAs) of Kaduna State, Nigeria. There is the possibility of faecal contamination, when rainwater wash off *Cryptosporidium* oocysts from faecal deposition in the environment into the surface water sources where animals drink freely. This could be one of the factors influencing the numerical increase of oocysts in the environment. This finding is in conformity with the 10 % prevalence reported by L a w a l [29] in dams within Zaria using microscopy. However, this prevalence is lower than previous reports which noted that the oocysts of *Cryptosporidium* species were continually present in virtually all surface waters [56] and 33.2 % reported by S i m o n – O k e et al. [51]. This variation could be due to season of the study (rainy or dry season), geographical location and type of water bodies (i.e. river, pond, stream, etc.). The observation that occurrence was significantly higher in pH ranges of 6.0–6.9 than those of 7.0–7.9 could be due to high quantity of organic matter/manure containing oocysts being washed from agricultural lands into these water bodies thereby reducing the pH due to the presence of organic acids in manure. Likewise, the higher occurrence observed in water bodies with 0.601–0.900 NTU than in other turbidity ranges may be due to high concentration of contaminants washed from the environment which might contain eggs/oocysts of parasites. This finding is also in conformity with report of S i m o n - O k e et al. [51], who observed that the occurrence of the *Cryptosporidium* oocysts in the water sources was affected by the water parameters such as temperature, pH, and turbidity.

CONCLUSIONS

The findings in this study are of veterinary and public health importance and they include:

Firstly, the prevalence of *Cryptosporidium* oocysts in cattle, sheep, goats and dog faeces were 18.3 %, 13.9 %, 4.2 %, 15.6 %, respectively, while the prevalence of *Cryptosporidium* oocysts in bulk milk and water bodies were 11.7 % and 13.3 %, respectively,

Secondly, the risk factors associated with the occurrence of *Cryptosporidium* infection in the animal species sampled for this study were as follows: cattle, age ($P = 0.002$); sex of the animals ($P = 0.007$); as well as body condition scores ($P < 0.001$) and local government areas (LGA) sampled ($P = 0.009$); sheep, LGAs sampled ($P < 0.001$); goats, only body condition score ($P = 0.008$); dogs, age ($P = 0.03$) were identified as risk factors,

Lastly, physico-chemical parameters such as pH ($P < 0.001$) and turbidity ($P = 0.003$) had a significant effect on the occurrence of *Cryptosporidium* oocysts in the water bodies.

We therefore recommend that given the prevalence status of *Cryptosporidium* oocysts in various animal species and environmental sources like bulk milk and water bodies, there should be increased monitoring and management practices implemented in livestock farming and water sanitation systems to reduce the contamination levels. Addressing the identified risk factors associated with *Cryptosporidium* infection in different animal species in this study is crucial and implementing targeted interventions, such as vaccination programs and improved animal husbandry practices, can help mitigate these risks.

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Table 1. Distribution of samples

Type of sample	Number of LGAs sampled	Number sampled in each LGA	Number of herds sampled in each LGA	Number of samples taken from each herd	Total sample for the study
Cattle faeces	6	40	5	8	240
Sheep faeces	6	30	5	BOA	180
Goat faeces	6	40	5	BOA	240
Dog faeces	6	15	5	BOA	90
Milk	6	20	5	4	120
Water	6	20	5	4	120

BOA – Based on availability

Table 3. Occurrence of *Cryptosporidium* oocysts in cattle and Livestock Husbandry practices of herdsmen in sedentary Fulani herds in selected Local Government Areas (LGAs) of Kaduna State, Nigeria

Practices	Number of herds (%)	Number of cattle examined	Number positive (%) for oocysts	χ2/Fishers exact	P
Number of herdsmen/herds					
One	9 (30.0)	72	8 (11.1)	3.77	0.152
Two	17 (56.7)	136	30 (22.1)		
Three	4 (13.3)	32	6 (18.8)		
Number of cattle in the herds					
<50	19 (63.3)	152	20 (13.2)	15.009	0.001
50-100	8 (26.7)	64	13 (20.3)		
>100	3 (10.0)	24	11 (45.8)		
Number of lactating animals					
< 20	23 (76.7)	184	22 (12.0)	21.417	< 0.001
21–40	7 (23.3)	56	22 (39.3)		
Other animals on the farm					
Sheep	15 (50.0)	120	20 (16.7)	1.085	0.897
Sheep and goats	6 (20.0)	48	9 (18.8)		
Sheep and dogs	6 (20.0)	48	11 (22.9)		
Dogs	2 (6.7)	16	3 (18.8)		
Sheep, goats and dogs	1 (3.3)	8	1 (12.5)		
Supplementary feeding					
Locally compounded	5 (83.3)	40	8 (20.0)	0.089	0.765
None	25 (16.7)	200	36 (18.0)		
Source of drinking water					
River	18 (60.0)	144	18 (12.5)	8.182	0.004
Lakes/ponds	12 (40.0)	96	26 (27.1)		
Any water treatment before giving to cattle to drink?					
Yes	0 (0.0)	0	0 (0.0)	-	-
No	30 (100.0)	240	44 (100.0)		
Animal on medication					
Yes	5 (16.7)	40	2 (5.0)	5.699	0.017
No	25 (83.3)	200	42 (21.0)		
Type of medication					
Antibiotics	3 (10.0)	24	1 (4.2)	5.727	0.057
Antiparasitic	2 (6.7)	16	1 (6.3)		
None	25 (83.3)	200	42 (21.0)		
Contact with other herds					
Yes	30 (100)	240	44 (18.3)	-	-
Nature of milking floor					
Soil	30 (100)	240	44 (18.3)	-	-
Milking environment					
Open field	30 (100)	240	44 (18.3)	-	

Table 2. Prevalence of *Cryptosporidium* oocysts in cattle faeces in sedentary Fulani herds in selected Local Government Areas of Kaduna State, Nigeria

Variables	Category	Number examined	Number positive	Specific rate (%)	χ2/Fishers exact	P
Age (years)	≤ 1	83	24	28.9	9.490	0.002
	> 1	157	20	12.7		
Sex	Male	98	10	10.2	7.311	0.007
	Female	142	34	23.9		
Breed	White Fulani	190	38	20.0	2.226	0.329
	Sokoto Gudali	45	6	13.3		
	Red Bororo	5	0	0.0		
Body condition score	Poor	65	21	32.3	21.499	< 0.001
	Medium	63	16	25.4		
	Good	112	7	6.3		
Consistency of faeces	Loose	55	13	23.6	1.34	0.247
	Firmly-formed	185	31	16.8		
LGAs	Zaria	40	9	22.5	15.250	0.009
	Sabon Gari	40	11	27.5		
	Giwa	40	11	27.5		
	Igabi	40	3	7.5		
	Soba	40	1	2.5		
	Kudan	40	9	22.5		
Total		240	44	18.3		

Table 4. Prevalence of *Cryptosporidium* oocysts in sheep faeces in sedentary Fulani herds in selected Local Government Areas of Kaduna State, Nigeria

Variables	Category	Number Examined	Number positive	Specific rate (%)	χ2/Fishers exact	P
Age (months)	≤ 6	62	12	19.4	2.363	0.124
	> 6	118	13	11.0		
Sex	Male	64	10	15.6	0.250	0.617
	Female	116	15	12.9		
Breed	Yankasa	138	18	13.0	1.104	0.576
	Balami	39	7	17.9		
	Uda	3	0	0.0		
Body condition score	Poor	54	7	13.0	3.119	0.210
	Medium	47	10	21.3		
	Good	79	8	10.1		
Consistency of faeces	Loose	41	9	22.0	2.886	0.089
	Firmly-formed	139	16	11.5		
LGAs	Zaria	30	1	3.3	28.66	< 0.001
	Sabon Gari	30	12	40.0		
	Giwa	30	6	20.0		
	Igabi	30	0	0.0		
	Soba	30	1	3.3		
	Kudan	30	5	16.7		
Total		180	25	13.9		

Table 5. Prevalence of *Cryptosporidium* oocysts in goat faeces in sedentary Fulani herds in selected Local Government Areas of Kaduna State, Nigeria

Variables	Category	Number examined	Number positive	Specific rate (%)	χ2/Fishers exact	P
Age (months)	≤ 6	81	6	7.4	3.216	0.073
	> 6	159	4	2.5		
Sex	Male	100	2	2.0	2.015	0.156
	Female	140	8	5.7		
Breed	Kano brown	188	7	3.7	0.888	0.641
	Red Sokoto	47	3	6.4		
	West African dwarf	5	0	0.0		
Body condition score	Poor	67	7	10.4	9.746	0.008
	Medium	61	2	3.3		
	Good	112	1	0.9		
Consistency of faeces	Loose	58	4	6.9	1.43	0.232
	Firmly-formed	182	6	3.3		
LGAs	Zaria	40	3	7.5	4.591	0.468
	Sabon Gari	40	2	5.0		
	Giwa	40	3	7.5		
	Igabi	40	1	2.5		
	Soba	40	1	2.5		
	Kudan	40	0	0.0		
Total		240	10	4.2		

Table 6. Prevalence of *Cryptosporidium* oocysts in dog faeces in sedentary Fulani herds in selected Local Government Areas of Kaduna State, Nigeria

Variables	Category	Number examined	Number positive	Specific rate (%)	χ2/Fishers exact	P
Age (months)	≤ 6	29	8	27.6	4.714	0.03
	> 6	61	6	9.8		
Sex	Male	64	12	18.8	1.721	0.190
	Female	26	2	7.7		
Breed	Local	87	14	16.1	0.572	0.450
	Crossbreed	3	0	0.0		
Body condition score	Poor	30	7	23.3	2.185	0.335
	Medium	21	2	9.5		
	Good	39	5	12.8		
Consistency of faeces	Loose	19	5	26.3	2.123	0.145
	Firmly-formed	71	9	12.7		
LGAs	Zaria	15	1	6.7	6.767	0.239
	Sabon Gari	15	2	13.3		
	Giwa	15	4	26.7		
	Igabi	15	0	0.0		
	Soba	15	3	20.0		
	Kudan	15	4	26.7		
Total		90	14	15.6		

Table 7. Distribution of *Cryptosporidium* oocysts in bulk milk sampled from sedentary Fulani herds in selected Local Government Areas (LGAs) of Kaduna State, Nigeria

LGA	Number of herds	Number examined	Number positive (%)	χ 2/Fishers exact	P-value
Zaria	5	20	2 (10.0)	2.588	0.763
Sabon gari	5	20	1 (5.0)		
Giwa	5	20	4 (20.0)		
Igabi	5	20	3 (15.0)		
Soba	5	20	2 (10.0)		
Kudan	5	20	2 (10.0)		
Total	30	120	14 (11.7)		

Table 8. Prevalence of *Cryptosporidium* oocysts in water bodies around sedentary Fulani herds in selected Local Government Areas (LGAs) of Kaduna State, Nigeria

Factors		Number Examined	Number Positive	Specific rate (%)	χ 2/ Fisher's exact	P-value
LGAs	Zaria	20	0	0.0	23.077	< 0.001
	Sabon gari	20	4	20.0		
	Giwa	20	0	0.0		
	Igabi	20	0	0.0		
	Soba	20	4	20.0		
	Kudan	20	8	40.0		
Temperature (°C)	23.1–25.0	40	8	20.0	2.308	0.129
	25.1–27.0	80	8	10.0		
pH	6.0–6.9	56	16	28.6	21.099	< 0.001
	7.0–7.9	64	0	0.0		
Turbidity (NTU)	0.001–0.300	40	0	0.0	11.403	0.003
	0.301–0.600	68	12	17.6		
	0.601–0.900	12	4	33.3		
Dissolved Oxygen ($mg.l^{-1}$)	1.1–3.0	20	4	20.0	0.923	0.337
	3.1–6.0	120	12	12.0		
	Total	120	16	13.3		

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