

Original papers

Cryptosporidium oocysts: prevalence in dogs in Abuja, Federal Capital Territory, Nigeria

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ABSTRACT. *Cryptosporidium* is known to be a zoonotic protozoan parasite, located mainly intracellularly causing the disease called cryptosporidiosis, a diarrheic disease of human and animals. Risk factors for dogs' illness includes exposure to contaminated drinking water from well, borehole and tap water. Hunting as an activity of some dogs serve as a major exposure. The objectives of this study is to determine the prevalence and risk factors associated with fecal shedding of *Cryptosporidium* oocysts in dogs in Abuja, Federal Capital Territory (FCT). This study was carried out from January to May 2018 in 6 Area Councils of FCT. A total of 400 dogs (213 males and 187 females) aged 0–14 years old were enrolled for this study. The fecal samples collected were examined using Modified Ziehl-Neelsen (MZN) technique. Overall, 91 samples out of 400 were positive, giving a prevalence of *Cryptosporidium* oocysts infection as 22.75% with 5% degree of freedom. Prevalence among male and female dogs were 27% and 17%, respectively which is statistically significant ($p=0.034$). Younger puppies had a higher infection rate compared to the older dogs. The infection is relatively higher in local breed of dogs than the cross and pure breeds. This is significantly different ($p=0.014$). Source of water was also significantly associated with *Cryptosporidium* infection but food type was found not to be associated. Some of the dogs infected with *Cryptosporidium* were presented with diarrhoea, though asymptomatic ones can still shed oocysts in the environment. These act as a possible source of infection for other animals.

Keywords: *Cryptosporidium* species, dogs, prevalence, Abuja, Nigeria

Introduction

Dogs habitually possess intestinal parasites causing cross infection to humans known as zoonosis. There is minimal reporting of the zoonotic risk of *Cryptosporidium* infections from infected dogs, though the isolation of *Cryptosporidium canis* from both dogs and children from the same household has been reported in Peru, which utmostly highlights cross infection between dogs and children [1]. Cryptosporidiosis in dogs is the predominant zoonotic parasite responsible for diarrhoea and gastroenteritis clinically, caused by opportunistic protozoan of the genus *Cryptosporidium* [2]. However, identified *Cryptosporidium* as an intracellular zoonotic parasite leading to diarrhoea both in humans and domestic animals [3].

Infants, immune-suppressed and the aged are the most vulnerable, although zoonotic parasite have been reported to cause death and unhealthy state in all age groups of humans and animals [1,4]. Xiao and Feng [5] suggested that dogs can be a remarkable source of human cryptosporidiosis. It is now recognized that *Cryptosporidium* species differ particularly in their host range while some are narrowed to particular species or type of host. It is reported that *C. parvum* and *C. hominis* are commonly found in humans, which includes *C. canis* [6,7]. Infection is more common in younger dogs of less than 6 months of age as compared to adult dogs. In addition to the above, severe diarrheic stool, malabsorption and weight loss especially in younger puppies are the clinical signs though some are asymptomatic [8,9].

Table 1. Prevalence of *Cryptosporidium* in dogs from Area Council

Area Council	Number examined	Number positive	Prevalence (%)
Kwali	66	16	24.2
Amac	68	11	16.2
Bwari	66	13	19.7
Abaji	66	19	28.8
Gwagwalada	66	19	28.8
Kuje	68	13	19.1
Total	400	91	22.75

$$\chi^2 = 5.35; \quad P = 0.374; \quad df = 5$$

Dogs have been reported to be infected naturally with *C. canis*, *C. parvum* and *C. meleagridis* [10, 11]. In Abuja, studies on *Cryptosporidium* in animals are slender. The only previous report by Olabanji et al. [12] from Abuja did not include three other Area Councils (Kuje, Bwari, and Gwagwalada), which were included in this study. Though the crucial nature of zoonotic parasites in dogs is known in most parts of the country, current research on *Cryptosporidium* infection in dogs remains sparse in Abuja. This investigation therefore aims to determine the prevalence and the risk factors associated with *Cryptosporidium* oocysts shedding in dogs, spread of infection to humans and other animals.

Materials and Methods

Study area. A cross-sectional study was conducted in the six Area Councils of Abuja (Kwali, AMAC, Gwagwalada, Bwari, Abaji and Kuje), the Federal Capital Territory of Nigeria. Three wards in each Area Council were selected using random sampling method. In Kwali 66 fecal samples were gotten from three wards viz: (26 sampled from Area Council Secretariat, 20 Yangoji, 20 Gumbo). In Abaji, 66 fecal samples in the three

wards viz: (20 from Area Council Secretariat, 25 from Abaji South East and 21 from Yaba). In Kuje, 68 fecal samples viz: (30 from Area Council Secretariat, 15 Kabi, 23 St. Kizito). In Gwagwalada, 66 samples viz: (20 from Area Council Secretariat, 24 from phase 3, 22 from phase 4). In Bwari, 66 in the three wards viz: (16 sampled from Dutse Alhaji, 30 Kubwa, 20 from Zuma 1). While in AMAC, 68 samples (30 from Vet world Bannex plaza, 10 Osun Crescent, and 28 from Jabi). A total of 400 samples were collected from 18 wards in the six Area Councils. Dogs presented to both government and private clinics were sampled. House to house collection was also done in some areas where necessary.

Study design. The study was conducted between January–May, 2018. All the dogs that visited the selected Veterinary Clinics were sampled (except those presented with suspected parvovirus enteritis). Samples were also collected from home to home in cases where Veterinary Clinics were limited.

Sample collection. Samples were collected directly from the rectum by using disposable latex gloves and for puppies, the topmost parts of freshly voided feces were collected. The samples were transferred immediately into labelled sterile wide-mouthed plastic bottles according to the protocols of Jongwutiwes et al. [13] and transported in cold chain to the parasitology laboratory of the Department of Veterinary Parasitology and Entomology, Faculty of Veterinary Medicine, University of Abuja for processing. The sample size was estimated using formulae developed by Fox et al. [14]. Therefore, 384 dogs were calculated, but to increase precision and get more accurate, the sample size was taken to be 400. A structured questionnaire was used to capture information of dogs and their management factors such as the type of food, routine check-up, water source, and confinement or roaming among others.

Risk factors. The risk factors studied were the age in months, summed up into (0–24, 25–48, >48),

Table 2. Relationship between *Cryptosporidium* oocysts shedding and breed

Variable	No of dogs examined	No positive for <i>Cryptosporidium</i>	Percentage (%)
Exotic breeds (GS, TR, LH, CA, BB Etc.)	200 (50%)	37 (40.7%)	18.5
Local breeds	146 (36.5%)	45 (49.5%)	30.8
Cross breeds	54 (13.5%)	9 (9.9%)	16.7
Total	400	91	22.75

Explanations: GS – German Shephard; TR – Terrier; LH – Lhasa; CA – Caucasian; BB – Boer boel; $\chi^2 = 8.61$; $P = 0.014$; $df = 2$

Table 3. Relationship between *Cryptosporidium* oocysts shedding and sex

Sex	Number of examined	Number of positive	Percentage prevalence	Number of negative	Prevalence
Males	213 (53.3%)	58	27%	158 (51.1%)	74.2%
Females	187 (46.8%)	33	17%	151 (48.9%)	80%
Total	400	91	22.75%	309	77.3%

$$\chi^2 = 4.50; P = 0.034; df = 1$$

gender (males and females), living conditions (confined, roam, security, hunting, pet), breed (pure, local and cross), infection with other parasites.

The fecal samples were treated using formol-ether concentration method and stained using Modified Ziehl-Neelsen (MZN) [15]. Microscopically, the stained slides were examined. However, if at least one *Cryptosporidium* oocysts was seen and pinpointed as a pinkish-red oval oocyst against a blue background, the sample was considered positive.

Statistical analysis. The data gotten were entered into an excel spreadsheet and tables and charts were used to represent it (descriptive statistics). Chi-square was used to check for an association between *Cryptosporidium* and sex, age, and other factors studied. Values of $p \leq 0.05$ were considered statistically significant.

Results

Ninety-one (22.8%) out of the 400 fecal samples examined using MZN staining technique were positive for *Cryptosporidium* oocysts. This is considered to be relatively high as shown in Table 1.

Abaji and Gwagwalada Area Council had the highest prevalence of (28.8%) each, while the least prevalence was recorded in AMAC (16.2%). Kwali, Bwari and Kuje Area Council had prevalence rates of 16 (24.2%), 13 (19.7%) and 13 (19.7%), respectively. There was no significant difference ($p=0.37$) between the occurrence of *Cryptosporidium* oocysts and Area Councils examined.

The prevalence was higher among local breeds 45 (30.8%) than exotic 37 (18.5%) and cross breed 9

Table 4. Age related prevalence of *Cryptosporidium* oocysts in dogs

Age in months	No of examined	No of positive	Prevalence (%)
0 – 12	168	47	28
13 – 24	76	16	21.1
25 – 36	56	8	14.3
37 – 48	40	8	20
49 – 60	24	5	20.8
61– 72	16	3	18.8
72 >	20	4	20
Total	400	91	22.75

$$\chi^2 = 5.47; P = 0.485; df = 6$$

(16.7%) of dogs. It was observed that breeds had a significant effect on the prevalence of *Cryptosporidium* and are associated ($p=0.014$) in Table 2. Males had higher infection 58 (27%) than females 33 (17%). The prevalence of *Cryptosporidium* between males and females were statistically significant ($p=0.034$) as shown in Table 3. The highest prevalence was recorded in the age group 0–12 months with a prevalence of 28% while the least prevalence was recorded in dogs between the ages of 25–36 months as seen in Table 4. However, when dogs were categorized into 3 age groups <24, 25–48, >48 months, it was observed that the prevalence of *Cryptosporidium* infection was yet highest in younger dogs where 244 were sampled with 63 (25.8%) positives, followed by the older dogs with 12 positives (20%) out of 60 sampled.

Table 5. Prevalence according to diarrheic dogs

	Total number of examined	Number with diarrhoea	Number of positive	Prevalence (%)	Number without diarrhoea	Number positive without diarrhoea	Prevalence (%)
Male	213	78	35	44.9	135	23	17
Female	187	58	23	39.7	129	10	7.8
Total	400	136	58	42.6	264	33	12.5

$$\chi^2 = 46.4; P = 0.000; df = 1$$

Table 6. Other factors investigated for *Cryptosporidium* infection

Factor	Number of examined	Number of positive	Prevalence (%)	P-value
<i>Confined</i>				
Yes	116	17	14.7	
No	284	74	26.1	0.014
Within the compound	131	28	21.4	
Outside the compound	47	12	25.5	
Both outside and within	106	34	32.1	0.019
<i>Water Source</i>				
Borehole	133	26	19.5	
Tap	181	38	21.0	
Well	86	27	31.4	0.093
<i>Type of food given</i>				
Combination	135	30	22.2	
Dry	140	39	27.9	
Wet	125	22	17.6	0.136
<i>Disease awareness</i>				
Yes	15	1	6.7	
No	385	90	23.4	0.130

The lowest are the dogs between the ages of 25–48 months with 16 (16.7%) positives out of 96 sampled. There was no statistical significance ($p=0.166$) between the age group. In table 5, there was also a higher prevalence for dogs with diarrhoea, 58 (42.6%) than from well-formed feces 33 (12.5%), for roam 74 (26.1%) than confined 17 (14.7%).

Table 6 shows that there were no associations between sources of water: borehole 26 (19.5%), tap 38 (21.0%) and well 27 (31.4%), food given: combination 30 (22.2%), dry 39 (27.9%), wet 22 (17.6%). There were associations ($p \leq 0.05$) between prevalence of *Cryptosporidium* and breed ($\chi^2=8.61$, $p=0.014$), sex ($\chi^2=4.50$, $p=0.034$), diarrhoea ($\chi^2=46.4$, $p=0.000$), confinement ($p=0.014$), and activities of dogs ($\chi^2=9.88$, $p=0.020$). These are statistically significant.

Discussion

The 22.8% rate recorded in this study was higher than the previous report of 5.4% reported by Olabanji et al. [12] in Abuja and 5.9% in Lusaka district [16]. These variances could be allotted to higher sample size, which most were gotten from the rural areas. However, the prevalence in this study is lower than 44% reported in South Africa [3]. *Cryptosporidium* prevalence varied from one Area Council to another, though not appreciably associated. Two Area Councils (Abaji and Gwagwalada) recorded the relatively higher prevalence of 28.8% each compared to AMAC with

a prevalence of 16.2%. Abaji is regarded as a rural settlement where dogs freely roam and are not well cared for. In Gwagwalada, the extreme climatic condition of the area might be enhancing the prevalence of the infection. However, the relatively lower prevalence in dogs sampled from AMAC doubtlessly obtain satisfactory care, clean and or safe food, Veterinary Services and shelter than other Area Councils, whose majority of owners are within middle/lower income class and cannot purvey conventional Veterinary services.

Breeds were found to be significantly associated with the prevalence of *Cryptosporidium* infections with higher prevalence among local breeds than in pure and cross breeds. This discovery was harmonious with the study by Titilincu et al. [17] but in contrast to the results obtained by Adejinmi et al. [18] that reported a higher prevalence in mixed breeds. However, in Egypt from the reports of Awadallah et al. [19] there was no correlation between dog breed and *Cryptosporidium*.

Sex susceptibility to the prevalence of *Cryptosporidium* is higher in males than females. An identical result was reported by Getahun and Addis [20] with a prevalence of 79.2% in male than 76.8% in female dogs. The result is in dispute with those reported by Olabanji et al. [12] with a prevalence more in females than in males. Males are known to be free-wanderers by going from one place to another in search of female complement. This makes them more prone to infections.

Age of dogs has no significant effect on the prevalence of *Cryptosporidium* infection in the

dogs. The highest prevalence was recorded among younger dogs of 0–12 months compared to older age group. Bajer et al. [21] accords this reports with higher prevalence in young animals. Unlike two reports proven by Olabanji et al. [12] and Mugala et al. [16] who found infection more in older dogs. However, host ability to withstand infections is usually acquired with age, probably due to single or repeated vulnerability [9]. Higher infection rates in older host can be attributed to degrees of infection with age.

Diarrheic dogs in this study had a strong association with *Cryptosporidium* shedding. This agrees with previous reports by Tariuwa et al. [22], who stipulated a higher prevalence of *Cryptosporidium* in dogs with diarrhoea than non-diarrheic dogs. However, Ramirez-Barrios et al. [9] disagreed with this study and they suggested that most infections in dogs are symptomless. Hence, dogs can harbour the parasites without symptoms but can still shed oocysts. Diarrhoea observed in cryptosporidiosis may be associated with the pathogenesis of the parasite and concurrent infection with other enteric pathogens such as *Ancylostoma*, *Isospora* etc.

Dogs that were roaming both within and outside the compound has a higher prevalence of *Cryptosporidium* infection of 26.1% compared to those that were completely confined (14.7%) with a significant difference. This strongly agrees with the report of Rambozzi et al. [23], who reported that outdoor cats and dogs were approximately five times more likely to be infected with *Cryptosporidium* specie than indoor ones. This can also be attributed to the fact that unconfined/non-sheltered dogs roam about in the streets where they may hunt, forage and eat adulterated food materials, thereby increasing the risk of getting infected with *Cryptosporidium*. Those that are confined and are positive maybe due to drinking fecally contaminated water. Also, contaminated feces can get on the dog's fur; when grooming himself, ingests the parasite. Water source given to the dogs was also investigated as a risk factor in this research. There is a borderline significant trend (0.093%) between water source and prevalence of *Cryptosporidium*. This leads to the understanding that all water sources are a potential risk. Well water has a higher prevalence rate of 31.4% compared to borehole and tap water with prevalence rates of 19.5% and 21.0%, respectively. Lisle and Rose [24] reported that between 5.6% and 87.1% of water source (i.e.

surface, spring, and groundwater samples not impacted by domestic and/or agricultural waste) tested contained 0.003 to 4.74 *Cryptosporidium* oocysts/L, while Korich et al. [25] reported *Cryptosporidium* to be resistant to chlorine treatment which is used commonly to treat tap water. Hence tap water undergoes treatment to some extent, it explains the lower prevalence seen in tap water in this study. *Cryptosporidium* may be more common in well water than borehole water because well waters are more vulnerable to direct infiltration and contamination from sewage discharges and runoff.

The current study showed that the type of food investigated had no significant influence on the prevalence of *Cryptosporidium* infection. However, the works of Abere et al. [26] disagrees with this finding and he however reported that feeding management had a significant influence on the prevalence of the gastrointestinal helminth infections but Quinn's report [27] highlighted the role of *Cryptosporidium* as a foodborne pathogen in uncooked meat products and uncooked or unwashed green onions. From this study, we found all food types (dry, wet and combination) as a potential risk to *Cryptosporidium* infections due to poor handling and poor sanitary measures by dog owners.

There is an association between *Cryptosporidium* and working activities of dogs. Dogs have been used as security, pet and hunting purposes. Hunting from this study had a high prevalence of 38.3% for *Cryptosporidium* and on exposure to other parasitic (*Ancylostoma*, *Isospora*, *Toxocara*, *Trichuris*, *Paragonimus*) infection 55.3% prevalence. This is extremely high compared to security with 18.6% prevalence for *Cryptosporidium* and 23.7% prevalence for other parasitic infections. This result is higher than Mustapha et al. [28] who reported an overall prevalence of 38% in hunting dogs but lower than Johnson et al. [29] with 86.4% prevalence. Generally, oocysts are resistant to a variety of environmental factors such as temperature. Closeness to other animals and other environmental contaminants such as watersheds, food crops and recreational waters serves as a source of infection to pets. This resistance enables oocysts to survive outside the host for extended periods of time, thus increasing the chances for the organisms to encounter new hosts.

The present study has shown that *Cryptosporidium* infections are present among dogs in Abuja, FCT. This finding is of public health importance as infected dogs can serve as a means of zoonotic

infection to humans. Infections are more in diarrheic dogs, and that local breeds are commonly infected compared to pure breeds of dogs. Hunting exposes dogs to a high level of infection both to *Cryptosporidium* and other parasitic infections. Awareness creation to educate dog owners on dog safekeeping practices is advocated in order to reduce the intestinal parasitic infections and cross-infections between dogs. Further studies are required on molecular identification, seasonality patterns of infections and development of more efficacious treatment for alleviating cryptosporidiosis in the system.

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