CRYPTOSPORIDIUM INFECTION IN ANIMALS AND HUMANS IN TANZANIA: A REVIEW

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ABSTRACT

Cryptosporidium is a zoonotic coccidian parasite causing infections in animals and humans. Two main species namely C. parvum and C. muris cause infections in mammalian hosts. C. parvum causes diarrhoea in both immunocompetemt and immunocompromised individuals with severe signs in the later. In Tanzania the parasite was first reported in 1984, in a diarrhoeic goat kid and subsequently in cattle, goats, sheep, pigs, dogs, wildlife and humans. Furthermore, Cryptosporidium has been associated with diarrhoea in cattle and pigs among domestic animals, as well as humans including HIV/AIDS patients with chronic diarrhoea. Two sizes of oocysts measuring 4-6mm and 6-7.4mm in diameter, resembling those of C. parvum and C. muris respectively, have been demonstrated in cattle. Cryptosporidium infections seem to be more prevalent in the cool climate areas of Tanzania. In various studies in Tanzania, neither details determining the association between human and animal cryptosporidiosis, nor the sources of infections to human and animals, were determined. The present paper highlights on the occurrence of Cryptosporidium in various domestic animals, wildlife animals and humans in Tanzania.

Keywords: *Cryptosporidium*, domestic animals, and wildlife, HIV/AIDS.

INTRODUCTION

Cryptosporidium is an obligate intracellular parasite causing infection in a wide range of animal species including man (Fayer and Ungar, 1986). The main *Cryptosporidium* spp. infecting mammalian hosts include *C. parvum*, with oocysts measuring 4.0 to 6µm in diameter and *C. muris* which measures 6.0 to 7.4µm in diameter (Fayer and Ungar, 1986). *C. parvum* is the major species known to cause clinical illnesses in human and other mammalian hosts (Current, 1988). The importance of *Cryptosporidium* was recognized after establishing its association with diarrhoea in cattle and humans (Panciera *et. al.*, 1971; Meisel *et. al.*, 1976; Nime *et. al.*, 1976). At present, *C. parvum* is recognized as an important zoonotic parasite occurring in a wide range of animal species and humans (Angus, 1983; Tzipori, 1983; Current, 1989, K-State, 2000).

The main clinical features of *C. parvum* infection is diarrhoea associated with profuse shedding of infective oocysts over 10^6 oocysts per gram of faeces (Blewett, 1988). The virulence of the strain, infective dose, susceptibility and the age of the host determine duration of diarrhoea (K-State, 2000). In immunocompromised patients, *C. parvum* causes a prolonged life-threatening cholera-like diarrhoea whereas, in immunocompetent individuals the disease is self-limiting (Fayer and Ungar, 1986; Current, 1989). With *C. muris* infections, the disease is characterized by inflammation of the stomach or abomasum but the affected

animals never develop diarrhoea. Infected animals may shed *C. muris* oocysts for several months and lose body weight (Fayer and Ungar, 1986).

Cryptosporidiosis has been reported in a number of countries world wide in a variety of animal species including bovine, goats, sheep, pigs, dogs, non-human primates, some wild herbivores and humans in both diarrhoeic and non diarrhoeic cases (Fayer and Ungar, 1986; K-State, 2000). Such reports include cryptosporidiosis in goats (Mason *et. al.,* 1981; Matovelo *et. al*,, 1984), in sheep (Barker *et. al.,* 1974; Anderson, 1982), in pigs (Tzipori *et. al.,* 1981; Moon and Bemrick., 1981) and in puppies (Wilson and Holscher, 1983). Other reports are in non-human primates (Kovatch and White, 1972; Cockrell *et. al.,* 1974), in wild herbivores (Angus, 1990; Mtambo *et. al.,* 1997) and in human beings (Fayer and Ungar, 1986). Limited studies have been conducted on cryptosporidiosis in African countries. Human cryptosporidiosis has been reported in Uganda, Zambia, Guinea Bissau, Rwanda and Tanzania. (Casemore *et. al.,* 1985; Sewankambo *et. al.,* 1987; Mølbak *et. al.,* 1990; Conlon *et. al.,* 1990; Tarimo, 1995).

In Tanzania, the parasite was first reported in diarrhoeic goat kids (Matovelo *et. al.,* 1984). Diarrhoea has been reported as the major cause calf mortalities in Tanzania, however, the main aetiological agents could not be determined (Shoo *et. al.,* 1992). In HIV/AIDS patients, chronic diarrhoea occur in 40% of cases being one of the major complaints (WHO, 1992). Although several reports indicate that *Cryptosporidium* infection is prevalent among animals and humans, the condition does not receive the required attention by both the medical and animal health personnel. The following is a review of cases of *Cryptosporidium* spp. demonstrated in various studies in Tanzania in order to indicate the magnitude of the problem in the country.

Morphology of *Cryptosporidium* oocysts

Cryptosporidium has a direct life cycle involving endogenous and exogenous stages in all hosts infected (Fayer and Ungar, 1986). Diagnosis of *Cryptosporidium* infection is based on detection of the exogenous stage or the oocyst in faecal specimen of infected animals or humans using staining techniques. Two types of oocysts have been reported in various studies including small oocysts associated with *C. parvum* and the larger *C. muris* oocysts (Mtambo *et. al.,* 1992; Scott, *et. al.,* 1995).

In Tanzania, Mpelumbe-Ngeleja (1997) demonstrated two types of oocysts in cattle faecal specimens, the small type, measuring 4.0 and 5.9 µm in diameter and the larger type, measuring between 6.8 and 7.4 µm. The small oocysts were similar to those of *Cryptosporidium parvum* whereas larger oocysts resembled those of *Cryptosporidium muris*. Based on oocysts sizes, both *C. parvum* and *C. muris* isolates appear to be prevalent in Morogoro, Tanzania (Mpelumbe-Ngeleja, 1997). Co-existence of *C. parvum* and *C. muris* has been reported in several studies in cattle (Anderson, 1988; Lorenzo *et. al.,* 1993).

Confirmation of the diagnosis of *Cryptosporidium* is made by using specific anti-*Cryptosporidium* monoclonal antibody technique using Enzyme labeled immunoassays (ELISA) or fluorescence antibody techniques (FAT). *Cryptosporidium* oocysts, appear as round to ovoid bodies with an apple -green fluorescence against a dark background. However, in a recent study using FAT utilizing specific anti-*Cryptosporidium* monoclonal antibody (Crypt-a-Glo Waterborne Inc., Los Angles, USA), only the smaller *C. parvum*-like oocysts from human stool specimens were positive whereas the larger oocysts resembling *C. muris* did not show any

fluorescence (Mpelumbe-Ngeleja, 1997). This suggests that the monoclonal antibody used in the study was specific for *C. parvum* isolates.

Cryptosporidium isolates have been reported in a variety of animal species in Tanzania, but limited studies have been carried out to characterize the organism (Kambarage *et. al.,* 1995; Tarimo, 1995; Esrony *et. al.,* 1996; Makene *et. al.,* 1996; Mtambo *et. al.,* 1997; Mpelumbe-Ngeleja, 1997; Mtambo, unpublished). Several *Cryptosporidium* spp. isolates with varying degree of virulence have been reported elsewhere (McDonald and Awad-El-Karim, 1995; Leng *et. al.,* 1996).

Epidemiology of *Cryptosporidium* in Tanzania

Reports of the occurrence of *Cryptosporidium* in Tanzania also involved the African buffalo, zebra and wildebeest, the species hardly associated with cryptosporidiosis in other reports (Mtambo *et. al.,* 1997). Table 1 shows the details in number of samples examined, number found with *Cryptosporidium* oocysts in different animals and study areas in Tanzania. Table 2 shows the overall prevalence of *Cryptosporidium* in different animal species in various studies in Tanzania.

Table 1: *Cryptosporidium* spp. oocysts in faecal samples/stool specimens examined from various studies in Tanzania.

	Domestic Animals					Wild animals		Human
Study Area	Cattle	Goats	Sheep	Pigs	Dogs	Herbivores	NHP*	
Morogoro [1]	242 (0)	367 (3)	121 (0)	-	-	-	-	-
Moshi [2]	-	-	-	-	-	-	-	90 (36)
Morogoro [3]	-	-	-	424 (33)	-	-	-	-
Morogoro [4]	-	-	-	-	235 (1)	-	-	-
Morogoro [5]	486 (26)	-	-	-		87 (22)	-	-
Morogoro [6]	1195 (280)	-	-	-	-	-	-	278 (7)
Kitulo [7]	284 (96)	-	-	-	-	-	-	-
NCA [7]	71 (33)	-	-	-	-	-	-	-
Gombe [7]	-	-	-	-	-	-	136 (2)	-
Total	2278 (435)	367 (3)	121 (0)	424 (33)	235 (1)	87 (22)	136 (2)	368 (43)

Sources of data are; [1] Kambarage *et. al.,* 1995; [2] Tarimo, 1995; [3] Esrony *et. al.,* 1996; [4] Makene *et. al.,* 1996; [5]Mtambo *et. al.,* 1997; [6] Mpelumbe-Ngeleja, 1997; [7]Mtambo (unpublished).
NHP = Non human primates; NCA = Ngorongoro conservation area.
( ) = data in parenthesis indicate *Cryptosporidium* positive specimens.

Table 2: *Cryptosporidium* oocysts isolation in faecal samples from different species (collective data for all the studies in the review

Species	Number examined	Positive samples	% positive
Cattle	2278	435	19.1
Goats	367	3	0.8
Sheep	121	0	0.0
Pigs	424	33	7.8
Dogs	235	1	0.4
Zebra *(Equus zebra)*	25	8	28.0
Wildebeest (*Connechaetes gnou*)	26	7	27.0
Buffalo (*Syncerus caffer*)	36	7	22.0
Non human primates	136	2	1.8
Human	368	43	11.7

Cryptosporidiosis in cattle

The prevalence of *Cryptosporidium* oocysts in cattle faeces as reported by various authors in Tanzania included 0% (Kambarage *et. al.,* 1995), 5.3 % (Mtambo *et. al.,*, 1997) and 23.4% (Mpelumbe-Ngeleja, 1997) in warm eastern zone of the country. Furthermore, *Cryptosporidium* oocysts were demonstrated in 34% and 46% in the cool areas in the Southern highland zone namely Kitulo and NCA in Northern highland zone respectively (Mtambo unpublished). Nevertheless, the prevalence of *Cryptosporidium* in cattle is much lower compared to that reported in the temperate countries with up to 94% of dairy and beef cattle shedding oocysts (Scott *et. al.*, 1995; Kemp *et. al.*, 1995).

On average, the oocysts per gram (OPG) detected in infected cattle ranged from 6.2×10^3 to 7.6×10^6 (Mpelumbe-Ngeleja, 1997). Of the 280 bovine faecal samples found positive for cryptosporidiosis in that particular study, the OPG range of 6.2×10^3 to 3.8×10^4 was in 58.2% (163 cattle) while 5.2×10^4 to 1.02×10^5 was detected in 28.6% (80 cattle). Other OPG ranges were 1.03×10^5 to 2.0×10^6 and 2.1×10^6 to 7.6×10^6 in 11.4% (32 cattle) and 1.8% (5 cattle) of the faecal samples examined, respectively. (Mpelumbe-Ngeleja, 1997). According to Kemp *et. al.*, (1995), adult cattle produce about 30 to 40 kgs of faeces per day. In that study, 155 adult cattle were detected to excrete *Cryptosporidium* oocysts in faeces in amounts ranging between 6.2×10^3 and 7.6×10^6 OPG. This suggests that the total number of oocysts shed in the environment per day could range between 1.86×10^8 and 2.28×10^{11} per animal producing about 30 kgs of faeces. This shows that *Cryptosporidium* infected cattle contribute significantly to environmental contamination with the oocysts. Shedding of *Cryptosporidium* oocysts by subclinical or apparently normal animals poses a great risk to both animals and human.

Bovine Cryptosporidiosis and diarrhoea

In their report, Mtambo *et. al.*, (1997) demonstrated *Cryptosporidium* oocysts in 12 (57%) out of 21 diarrhoeic cattle. According to their report, occurrence of oocysts in diarrhoeic cattle was significantly high. Furthermore, it was observed that, out of 12 diarrhoeic cattle shedding *Cryptosporidium* oocysts, nine were below three months old. In another study conducted in the same area, *Cryptosporidium* oocysts were demonstrated in 25.9% (n = 58) of

the diarrhoeic and 23.3% (n = 265) of the non-diarrhoeic faecal samples (Mpelumbe-Ngeleja, 1997).

Various studies have associated *Cryptosporidium* infection with diarrhoea especially in calves where the parasite was observed in 16% to 68% of diarrhoeic calves (Tzipori *et. al.,* 1980; Fayer and Ungar, 1986). Clinical cases of cryptosporidiosis associated with diarrhoea have been shown to produce between 10^6 and 10^8 OPG faeces (Blewett, 1988). In the study by Mpelumbe-Ngeleja (1997), average OPG was $2.3x10^6$ in one calf with severe watery diarrhoea, $1.5x10^5$ in two adults with moderate diarrhoea. Based on oocyst count in the three diarrhoeic cattle excreting between 1.03×10^5 and 7.6×10^6 OPG of faeces, diarrhoea could strongly be associated with *Cryptosporidium* spp. infection. Since other agents likely to cause diarrhoea in cattle were not determined, it is difficult to incriminate *Cryptosporidium* as the sole cause of diarrhoea in cattle in Tanzania.

Cryptosporidiosis in small ruminants

Based on the available reports, the prevalence of the parasite in small ruminants is relatively low. Kambarage *et. al.,* (1995); reported prevalence rate of 0.8% in goats and 0% in sheep. However, limited studies have been conducted to establish the occurrence of *Cryptosporidium* in small ruminants in various parts of Tanzania. Elsewhere, *Cryptosporidium* has been associated with diarrhoea in goat kids and lambs (Barker *et. al.,* 1974; Mason *et. al.,* 1981; Anderson, 1982; Matovelo *et. al.,* 1984). The age and health status of the three goats found positive for *Cryptosporidium* in a previous study conducted in Tanzania (Kambarage *et. al.,* 1995); was not reported. Further studies need to be conducted so as to establish the actual status of cryptosporidiosis in small ruminants in Tanzania.

Cryptosporidiosis and its association with diarrhoea in pigs in Tanzania

In pigs, *Cryptosporidium* was more prevalent in cool climate area of Mgeta where it was demonstrated in 25% of the samples whereas in the warm areas of Morogoro municipality the prevalence of 0.8%, was reported (Esrony *et. al*., 1996). No other reports are available on *Cryptosporidium* in pigs in other areas of Tanzania. Studies elsewhere demonstrated *Cryptosporidium* oocysts in 5.3% (n=3491) in Canada, 5% (n=200) in California, and 19.6% in Trinidad and Tobago (Moon and Bemrick, 1981; Tacal *et. al*., 1987; Kaminjolo *et. al.,* 1993). In those studies, 30-50% of the diarrhoeic pigs excreted *Cryptosporidium* oocysts (Moon and Bemrick, 1981; Tacal *et. al*., 1987; Kaminjolo *et. al.,* 1993; Esrony, unpublished).

Cryptosporidiosis in dogs

Occurrence of *Cryptosporidium* oocysts in 0.4% of dogs (n=235) shows the rare occurrence of the parasite in this species (Makene *et. al.,* 1996). Similar observations were made in Finland and Germany whereby of the 57 and 200 dogs surveyed, none was found with the parasite (Pohjola 1984; Augustin *et. al.,* 1984). However, in North America, four clinically ill puppies aged between one week and three months were found positive for *Cryptosporidium* infection (Wilson and Holscher, 1983). However, dogs surveyed in the study in Tanzania were those brought to the Sokoine University of Agriculture veterinary clinic for some other problems (Makene *et. al.,* 1996).

Cryptosporidiosis in wild herbivores

Cryptosporidium oocysts were detected in 25% of the wild herbivores faecal samples from Mikumi National Park Tanzania (Mtambo *et. al.*, 1997). There is little known on *Cryptosporidium* oocysts shedding in other African wildlife species for comparison purposes.

Cryptosporidiosis in non human primates

Mtambo (unpublished), demonstrated *Cryptosporidium* oocysts in 1.4% (n=1360 of baboons and chimpanzee from Gombe in Tanzania. Occurrence of the parasite in non-human primates has been reported elsewhere by Kovatch and White, (1972), Cockrell *et. al.*, (1974) and Wilson *et. al.*, (1984) Most of the reports from the temperate countries have been in infants or juveniles housed in primate centers and all were associated with diarrhoea (Kovatch *et. al.*, 1972; Cockrell *et. al.*, 1974; Wilson *et. al.*, 1983). In the study in Tanzania, the non-human primates were found in the wild and it is not known whether there was a history of diarrhoea in the individuals found with *Cryptosporidium* oocysts.

Cryptosporidiosis in humans

Mpelumbe-Ngeleja (1997) has shown the occurrence of *Cryptosporidium* spp. oocysts in 2.5% of humans in Morogoro district in Tanzania. In that study, three sources of stool specimens included healthy primary school pupils aged between 7 to 18 years old and patients with gastrointestinal problems from a dispensary and a regional hospital. Out of 278 stool specimens screened, only 65 were from the diarrhoeic patients. There was a relatively higher number (4.6%) of diarrhoeic patients who excreted *Cryptosporidium* oocysts compared to 1.9% of the patients who did not show signs of diarrhoea. This strongly suggests that *Cryptosporidium* is more common in diarrhoeic individuals than the non-diarrhoeic ones.

Cryptosporidiosis in adult HIV patients with chronic diarrhoea

The most common parasite isolated from African AIDS patients with chronic diarrhoea are *Cryptosporidium* spp. and *Isospora belli* (Fleming, 1990). Both or one of these opportunistic parasites have been found in 61%, of HIV patients with chronic diarrhoea in Uganda and Haiti (Sewankambo *et. al.*, 1987; DeHovirtz *et. al.*, 1986) whereas in Zambia, *Cryptosporidium* was found in 32% of such patients. *Cryptosporidium* has been found to be the most common parasite followed by *Isospora belli* which were found in 40% and 12%, respectively of HIV/AIDS patients with chronic diarrhoea in Tanzania (Tarimo, 1995).

Chronic diarrhoea is one of the major complains of AIDS patients occurring in 40% of cases (World health organization 1992). In Tanzania, Tarimo (1995), demonstrated that about 75% of adult HIV patients, of which, 45% (n=352) succumb to chronic diarrhoea cases whereas 29.8% (n=352) had history of diarrhoea in the past three months. Similar observations were made by Howlett *et. al.*, (1989) who reported chronic diarrhoea in 75% of AIDS patients. However in Mwanza region in Tanzania, chronic diarrhoea was not found to be common among HIV positive workers (Temu *et. al.*, 1994).

The findings by Tarimo (1995) are in agreement with similar studies involving HIV patients with chronic diarrhoea conducted in other African countries including Uganda (Sewankambo *et. al.*, 1987), Democratic Republic of Congo (former Zaire) (Colebunders *et. al.*, 1987), Rwanda (Soave and Johnson, 1988) and Zambia (Conlon *et. al.*, 1990).

In Tanzania, the prevalence of HIV/AIDS is increasing in many parts of the country (Mhalu *et. al.,* 1987; Kilewo *et. al.,* 1990; Shao *et. al.,* 1994; Ministry of Health, 1994). Nevertheless, the sources of infection of *Cryptosporidium* oocysts for humans involved in the NorthEastern Tanzania and Morogoro were not determined. However, the *Cryptosporidium* oocysts isolated from both human and bovine faecal specimens measured 4.0 to 5.9 µm in diameter, similar to those of *C. parvum* (Mpelumbe-Ngeleja, 1997).

Occurrence of *Cryptosporidium* in relation to climatical features in Tanzania

The highest prevalence of *Cryptosporidium* infection reported in the studies conducted in the warm climatic zone of Tanzania was 25% in wild herbivores (Mtambo *et. al.,* 1997) and 23.4% in cattle (Mpelumbe-Ngeleja, 1997). Other studies in the same area reported prevalence of less than 10% (Kambarage *et. al.,* 1995; Esrony *et. al.,* 1996; Makene *et. al.,* 1996; Mtambo *et. al.,* 1997; Mpelumbe-Ngeleja, 1997) in domestic animals and humans. On the other hand, studies conducted in cool tropical highland areas involving pigs in Mgeta and bovine in Kitulo and Ngorongoro showed prevalence of more than 25% (Esrony *et. al.,* 1996; Mtambo unpublished). Results from the study conducted in different species in Tanzania are presented in Table 3. The climatic factors affecting the survival or viability of the oocysts in the environment (Campbell *et. al.,* 1992; Robertson *et. al.,* 1992) could account for the differences recorded.

Table 3: Studies showing *Cryptosporidium* oocysts excretion in different species in warm and cool climate areas of Tanzania

Species and study area	Cool climate			Warm climate		
	No. examined	No. positive	% positive	No. examined	No. positive	% positive
Cattle Morogoro. Municipality.						
[1]	-	-	-	242	0	0
[2]	-	-	-	486	26	5.3
[3]	-	-	-	1195	280	23.4
Goats Morogoro municipality[1]	-	-	-	367	3	0.7
Sheep Morogoro municipality[1]	-	-	-	121	0	0
*Pigs Morogoro municipality[4]	-	-	-	296	1	0.7
Dogs Morogoro municipality[5]	-	-	-	235	1	0.4
Human Morogoro Municipality[3]	-	-	-	278	7	2.5
Wild Herbivores Mikumi[2]	-	-	-	87	22	25
*Pigs - Mgeta[4]	128	32	25	-	-	-
Cattle - Kitulo[6]	284	96	34	-	-	-
Cattle - NCA[6]	71	33	46	-	-	-

*One study; Sources are;[1] Kambarage *et. al.*,1995; [2]Mtambo *et. al.,* 1997; [3]Mpelumbe-Ngeleja, 1997; [4]Esrony *et. al.,* 1996; [5]Makene *et. al.,* 1996; [6]Mtambo(unpublished).

DISCUSSION

The studies on *Cryptosporidium* conducted in Tanzania so far demonstrate the existence of this important zoonotic pathogen in the country. It has been shown that *Cryptosporidium* is common in cattle where huge numbers of oocysts that are excreted contaminating the environment. This is of great concern as effluents from farming activities here been associated with outbreaks of diarrhoea due to Cryptosporidiosis in the developed countries

(Smith *et. al*., 1988; Barbara *et. al*., 1990). *Cryptosporidium* has also been found more in the diarrhoeic calves compared to the adults. This findings support observations reported in other countries (Tzipori *et. al.,* 1980; Sanford and Josephson, 1982). The finding of two types of oocysts, namely the small resembling *C. parvum* and the larger *C. muris* like oocysts also do suggest the occurrence of various *Cryptosporidium* isolates obtained in the country in relation to the clinical signs.

It has been observed in the studies on *Cryptosporidium* conducted in Tanzania that higher prevalence of the parasite could be influenced by the cool climatical condition. As observed in the studies on cattle and pigs, higher number of cases of *Cryptosporidium* infection were reported in the cool climatical areas as opposed to warm climate. This could be attributed to the survival of *Cryptosporidium* oocysts in the environment whereby oocysts lose viability reading in warm environment compared to moderately cool climatical condition (Campbell *et. al.,* 1992; Robertson *et. al.,* 1992). This could also explain the higher prevalence and oocysts burden in animals in the temperate climate (Scott *et al* 1995).

Reports of *Cryptosporidium* in wild herbivores is of great interest and suggests that the parasite is widely spread in wildlife. However, limited studies have been conducted on *Cryptosporidium* in other wildlife species. As observed, *Cryptosporidium* has also been demonstrated in non-human primates in Gombe National Park, Tanzania. The ecosystem of Gombe allows close contact between fishermen and the non-human primates to the extent of sharing feeds and some habitats. Such a situation allows easy transfer of pathogens between humans and the non-human primates. Nevertheless, non-human stool specimen has been screened for *Cryptosporidium* oocysts. Further studies are therefore necessary to demonstrate the source of *Cryptosporidium* infection to the non-human primates. Molecular biology techniques are also required in order to characterize the *Cryptosporidium* isolates in the non-human primates and other animal species.

CONCLUSION

Cryptosporidium has been strongly associated with diarrhoea in AIDS patients. This calls for a serious attention in determining the sources of human infections in order to control the spread of the parasite. The present review of studies on *Cryptosporidium* infection in Tanzania has highlighted on.

Further studies using a battery of anti-*Cryptosporidium* monoclonal antibody against both *Cryptosporidium* isolates are required in order to characterize the *Cryptosporidium* species present in domestic, wild animals and humans in Tanzania.

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