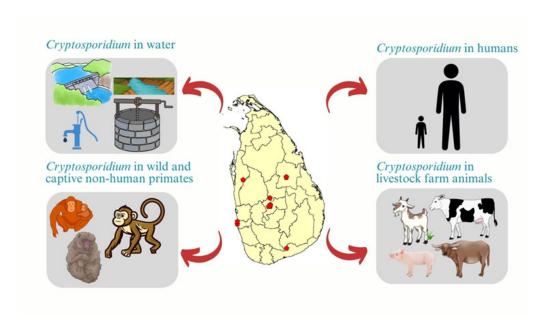
# **REVIEW ARTICLE**

# Cryptosporidium infections in Sri Lanka: A Systematic Review

Fazla Fareed, P.T. Amandi Thilakarathna, S.H.P. Parakrama Karunaratne and Faseeha Noordeen\*



# Highlights

- Cryptosporidium, a protozoan parasite causing gastroenteritis in humans and animals, is widespread in Sri Lanka.
- Livestock being a primary source of infection, goats, cattle, and buffaloes are the potential source for zoonotic transmission to humans.
- *Cryptosporidium* infections are a prevalent and significant cause of diarrhoea in children under the age of five years.
- High prevalence of *Cryptosporidium* oocysts in surface waters contaminated with faecal matter from domestic, agricultural, and wildlife animals.

### REVIEW ARTICLE

# Cryptosporidium infections in Sri Lanka: A Systematic Review

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Abstract: Cryptosporidium is a protozoan parasite that causes gastroenteritis in both humans and animals. Cryptosporidium infections in humans and animals have been reported from many parts of Sri Lanka. During the present study, five international electronic databases were extensively searched for peer-reviewed research papers published on the prevalence of Cryptosporidium infections and their occurrence in probable sources in Sri Lanka, within the time frame from 1987 to 2023. Collected information revealed that Cryptosporidium oocysts are more commonly found in surface water compared to well water, and shallow wells have a higher occurrence than deep wells. Contamination of river water with Cryptosporidium is mainly from the faecal matter coming from various domestic, agricultural and wildlife animal species. In Sri Lanka, Cryptosporidium infections in captive non-human primates is possibly due to contaminated food and human interaction. In wild animals, infections are more common in species that feed on the ground, suggesting contaminated soil and water as the source of infection. Most of the infected animals are asymptomatic, and many had co-infections with other enteric parasites. Molecular analysis of Cryptosporidium samples from infected primates revealed the presence of four major clades of C. parvum, with some isolates closely related to zoonotic C. parvum genotypes. Information on the habitats of the infected primates suggests that livestock is the primary source of infection. Several studies have been conducted to investigate the prevalence of Cryptosporidium infection in various livestock animals, particularly in goats, cattle, buffaloes, and swine in Sri Lanka. These studies reveal high prevalence of asymptomatic Cryptosporidium infection in goats and the potential for the transmission of zoonotic C. parvum from goats, cattle and buffaloes to humans. Molecular epidemiological analysis identified new genotypes of Cryptosporidium in domestic bovids (cattle and water buffalo), with no evidence of the commonly reported zoonotic species C. parvum. Waterborne transmission is the most common mode of infection of Cryptosporidium that affect both humans and animals. Studies conducted in human populations are primarily based in hospitals and pre-schools, and conclude that Cryptosporidium infections are a common cause of diarrhoea in children under the age of five. In conclusion, Cryptosporidium infections occur in different species of animals and humans in Sri Lanka and the oocysts have been detected in surface water, which might be an important source of infection for animals and humans in the country.

Keywords: Cryptosporidium; cryptosporidiosis; livestock; wildlife; water pollution; Sri Lanka

### INTRODUCTION

Cryptosporidium is a parasitic protozoan that causes gastroenteritis in humans and animals. The five primary species of Cryptosporidium known to cause infections are C. hominis, C. parvum, C. meleagridis, C. felis and C. canis. Among these species, C. hominis and C. parvum are the most frequently encountered species causing infections in humans (Sardar et al., 2021; Sharma et al., 2023).

Cryptosporidium, first discovered by Tyzzer in 1907, was initially considered non-pathogenic (Tyzzer, 1907). However, in 1976, identification of two human cases of cryptosporidiosis with diarrhoea showed its potential as an opportunistic pathogen. It was in 1982 that Cryptosporidium was recognized as a cause of self-limiting diarrhoea in healthy populations (Navin and Juranek, 1984). Its pathogenic role was then acknowledged as more severe and persistent in immuno-suppressed populations such as those with HIV/AIDS or undergoing chemotherapy. It was also reported that children, especially those under the age of five, are more susceptible to Cryptosporidium infection (Gerace et al., 2019). In 2015, cryptosporidiosis was ranked as the fourth leading cause of death in children under the age of 5, resulting in 1.3 million global fatalities. This accounted for approximately 12.1% of all deaths in children under the age of five worldwide as children are more vulnerable to infections due to the lack of exposure and resultant immunity (Troeger et al., 2017; Hassan et al., 2021).

Cryptosporidium has a broad host range which includes mammals, birds, reptiles, amphibians and fish (Fayer, 2004; Santín, 2013). Cryptosporidia are host-specific and different species are known to infect different host species. Symptomatic infections were first observed in turkeys in 1955 and Cryptosporidium associated neonatal diarrhoea in ruminants has been reported in the 1970s and 1980s (Panciera et al., 1971; Mason et al., 1981; Angus et al., 1982). Currently, cryptosporidiosis is a significant cause of diarrhoea in farm animals contributing to economic losses. Clinical signs in animals range from mild to severe diarrhoea leading to possible fatal outcomes (de Graaf et

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al., 1999; Noordeen et al., 2000; Noordeen et al., 2001; Noordeen et al., 2012).

Cryptosporidium infection can be transmitted to humans through direct contact from infected individuals or animals and indirectly through the consumption of contaminated food or water. Person-to-person and waterborne transmission are the most common modes of spread reported for outbreaks (Donnelly and Stentiford, 1997; Carmena, 2010). Cryptosporidium can enter water sources through faecal contaminations from infected humans or animals, particularly from livestock such as cows, goats and sheep (Fayer, 2004). Runoff from agricultural areas, sewage overflows, and faulty septic systems can contribute to the contamination of water sources. From 1946 to 2016, Cryptosporidium was responsible for 58% of all reported waterborne outbreaks caused by protozoa globally (Toledo et al., 2017). The largest recorded drinking waterrelated outbreak of cryptosporidiosis occurred in 1993 in Milwaukee, Wisconsin (USA). It affected approximately half of the population in the area resulting over 100 deaths (Mac Kenzie et al., 1994). Numerous other outbreaks have been associated with contaminated recreational water. Swimming in contaminated rivers, lakes, or swimming pools has been recognized as a significant transmission route for Cryptosporidium infection (Fayer et al., 2000; Fayer, 2004).

Cryptosporidium oocysts are resistant to adverse environmental conditions, allowing them to be present in diverse reservoirs including water sources. Presence of zoonotic species of Cryptosporidium has expanded the range of potential hosts, thus increasing the possibility of water source contamination (Smith & Rose, 1998; Hassan et al., 2021). Cryptosporidium infections affecting human and animal health have been reported from different areas of Sri Lanka. The reported cases include the cases from wild and livestock animals, as well as from humans, particularly from children. In this review, we provide an overview of Cryptosporidium infection in Sri Lanka and its impact by analyzing peer-reviewed research articles.

## MATERIALS AND METHODS

# Study protocol

This systematic review was focused on the occurrence and prevalence of *Cryptosporidium* in Sri Lanka adhering to PRISMA guidelines for systematic review and meta-analysis.

## Search strategy

Peer reviewed research articles published from 1987 to 2023, were extensively reviewed after retrieving them from five international online databases i.e., Google Scholar, PubMed, Medline, Web of Science and Scopus using "Cryptosporidium" and "Sri Lanka" as key words and MeSH terms ("Cryptosporidium" and "Sri Lanka"). After evaluating the inclusion and exclusion criteria, the review was done using the selected papers.

## Eligibility criteria

Freely accessible peer-reviewed full-text papers published

in the English-language were considered for review based on the inclusion and exclusion criteria. Two researchers examined the abstracts that were selected by referencing keywords. The following criteria had to be met: 1) the study should have been carried out in Sri Lanka; and 2) tested samples should have been collected from water sources, humans and animals. Review articles, editorials, comments and abstracts were excluded. Duplicates and those with no references in the English language were also eliminated (Figure 1).

# Screening and data extraction

Data extraction was done by two researchers independently into a pre-made excel document. The following information was included in the retrieved data: category/research sample (humans, primates, water, or other animals), paper title, author names, year of publication, study region, sample size, methods, and results (number of positive samples, prevalence, and major study findings).

#### **RESULTS**

Overall, 17 research articles, from Google Scholar and PubMed databases, were selected for the review (Table 1).

## Cryptosporidium in water

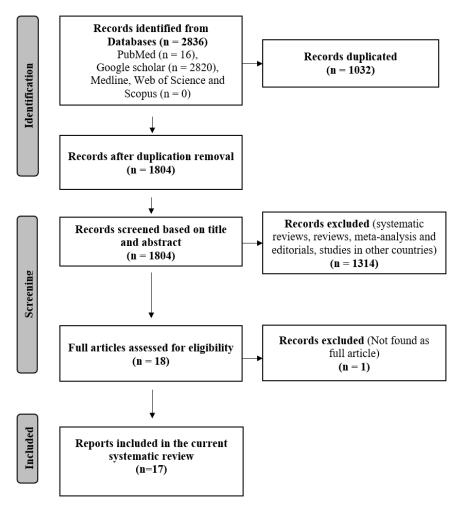
During our search for Cryptosporidium in water, we came across only one study in the literature, which was conducted in 2000 in the Southern region of Sri Lanka, near Sooriyawewa (W1) (Shortt et al., 2006). This is the only investigation on the detection of Cryptosporidium oocysts in Sri Lankan ground and surface waters. In order to detect oocysts, 49 Ls of water from each of four different types of water sources i.e., a shallow well, a canal, a reservoir, and a tube well have been sampled. The results revealed the presence of Cryptosporidium oocysts in all water sources. Notably, surface water exhibited a higher concentration of oocysts compared to the groundwater, while deep water sources showed significantly lower concentrations when compared to shallow water sources. As per the study, oocyst counts varied significantly based on the type of the water source (p<0.05). Moreover, no correlation was noted for the monthly variations in the oocyst numbers throughout the study period.

### Cryptosporidium in wild and captive non-human primates

Cryptosporidium infections are widespread among animals worldwide. It is commonly encountered protozoa in both wild and captive non-human primates. In Sri Lanka, only one study (Shortt et al., 2006) has been carried out so far to identify the parasite Cryptosporidium in the captive non-human primates. Eighty-five faecal samples from 15 primates were tested during the study period while restricting new animals to enter the experimental population. Out of the tested samples, only four primate species were positive for Cryptosporidium oocysts. The potential sources of contamination might be human-primate interactions and consumption of Cryptosporidium oocysts contaminated food.

Study of PW1 is the first paper that extensively reported *Cryptosporidium* infection in wild non-human primates

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**Figure 1:** Article selection for the current systematic review.

in Sri Lanka, from the Polonnaruwa Nature sanctuary and archaeological reserve. Faecal samples from three types of primates; toque macaques, gray langurs and highly arboreal purple-faced langurs were tested for the presence of oocysts using both microscopy and PCR. Based on the PCR more than one third of the primates were excreting Cryptosporidium oocysts. Toque macaques and gray langurs extensively feed on the ground whereas purplefaced langurs (highly arboreal) rarely feed on the ground. Hence, the infection might have been transmitted on the exposure to contaminated soiled ground and/or water. From toque macaques and gray langurs, individuals who exposed themselves to contaminated ground and water showed a higher prevalence of infection compared to the prevalence of infection shown by those from cleaner environments. All the positive animals were asymptomatic and 96% of the macaques, which were positive for Cryptosporidium oocysts, were co-infected with at least one species of another enteric parasite.

A molecular analysis of these infected animals was done in PW2, to identify the species and genotypes of *Cryptosporidium* in non-human primates. The study found that the three primate species were infected with four major clades of *C. parvum*, which were closely related to *C. parvum* type A (bovine or zoonotic) and type B genotypes. Those from uncleaned habitats with soiled grounds harboured isolates which were closely related to *C. parvum* 

type A while those from clean habitats harboured isolates closely related to *C. parvum* type B (not bovine type), suggesting livestock as the primary source of infection for wild primates. Individual primates, their social groups, and different species shared multiple genotypes of *C. parvum*, indicating the potential for the cross-species transmission of the parasite.

The PW3 study included two subspecies of toque macaquea: *Macaca sinica* from Polonnaruwa and *Macaca sinica aurifrons* from University of Peradeniya premises. More than 10% of both species were infected and, contaminated food and water appear to be the source of infection.

# Cryptosporidium in Livestock Farm Animals

Cryptosporidium was initially discovered in different farm animals, but its significance was overlooked until the early 1980s when it was identified as a primary cause of diarrhoea in farm animals (de Graaf, 1999). A comprehensive work undertaken by G1, G2, G3 and G4 in goats in Sri Lanka from 1998 to 2000 (Table 2) has made important findings on Cryptosporidium infection in goats in Sri Lanka. G1 study was done in dry zone of Sri Lanka. More than half of the animals examined excreted oocysts and more than 90% of them were asymptomatic. Prevalence of Cryptosporidium oocysts in three agro-climatic zones of Sri Lanka was observed in relation to the age of the animals (G2). More than one fourth of the goats from all three agro-

**Table 1:** Details of the articles used in the current review on *Cryptosporidium* infection in Sri Lanka.

Category	Paper ID	Authors	Location	Study Period
Water	W1	Shortt et al., 2006	UdaWalawe	August, September and November 2000
Wild and captive Primates	PW1	Ekanayake et al., 2006	Polonnaruwa	March to June 2001
	PW2	Ekanayake et al., 2007	Polonnaruwa	March to June 2001
	PC2	Umanga et al., 2012	National Zoological Gardens	August 2009 to February 2010
	PW3	Thilakarathne et al., 2021	University of Peradeniya and Polonnaruwa	June 2018 to September 2019
Livestock Farm Animal	G1	Noordeen et al., 1999	Puttalam and Anamaduwa	December 1998 to June 1999
	G2	Noordeen et al., 2000	Dry, intermediate and wet zones	October 1999
	G3	Noordeen et al., 2001	Anamaduwa	May 1999 to April 2000
	G4	Noordeen et al., 2002		
	S1	Senasinghe et al., 2002 (a)	Swine-belt area	
	CB1	Senasinghe et al., 2002 (b)	Dry zone	
	DC1	Abeywardena et al., 2014	Wet zone	
Human	H1	de Silva et al., 1994	Teaching Hospital Peradeniya	October 1992 &July 1993
	H2	Perera et al., 1999	Lady Ridgeway Children's Hospital	August 1987 to April 1989
	Н3	Sirisena et al., 2014	Kandy, Peradeniya, Matale hospitals and SirimawoBandaranayake Childrens' Hospital	August 2011 &February 2013
	H4	Rathnayake et al., 2015	Lady Ridgeway Hospital	September to November 2012
	Н5	Galgamuwa et al., 2016	Hanthana and Heerassagala	November 2013 to March 2014

climatic zones were infected with Cryptosporidium with a higher prevalence in the dry zone. Cryptosporidium oocyst counts were significantly higher in goat kids of <12 months age compared with goats >12 months of age in the dry and intermediate zones. Only 3% of the infected goat kids had diarrhoea and excreted oocysts in large numbers. A shot longitudinal study was done in Anamaduwa in the dry zone of Sri Lanka to examine seasonal variation and age of the animals in relation to Cryptosporidium infections in a group of goat kids recruited from 1 month of age and these animals were followed for one year (G3). A significant correlation between the prevalence of Cryptosporidium infection and the age of the animal was observed (p<0.01). However, Cryptosporidium oocyst excretion occurred irrespective of the rainfall pattern. A laboratory based study (G4) was conducted to examine the infectivity, oocyst shedding pattern, clinical manifestations, and intestinal morphological changes in goat kids infected with Cryptosporidium oocysts, isolated from naturally infected asymptomatic adult goats. Cryptosporidium oocysts obtained from goats were inoculated into mice too. Infected goat kids started to excrete oocysts 3-6 days after inoculation and reached its

maximum in 10-17 days followed by gradual decline up to 22 days of post inoculation. Similarly, the infected mice also started to excrete oocytes 3 days after inoculation. Ileum of infected goat kids showed endogenous stages of *C. parvum* on the brush border of the enterocytes, and infiltration of neutrophils and mononuclear cells into the lamina propria. Ileum also showed morphological changes, which included atrophy, stunting, fusion and denudation of villi. Similarly, histological sections of mice showed the presence of endogenous stages of *Cryptosporidium* on the microvilli of the ileum. Findings of the study indicated that *Cryptosporidium* oocysts, isolated from asymptomatic adult goats, can infect both mice and goat kids emphasizing the source of transmission and public health significance.

In 2002, presence of *Cryptosporidium* oocysts in cattle and buffaloes, managed extensively in a mixed farm located in the dry zone, was investigated (CB1). All animals shedding oocysts in the faeces were asymptomatic and exclusively had Genotype 2 of *C. parvum*. As these animals lived closely with children and shared common surface water sources with humans, there was an increased risk of

transmission of this zoonotic Genotype 2 of the *C. parvum* from animals to susceptible humans.

Molecular epidemiological analysis was conducted (DC1) in domestic bovids in selected regions of Sri Lanka to observe whether they excrete *Cryptosporidium* of zoonotic potential. Based on the phylogenetic analysis of a few samples, new genotypes of *Cryptosporidium* were recorded: *C. bovis, C. ryanae* and new six genotypes in cattle and two other new genotypes in water buffaloes. This study found no evidence of *C. parvum*, the most commonly reported zoonotic species of *Cryptosporidium* from bovine calves globally.

Another study (Senasinghe et al., 2002a) carried out on the prevalence of *Cryptosporidium* infection in pigs along the pig belt area of Sri Lanka showed *Cryptosporidium* oocysts positivity in a few animals, and the oocysts were confirmed to be *C. parvum*.

## Cryptosporidium in humans

Cryptosporidiosis plays a major role in childhood diarrhoea in developing countries. However, only a few studies have been carried out on human excretion of Cryptosporidium oocysts in association with diarrhoea in Sri Lanka. A hospital-based study was conducted focusing on densely populated low-income areas close to the hospitals in Colombo, Kandy and Matale. The study revealed that children under the age of 12 excrete Cryptosporidium oocysts. The mean age at risk of getting the infection is 2.2 years. Stools with blood and mucus could be observed when Cryptosporidium was associated with bacterial or viral pathogens. Infection was high when these children did not use boiled cooled water for drinking, and were associated with pets including dogs, cats, goats and chicken. In the H3 study, the pathogen was confirmed to be C. Parvum by PCR, showing the potential of zoonotic implications.

**Table 2:** Data on *Cryptosporidium* infection in Sri Lanka from selected studies.

Paper ID	Sample size	Method	Prevalence by category	Mean oocysts counts (oocysts per L or grams)
W1	12 water samples	Filtration sugar flotation MZN staining	Overall prevalence -83% Irrigation reservoir - 100% Canal - 100% Shallow wells - 100% Tube wells - 33%	Irrigation reservoir and canal - 8 Tube wells and shallow wells - 1
PW1	125 monkeys 89 toque macaques 21 gray langurs 15 purple-faced langurs	Sheather's sucrose solution MZN staining Nested PCR	Overall prevalence – by microscopy (M) – 27% Overall prevalence by PCR – 40% Toque macaques - microscopy – 29%, PCR - 44% Gray langurs – microscopy- 38%, PCR - 48% Purple-faced langur – microscopy - 0%, PCR - 26%	Toque macaques - 3,633 Gray langurs - 2,407 Juvenile toque monkey - 34,250
PC2	15 species of primate, fecal samples	MZN staining	Overall prevalence –26.7%	
PW3	98 monkeys	Sheather's sucrose floatation method	Overall prevalence –11.2% Macacasinica- 10.2% Macacasinica aurifrons-12.2%	Overall - 1.36  Macacasinica- 1.20  Macacasinica aurifron- 1.50
G1	200 goats	sucrose flotation technique MZN staining	Overall prevalence -55% 0-11 months - 53% 12-48 months - 30%	Non-diarrhoeic animals (91%) - 250-5000 diarrhoeic animals (9%) - 5000-25000
G2	1020 goats dry zone – 512 intermediate zone – 287 wet zone – 221	Sheather's sucrose solution MZN staining	Overall prevalence –28.5% Dry zone – 33.6% Intermediate zone– 24.7% Wet zone– 21.7% <12 months – Dry zone – 44.15, Intermediate zone – 27.3% and Wet zone – 19.05% >12 months – Dry zone - 22.8%, Intermediate zone – 10.5% and Wet zone – 25.2%	Non-diarrhoeic animals - 383 diarrhoeic animals - 6814

G3	72 goats	MZN staining	Overall prevalence $-34\%1-6$ months $-36.7\%$ 7-12 months $-9.7%$	633
S1	72 faecal samples	Salt flotation MZN staining PCR	Overall prevalence –9%	429
CB1	50 fecal samples, 20 cattle, 20 buffalos and 10 goats	Salt flotation method MZN staining PCR	Cattle–65% Buffaloes–25% Goats–0%	Cattle - 1900±770 Buffaloes -190±123
DC1	cattle 340 and water buffaloes 297	PCR sequencing phylogenetic approach	Cattle (<3 months) −62.1% Water buffaloes −9.8 % (<6 months − 8.4% ≥6 months − 1.3%)	ND
H1	192 adults and 354 children	Cold Ziehi-Neelsen staining	Adult stool samples –0% Children–0.3%	ND
Н2	940 diarrhoea samples 260 controls samples	MZN staining	Cases – 2.7% Control – 0.4% Cryptosporidium as the only parasite – 1.8% Cryptosporidium + Rotavirus – 0.6% (50% had blood and mucus diarrhoea) Cryptosporidium + Salmonella spp.– 0.3%	ND
Н3	138 fecal samples (children under 12 years of age)	Modified formalin ethyl acetate floatation MZN staining PCR	Overall prevalence – 5.8%  Overall prevalence – 5.1% were confirmed <i>C. parvum</i> by PCR 5.1% had watery diarrhoea 4.3% had contact with dogs, cats, goats and chicken 4.3% had fever 1.4% did not drink boiled cooled water	ND
H4	145 stool samples of Children (less than 12 years)	MZN staining	Overall prevalence – 33.1% <1 years – 27.1% 1 – 5 years – 12.2% 7 – 11 years – 2.5%	ND
H5	489 children (Children below 12 years of old)	Formaline- ethyl acetate concentration MZN staining	Overall prevalence – 0%.	ND

 $MZN-Modified\ Ziehi-Neelsen;\ PCR-Polymerase\ chain\ reaction;\ ND-Oocysts\ counts\ not\ done.$ 

## **DISCUSSION**

Cryptosporidiosis is a global health concern due to its zoonotic nature and contagiousness. *Cryptosporidium* infection in animals and humans causes diarrhoea, abdominal cramps, nausea, and vomiting (Abeywardena et al., 2015). In Sri Lanka, the prevalence of *Cryptosporidium* infection varies, and the variation is influenced by factors like water quality, sanitation practices, and standards of hygiene. Proper management of these factors is crucial to effectively control and reduce the transmission of *Cryptosporidium* infection.

In Udawalawa (W1), *Cryptosporidium* oocysts were examined from a variety of water sources, including a canal, a small reservoir, a tube well, and a shallow well

(W1). All four water sources were contaminated with oocysts and the contamination was higher in surface water than in well water. *Cryptosporidium* oocyst contamination was higher in shallow well compared to the deep well. The contamination of river water was attributed to agricultural land use along the water bodies, especially where manure has been used as fertilizer or for cattle farming (Farizawati et al., 2005; Kistemann et al., 2016; Hamilton et al., 2018). Faecal contamination from various animals, including domestic, agricultural, and wildlife species, is the primary cause for the presence of oocysts in river water (Kistemann et al., 2016). This pattern of contamination is evident in the areas, where major paddy cultivation occurs, and water buffalos bath and contaminate water. Shallow wells, canals, and reservoirs have been preferred for obtaining residential

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water over tube wells due to the high levels of salt, iron, and fluoride content in the tube well water. Although some families choose to boil the shallow-well water before consuming, water is usually consumed untreated by the majority and this poses a risk of waterborne transmission of *Cryptosporidium* oocysts.

The findings of the review show that waterborne transmission is the most prevalent mode of infection with Cryptosporidium, which is common in humans as well as in animals (Shortt et al., 2006; Sirisena et al., 2014). The human studies were mainly conducted in hospitalized patients (H1, H2, H3 and H4), and pre-school children (H1). Children are used as the frequent study population (92.4%) compared to the adult population probably due to their agerelated risk of acquiring the infection. In accordance to the studies (H1 and H4), Cryptosporidium causes serious infections in immuno-compromised individuals and is a common cause of diarrhoea in children below the age of 5 years. The clinical manifestations of cryptosporidiosis are also seen in association with other pathogens like Salmonella or rotavirus infections, which trigger blood and mucus diarrhoea (H1). According to the GBD Diarrhoeal Diseases Collaborators in 2017, cryptosporidiosis is the fourth leading cause of death among children under the age of 5 years. It was the cause of 12.1% of all fatalities in children under the age of 5 years in 2015 and this shows the impact of Cryptosporidium infection on childhood mortality worldwide.

Many studies have been conducted in different countries to obtain the prevalence of Cryptosporidium infection in animals. The studies conducted in Iran emphasized the influence of geographical region and climate on the prevalence of animal cryptosporidiosis, highlighting the variability of Cryptosporidium species across different regions (Haghi et al., 2020). Five studies revealed the situation in goats (G1, G2 and G3), goats and buffaloes (CB1) as well as goats and mice (G4). A total of 1295 goats were used in 3 studies conducted in north-western region and dry-wet zones of Sri Lanka. G1 and G2 studies concluded that 91% infected goats were asymptomatic while high oocyst counts indicated symptomatic Cryptosporidium infection. Moreover, prevalence of Cryptosporidium infection and the genotypes of the parasite (DC1) have been reported in buffaloes and cattle in dry zone (CB1) and wet zone (DC1). This work emphasized the high prevalence of Cryptosporidium infection in cattle (50% in CB1) in Sri Lanka. A similar study was conducted in Japan highlighting the high prevalence of Cryptosporidium infections in cattle (El-Alfy and Nishikawa, 2020). There were notable differences in the species and prevalence among the various breeds of cattle, indicating breed, age, and regional correlations (Gong et al., 2017).

The modified Ziehl-Neelsen staining followed by microscopy has been used for the identification of *Cryptosporidium* oocysts by all selected studies for the current review. Some studies have used PCR for identifying the *Cryptosporidium* species and genotypes of the parasite. As it is evident, microscopy alone is insufficient to interpret the absence of *Cryptosporidium* infection as low oocysts

numbers may not be detected by microscopy (PW1).

Overall, cryptosporidiosis is a parasitic disease that is widespread and has an impact on animal and human health, and thereby on one health. Advanced PCR techniques must be used to screen suspected animal and human samples, and water samples for *Cryptosporidium* infection in Sri Lanka. A multi-faceted strategy involving improved water supply, personal hygiene and health promotion measures is essential to prevent and control *Cryptosporidium* infections.

### **CONCLUSION**

This review offers an in-depth summary of the existing knowledge on *Cryptosporidium* infections in animals and humans, and contaminations in the water bodies in Sri Lanka. The findings of this review highlight the need for future research to expand our knowledge on the subject and the use of effective screening methods for parasite identification to prevent the spread of *Cryptosporidium* species to minimize their negative impacts on One Health.

#### REFERENCES

Abeywardena, H., Jex, A.R., Koehler, A.V., Rajapakse, R.J., Udayawarna, K., Haydon, Stevens, M.A. and Gasser, R.B., (2014). First molecular characterization of Cryptosporidium and Giardia from bovines (Bos taurus and Bubalus bubalis) in Sri Lanka: unexpected absence of C. parvum from pre-weaned calves. *Parasites & Vectors*, 7(1), 1-10.

Abeywardena, H., Jex, A.R. and Gasser, R.B., (2015). A Perspective on *Cryptosporidium* and *Giardia*, with an Emphasis on Bovines and Recent Epidemiological Findings. *Advances in Parasitology*, **88**, 243–301. https://doi.org/10.1016/bs.apar.2015.02.001

Angus, K.W., Tzipori, S. and Gray, E.W., (1982). Intestinal lesions in specific-pathogen-free lambs associated with a *Cryptosporidium* from calves with diarrhoea. *Veterinary Pathology*, 19(1), 67-78. https:// doi.org/10.1177/030098588201900110

Carmena, D., (2010). Waterborne transmission of *Cryptosporidium* and *Giardia*: detection, surveillance and implications for public health. *Current Research, Technology and Education Topics in Applied Microbiology and Microbial Biotechnology*, **20**, 3-4.

De Silva, N.R., De Silva, H.J. and Jayapani, V.P.P., (1994). Intestinal parasitoses in the Kandy area, Sri Lanka. Southeast Asian Journal of Tropical Medicine and Public Health, 25, 469-469.

Donnelly, J.K. and Stentiford, E.I., (1997). The *Cryptosporidium* problem in water and food supplies. *LWT-Food Science and Technology*, **30**(2), 111-120. https://doi.org/10.1006/fstl.1996.0168

Ekanayake, D.K., Arulkanthan, A., Horadagoda, N.U., Sanjeevani, G.M., Kieft, R., Gunatilake, S. and Dittus, W.P., (2006). Prevalence of *Cryptosporidium* and other enteric parasites among wild non-human primates in Polonnaruwa, Sri Lanka. *The American Journal of Tropical Medicine and Hygiene*, **74**(2), 322-329. https://doi.org/10.4269/ajtmh.2006.74.322

Ekanayake, D.K., Welch, D.M., Kieft, R., Hajduk, S.

- and Dittus, W.P., (2007). Transmission dynamics of *Cryptosporidium* infection in a natural population of non-human primates at Polonnaruwa, Sri Lanka. *The American Journal of Tropical Medicine and Hygiene*, 77(5), 818-822. https://doi.org/10.4269/ajtmh.2007.77.818
- El-Alfy, E. S., & Nishikawa, Y. (2020). *Cryptosporidium* species and cryptosporidiosis in Japan: a literature review and insights into the role played by animals in its transmission. *The Journal of Veterinary Medical Science*, **82**(8), 1051–1067. https://doi.org/10.1292/jvms.20-0151
- Farizawati, S., Lim, Y.A.L., Ahmad, R.A., Fatimah, C.T.N.I. and Siti-Nor, Y., (2005). Contribution of cattle farms towards river contamination with Giardia cysts and *Cryptosporidium* oocysts in Sungai Langat Basin. *Tropical Biomedicine*, **22**(2), 89-98.
- Fayer, R., Morgan, U. and Upton, S.J., (2000). Epidemiology of *Cryptosporidium*: transmission, detection and identification. *International Journal for Parasitology*, 30(12-13), 1305-1322. https://doi.org/10.1016/S0020-7519(00)00135-1
- Fayer, R., (2004). *Cryptosporidium*: a water-borne zoonotic parasite. *Veterinary Parasitology*, **126**(1-2), 37-56. https://doi.org/10.1016/j.vetpar.2004.09.004
- Galgamuwa, L.S., Iddawela, D. and Dharmaratne, S.D., (2016). Intestinal protozoa infections, associated risk factors and clinical features among children in a low-income tea plantation community in Sri Lanka. *International Journal of Community Med Public Health*, **3**(9), 2452-2458. DOI: http://dx.doi.org/10.18203/2394-6040.ijcmph20163053
- GBD Diarrhoeal Diseases Collaborators (2017). Estimates of global, regional, andnational morbidity, mortality, and aetiologies of diarrhoeal diseases: a systematic analysis for the Global Burden of Disease Study 2015. Lancet Infect. Dis.17, 909–948. https://doi.org/10.1016/S1473-3099(17)30276-1
- Gerace, E., Presti, V.D.M.L. and Biondo, C., (2019). Cryptosporidium infection: epidemiology, pathogenesis, and differential diagnosis. European Journal of Microbiology and Immunology, 9(4), 119-123. DOI: https://doi.org/10.1556/1886.2019.00019
- deGraaf, D.C., Vanopdenbosch, E., Ortega-Mora, L.M., Abbassi, H. and Peeters, J.E., (1999). A review of the importance of cryptosporidiosis in farm animals. *International Journal for Parasitology*, **29**(8), 1269-1287. https://doi.org/10.1016/S0020-7519(99)00076-4
- Gong, C., Cao, X. F., Deng, L., Li, W., Huang, X. M., Lan, J. C., Xiao, Q. C., Zhong, Z. J., Feng, F., Zhang, Y., Wang, W. B., Guo, P., Wu, K. J., & Peng, G. N. (2017). Epidemiology of *Cryptosporidium* infection in cattle in China: A review. *Parasite (Paris, France)*, 24, 1. https://doi.org/10.1051/parasite/2017001
- Haghi, M. M., Khorshidvand, Z., Khazaei, S., Foroughi-Parvar, F., Sarmadian, H., Barati, N., Etemadifar, F., &Ghasemikhah, R. (2020). *Cryptosporidium* animal species in Iran: a systematic review and meta-analysis. *Tropical Medicine and Health*, 48(1), 97. https://doi.org/10.1186/s41182-020-00278-9

- Hamilton, K.A., Waso, M., Reyneke, B., Saeidi, N., Levine, A., Lalancette, C., Besner, M.C., Khan, W. and Ahmed, W., (2018). *Cryptosporidium* and *Giardia* in wastewater and surface water environments. *Journal of Environmental Quality*, 47(5), 1006-1023. https://doi. org/10.2134/jeq2018.04.0132
- Hassan, E.M., Örmeci, B., DeRosa, M.C., Dixon, B.R., Sattar, S.A. and Iqbal, A., (2021). A review of *Cryptosporidium* spp. and their detection in water. *Water Science and Technology*, **83**(1), 1-25. https://doi.org/10.2166/wst.2020.515
- Kistemann, T., Schmidt, A. and Flemming, H.C., (2016). Post-industrial river water quality- Fit for bathing again? *International Journal of Hygiene and Environmental Health*, **219**(7), 629-642. https://doi.org/10.1016/j.ijheh.2016.07.007
- Mac Kenzie, W.R., Hoxie, N.J., Proctor, M.E., Gradus, M.S., Blair, K.A., Peterson, D.E., Kazmierczak, J.J., Addiss, D.G., Fox, K.R., Rose, J.B. and Davis, J.P., (1994). A massive outbreak in Milwaukee of *Cryptosporidium* infection transmitted through the public water supply. *New England Journal of Medicine*, 331(3), 161-167. https://doi.org/10.1056/NEJM199407213310304
- Mason, R.W., Hartley, W.J. and Tilt, L., (1981). Intestinal cryptosporidiosis in a kid goat. *Australian Veterinary Journal*, **57**(8), 386-388. https://doi.org/10.1111/j.1751-0813.1981.tb00529.x
- Navin, T.R. and Juranek, D.D., (1984). Cryptosporidiosis: clinical, epidemiologic, and parasitologic review. *Reviews of Infectious Diseases*, **6**(3), pp.313-327. https://doi.org/10.1093/clinids/6.3.313
- Noordeen, F., Rajapakse, R.P.V.J., Faizal, A.C.M. and Horadagoda, N.U., 1999. Prevalence of *Cryptosporidium* infection among goats in selected areas of the dry zone of Sri Lanka. *Tropical Agricultural Research*, 11, 365-371.
- Noordeen, F., Rajapakse, R.P.V.J., Faizal, A.C.M., Horadagoda, N.U. and Arulkanthan, A., (2000). Prevalence of *Cryptosporidium* infection in goats in selected locations in three agroclimatic zones of Sri Lanka. *Veterinary Parasitology*, 93(2), 95-101. https:// doi.org/10.1016/S0304-4017(00)00361-7
- Noordeen, F., Faizal, A.C.M., Rajapakse, R.P.V.J., Horadagoda, N.U. and Arulkanthan, A., (2001). Excretion of *Cryptosporidium* oocysts by goats in relation to age and season in the dry zone of Sri Lanka. *Veterinary Parasitology*, **99**(1), 79-85. https://doi.org/10.1016/S0304-4017(01)00449-6
- Noordeen, F., Horadagoda, N.U., Faizal, A.C.M., Rajapakse, R.P.V.J., Razak, M.A.A. and Arulkanthan, A., (2002). Infectivity of *Cryptosporidium parvum* isolated from asymptomatic adult goats to mice and goat kids. *Veterinary Parasitology*, **103**(3), 217-225. https://doi.org/10.1016/S0304-4017(01)00578-7
- Noordeen, F., Rajapakse, R.P.V.J., Horadagoda, N.U., Abdul-Careem, M.F. and Arulkanthan, A., (2012). *Cryptosporidium*, an important enteric pathogen in goats—A review. *Small Ruminant Research*, **106**(2-3), 77-82. https://doi.org/10.1016/j. smallrumres.2012.03.012

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Panciera, R.J., Thomassen, R.W. and Garner, F.M., (1971). Cryptosporidial infection in a calf. *Veterinary Pathology*, **8**(5-6), 479-484. https://doi.org/10.1177/0300985871008005-00610

- Perera, J., Jayawardene, I., Mendis, L. and Abeyratne, K., (1999). Intestinal parasites and diarrhoea in a children's hospital in Sri Lanka. *Ceylon Journal of Medical Science*, **42**(1). 7-12. https://doi.org/10.4038/cjms.v42i1.4883
- Rathnayake, R.G.L., Ranchigoda, C.M., Roshan, R.G., Samarasinghe, S. and Banneheke, B.M.H.A., (2015). Laboratory investigation of cryptosporidiosis and giardiasis in children with Diarrhoea: a hospitalbased study. Proceedings of 8th International Research Conference, KDU, Published November 2015.
- Santín, M., (2013). Clinical and subclinical infections with *Cryptosporidium* in animals. *New Zealand Veterinary Journal*, **61**(1), 1-10. https://doi.org/10.1080/0048016 9.2012.731681
- Sardar, S.K., Ghosal, A., Saito-Nakano, Y., Dutta, S., Nozaki, T. and Ganguly, S., (2021). Molecular Identification of *Cryptosporidium viatorum* infection in a patient suffering from unusual cryptosporidiosis in West Bengal, India. *The Korean Journal of Parasitology*, **59**(4), 409. https://doi.org/10.3347/kjp.2021.59.4.409
- Sharma, K., Srivastava, S., & Tak, V. (2023). Cryptosporidiosis in India and the World: A Review. Infectious Disorders-Drug Targets (Formerly Current Drug Targets-Infectious Disorders), 23(5), 46-52. https://doi.org/10.2174/1871526523666230403103344
- Senasinghe, N.D., Rajapaksha, W.R.A.K.J.S., Faizal, A.C.M., Rajapakse, R.P.V.J. and Widmer, G., (2002). Occurrence of zoontic genotype of *Cryptosporidium parvum* in cattle an buffaloes managed extensively in mixed livestock farms in the dry zone. Tropical Agricultural Research, **14**, 357-361.
- Senasinghe, N.D., Rajapaksha, W.R.A.K.J.S., Faizal, R.P.V.J and Widmer, G., (2002). Identification of *Cryptosporidium parvum* oocysts isolated from swine by PCR technique. *Tropical Agricultural Research*, **14**, 362-364.

- Shortt, R.L., Boelee, E., Matsuno, Y., Madramootoo, C., Van Der Hoek, W. and Faubert, G., (2006). Cryptosporidium and Giardia as determinants for selection of an appropriate source of drinking water in southern Sri Lanka. Journal of Health, Population and Nutrition, 24(1), 64-70.
- Smith, H.V. and Rose, J.B., (1998). Waterborne cryptosporidiosis: current status. *Parasitology Today*, **14**(1), 14-22. https://doi.org/10.1016/S0169-4758(97)01150-2
- Sirisena, U.M., Iddawela, W.M.D.R., Noordeen, F. and Wickramasinghe, S., (2014). Prevalence and identification of *Cryptosporidium* species in paediatric patients with diarrhoea. *Ceylon Medical Journal*, **59**(3), 75-78. https://doi.org/10.4038/cmj.v59i3.7467
- dos Santos Toledo, R., Martins, F.D.C. and Freire, R.L., (2017). Waterborne *Giardia* and *Cryptosporidium*: contamination of human drinking water by sewage and cattle faeces. *Semina: Ciências Agrárias*, **38**(5), 3395-3415. https://doi.org/10.5433/1679-0359.2017v38n5p3395
- Thilakarathne, S.S., Rajakaruna, R.S., Fernando, D.D., Rajapakse, R.P.V.J. and Perera, P.K., (2021). Gastro-intestinal parasites in two subspecies of toque macaque (*Macacasinica*) in Sri Lanka and their zoonotic potential. *Veterinary Parasitology: Regional Studies and Reports*, **24**, 100558. https://doi.org/10.1016/j. vprsr.2021.100558
- Troeger, C., Forouzanfar, M., Rao, P.C., Khalil, I., Brown, A., Reiner, R.C., Fullman, N., Thompson, R.L., Abajobir, A., Ahmed, M. and Alemayohu, M.A., (2017). Estimates of global, regional, and national morbidity, mortality, and aetiologies of diarrhoeal diseases: a systematic analysis for the Global Burden of Disease Study 2015. *The Lancet Infectious Diseases*, 17(9), 909-948. https://doi.org/10.1016/S1473-3099(17)30276-1
- Tyzzer, E.E., (1907). A sporozoan found in the peptic glands of the common mouse. *Proceedings of the Society for Experimental Biology and Medicine*, **5**(1), 12-13. https://doi.org/10.3181/00379727-5-5