

## Prevalence of Cryptosporidiosis Among School Going Children in Kathmandu, Nepal

Shardulendra Prasad Sherchand<sup>1,7</sup>, Dev Raj Joshi<sup>2</sup>, Nabaraj Adhikari<sup>3</sup>, Ram Chandra Poudel<sup>1</sup>, Krishna Prasad Pant<sup>4</sup>, Madhav KC<sup>5</sup>, Dipendra Shrestha<sup>1</sup> and Samendra Sherchan<sup>6\*</sup>

<sup>1</sup>National College, Khusibu, Kathmandu, Nepal

<sup>2</sup>Central Department of Microbiology, Kirtipur, Kathmandu, Nepal

<sup>3</sup>Kantipur College of Medical Sciences, Sitapila, Kathmandu, Nepal

<sup>4</sup>Department of Microbiology, Siddhanath Science Campus, Mahendranagar, Kanchanpur, Nepal

<sup>5</sup>Louisiana State University Health Sciences Center School of Public Health, New Orleans, LA, 70112, United States

<sup>6</sup>Department of Global Environmental Health Sciences, Tulane University, New Orleans, LA, 70112, United States

<sup>7</sup>Department of Microbiology, Immunology and Parasitology, Louisiana State University Health Sciences Center, New Orleans, LA, 70112, United States

**\*Corresponding Author:** Samendra Sherchan, Assistant Professor, Department of Global Environmental Health Sciences, Tulane University, New Orleans, LA, 70112, United States.

**Received:** September 19, 2016; **Published:** October 31, 2016

### Abstract

**Background:** Intestinal parasitic infection among children poses a critical public health issue in Nepal. This study was conducted to determine cryptosporidiosis among school going children in Kathmandu, Nepal.

**Methodology:** A total of 187 collected stool samples were subjected to direct microscopy, formal-ether concentration technique, Sheather's sucrose floatation technique and modified acid fast staining technique.

**Results:** The occurrence of cryptosporidiosis was 29.4%. Cryptosporidiosis was high in female (35.2%) without any significance ( $p = 0.100$ ). Similarly, cryptosporidiosis was highest in age group 5 - 8 (31.8%) which was not statistically significant ( $p = 0.619$ ). The cryptosporidiosis was higher in symptomatic case (31.0%) than asymptomatic case, which was insignificant ( $p = 0.475$ ). The cryptosporidiosis was higher in children not following hygienic practice (35.6%) with significance ( $p = 0.015$ ).

**Conclusion:** The outcome of this study revealed that cryptosporidiosis infections remain highly endemic among school children in Kathmandu, Nepal. This suggests proper diagnosis; specific treatment and adequate WaSH (Water, Sanitation and Hygiene) practices are required to lower the parasitic burden in low-resource settings.

**Keywords:** Cryptosporidiosis; Children; Enteric Parasites; Nepal

### Introduction

*Cryptosporidium* spp. is a leading cause of persistent diarrhea in developing countries. This coccidian parasite causes a self-limiting diarrhoeal infection in immunocompetent individuals, and full recovery is expected with proper oral hydration therapy [1]. The organism can cause illnesses lasting longer than 1 or 2 weeks in previously healthy persons or indefinitely in immunocompromised patients;

furthermore, in young children in developing countries, cryptosporidiosis predisposes to substantially increased diarrhoeal illnesses. The first case of human cryptosporidiosis was reported in 1976, and since then, it has been widely studied. Several studies suggest that it spreads in daycare centers and possibly in widely distributed water supplies, public pools, and institutions such as hospitals and extended care facilities for the elderly [2].

There are 27 species of *Cryptosporidium* that are pathogenic to humans [3,4]. The oocyst stage is of primary importance for the dispersal, survival, and infectivity of the parasite and is of major importance for detection and identification [5]. In WHO bulletin, the reported prevalence of *Cryptosporidium parvum* is 1 - 4% in Europe and North America; and 3 - 20% in Africa, Asia, Australia, South and Central America. Peaks in the prevalence in developed countries are observed in spring and in the late summer. Numbers of asymptomatic carriers, as determined by stool surveys, are generally very low (< 1%) in industrialized countries, although higher rates have been reported in day-care centers. Routine bile endoscopy suggests a higher prevalence of asymptomatic carriage: 13% of non-diarrheic patients were shown to carry *Cryptosporidium parvum* oocysts. High rates of asymptomatic carriage (10 - 30%) are common in non-industrialized countries. Seroprevalence rates are generally higher than fecal carriage rates, from 25 - 35% in industrialized countries to 95% in South America increase with age and are relatively high in dairy farmers and day-care centre attendants [6].

Intestinal parasitic infection is still a serious public health in Nepal and there is little data on the prevalence of intestinal parasites among school going children in Nepal [7,8].

Therefore, the present research has been designed to study the prevalence of cryptosporidiosis among school going children in Kathmandu, Nepal and its relation with behavioral pattern and life style of the children relating to health.

**Methods**

A total of 187 stool samples were collected from school going children (n = 187) of Kathmandu valley in a screw capped plastic container. All the samples were processed in Microbiology Laboratory of National College. This study was approved by the Institutional Review Board, National College and the informed consents were obtained from the participants. A questionnaire on age, sex, hygienic practice (hand-washing) and gastrointestinal symptoms (diarrhea) were filled and used for analysis. Each fresh stool sample was examined macroscopically and microscopically for the detection of *Cryptosporidium* Spp. Microscopic examination of sample was done by standard formol- ether concentration method. Sheather’s sucrose floatation method followed by modified acid fast staining was conducted for detecting oocysts of *Cryptosporidium*. All the *Cryptosporidium* positive specimens were stored at ambient temperature in 2.5% potassium dichromate for about 10 days and were examined for sporulation as described by Ghimire., *et al.* [9]. The data obtained were analyzed by statistical software SPSS (version 11.5).

**Results**

The Cryptosporidiosis was found as high as 29.4%. The gender wise distribution of *Cryptosporidium* spp. in school going children revealed higher prevalence in female school going children (35.2%) than in male school going children (24.2%) with no significant difference (Table 1). *Cryptosporidium* spp. was present highest in age group 5 - 8 (31.8%). The occurrence of *Cryptosporidium* spp. in different age groups was also found statistically insignificant (Table 2).

Sex	Frequency (n)	Positive (n)	%	p-value
Male	99	24	24.2	p = 0.100
Female	88	31	35.2	
Total	187	55	29.4	

**Table 1:** Occurrence of *Cryptosporidium* spp. with gender of school going children.

Age	Frequency (n)	Positive (n)	%	p-value
3-5 years	46	11	23.9	p = 0.619
5-8 years	107	34	31.8	
8-12 years	34	10	29.4	
Total	187	55	29.4	

Table 2: Distribution of *Cryptosporidium* spp. with age.

The occurrence of *Cryptosporidium* spp. in symptomatic case (31.0%) was higher than asymptomatic case (25.9%) with no statistically significance (Table 3). The occurrence of *Cryptosporidium* spp. in school going children not following hygienic practice (35.6%) was higher than with following good hygiene practice (18.8%) and the result was found statistically significant (Table 4).

Symptoms	Frequency (n)	Positive (n)	%	p-value
Symptomatic	129	40	31.0	P = 0.475
Asymptomatic	58	15	25.9	
Total	187	55	29.4	

Table 3: Symptom wise distribution of *Cryptosporidium* spp.

Hygienic Practice	Frequency (n)	Positive (n)	%	p-value
Following	69	13	18.8	p = 0.015
Not Following	118	42	35.6	
Total	187	55	29.4	

Table 4: Distribution of *Cryptosporidium* spp. with hygiene practice.

### Discussion

In this present study, prevalence of *Cryptosporidium* spp. was detected as 29.4%. This high prevalence rate may be due to various factors such as geographical factors, age of the school going children, lack of proper sanitation, lack of proper awareness among school going children, lack of personal hygiene, lack of properly treated water, inadequately washed hand, lack of specific treatment, consumption of undercooked food, consumption of contaminated fruits and overall diminished socioeconomic condition of pupils of different schools of Kathmandu Valley who were included in this study. The current prevalence figure was higher than figures revealed by different studies in Nepal [9-13]. Similarly, the present finding was higher than the findings of Filipe., *et al.* [14], Bern., *et al.* [15], Nimri [16], Easow., *et al.* [17] and Latif and Rossle [21] where they found the infection rates to be 9.3%, 1.2%, 8%, 1%, 8.6% and 4.62% respectively. However, the current finding was more or less similar to the findings of Laubach., *et al.* [18], Mahgoub., *et al.* [20] and Munoz-Antoli., *et al.* [22], where they found the prevalence of cryptosporidiosis as high as 32%, 37.3% and 35.7% in Gautemala, Jordan and Nicaragua respectively.

This endemic, zoonotic coccidian parasitosis is highly prevalent in third-world countries that have waterborne fecal contamination of food and water with oocysts and where there is direct infection by person-to-person contact as the common method of transmission of this enteric protozoan [19]. These results suggest that there is heavy contamination of water, food, animal and soil with the oocysts of *Cryptosporidium* which could be the potential source of infection.

The gender wise distribution of Cryptosporidiosis in school going children revealed higher prevalence in female school going children (35.2%) than in male school going children (24.2%), which was statistically insignificant (p = 0.100). Although there is no sexual predi-

lection in the cryptosporidiosis, but the current finding clearly suggests higher prevalence among female school going children. The exact reason behind the high prevalence among female school going children is unknown and it may be associated with cooking and household work such as handling livestock and storing water [20]. Cryptosporidiosis was present highest in age group 5 - 8 (31.8%) but it was found statistically insignificant ( $p = 0.619$ ). The finding of higher prevalence of cryptosporidiosis in children with age less than 8 is similar to the findings of Bern., *et al.* [15], Easow., *et al.* [17] and Sherchand and Shrestha [9]. The high prevalence of infection in children under 8 years old could be attributed to the extensive use of untreated water, unsafe sanitation and contaminated food in Nepal [21]. Recent studies have shown that the gut microbiome plays a vital role in children's health [23-27] including protection against pathogens and contributing to metabolic functions. In this context, the occurrence of Cryptosporidiosis in symptomatic case (31.0%) was higher than asymptomatic case (25.9%) but it was found statistically insignificant ( $p = 0.475$ ). This insignificant result may be due to the limitation in conventional microscopy diagnostics. PCR-based methods would probably increase this figure considerably.

The occurrence of *Cryptosporidium* spp. in school going children not following hygienic practice (35.6%) was higher than with following hygienic practice (18.8%) and the result was found statistically significant ( $p = 0.015$ ). *Cryptosporidium* oocysts can be directly transmitted by fecal oral route as a result of which good hygiene practice (handwashing practice) is the best way to prevent cryptosporidiosis.

### Conclusion

Cryptosporidiosis is one of the most prevalent intestinal protozoal parasites, which is endemic in Nepal. The present research reveals that cryptosporidiosis is abundant among school going children in Kathmandu, Nepal. Effective control strategies, proper hygienic education, improved sanitation and safe water supply are necessary to prevent *Cryptosporidium* infections in low-resource settings.

### Acknowledgements

The authors extend their honest gratitude to school going children of Ganesh Himal Secondary School, Samakhusi, Kathmandu; Kopila English Secondary School, Siddhipur, Lalitpur; and Samata Shikshya Niketan, Kamalvinayak, Bhaktapur for providing stool samples and National College for providing the laboratory facilities.

### Disclosures

The authors declare no potential/perceived conflicts of interest in the study.

### Bibliography

1. White AC Jr. "Cryptosporidiosis (*Cryptosporidium* species). Bennett JE, Dolin R, Blaser MK, eds. Principles and Practice of Infectious Diseases. 8<sup>th</sup> ed. Philadelphia". *Pa: Elsevier Churchill Livingstone* (2015): 3173-3183.
2. Checkley W., *et al.* "A review of the global burden, novel diagnostics, therapeutics, and vaccine targets for cryptosporidium". *Lancet Infectious Diseases* 15.1 (2015): 85-94.
3. Ryan., *et al.* "Cryptosporidium species in humans and animals: current understanding and research needs". *Parasitology* 141.13 (2014): 1667-1685.
4. Xiao. "Molecular epidemiology of cryptosporidiosis: an update". *Experimental Parasitology* 124.1 (2010): 80-89.
5. Zahedi., *et al.* "Public health significance of zoonotic *Cryptosporidium* species in wildlife: Critical insights into better drinking water management". *International Journal for Parasitology: Parasites and Wildlife* 5.1 (2016): 88-109.
6. Peletz., *et al.* "Preventing cryptosporidiosis: the need for safe drinking water". *Bulletin of the World Health Organization* 91.4 (2013): 238.

7. Tandukar, *et al.* "Intestinal parasitosis in school children of Lalitpur district of Nepal". *BMC Research Notes* 6 (2013): 449.
8. Bhandari, *et al.* "Cyclospora Infection among School Children in Kathmandu, Nepal: Prevalence and Associated Risk Factors". *Tropical Medicine and Health* 43.4 (2015): 211-216.
9. Ghimire, *et al.* "Enteric parasitic infection among HIV-infected patients visiting Tribhuvan University Teaching Hospital, Nepal". *BMC Res Notes* 9 (2016): 204.
10. Adhikari N., *et al.* "Intestinal parasitic infections among HIV seropositive and high risk group subjects for HIV infection in Nepal". *Nepal Medical College Journal* 8.3 (2006): 166-170.
11. Sapkota D., *et al.* "Enteric parasitosis with HIV-infection and AIDS in Nepal". *Journal of Nepal Health Research Council* 2 (2004): 9-13.
12. Rai SK., *et al.* "Status of intestinal parasitosis at TU Teaching Hospital". *Institute of Medicine, Nepal* 17 (1993): 134-142.
13. Dhakal DN., *et al.* "Cryptosporidium parvum: An observational study in Kanti Children Hospital, Kathmandu Nepal". *Journal of Nepal Health Research Council* 2.1 (2004): 25-29.
14. Filipe Anibal Carvalho-Costa., *et al.* "Detection of Cryptosporidium spp and other intestinal parasites in children with acute diarrhea and severe dehydration in Rio de Janeiro". *Revista da Sociedade Brasileira de Medicina Tropical* 40.3 (2007): 346-348.
15. Bern C., *et al.* "The contrasting epidemiology Of Cyclospora and Cryptosporidium among outpatients in Guatemala". *American Journal of Tropical Medicine and Hygiene* 63.5-6 (2000): 231-235.
16. Nimri LF. "Cyclospora cayetanensis and other intestinal parasites associated with diarrhea". *International Microbiology* 6.2 (2003): 131-135.
17. Easow JM., *et al.* "Emerging opportunistic protozoa and intestinal pathogenic protozoal infestation profile in children of western Nepal". *Nepal Medical College Journal* 7.2 (2005): 134-137.
18. Laubach HE., *et al.* "A Study of Risk Factors Associated With the Prevalence of Cryptosporidium in Villages Around Lake Atitlan, Guatemala". *The Brazilian Journal of Infectious Diseases* 8.4 (2004): 319-323.
19. Alles AJ., *et al.* "Prospective comparison of direct immunofluorescence and conventional staining methods for detection of Giardia and Cryptosporidium spp. in human fecal specimens". *Journal of Clinical Microbiology* 33.6 (1995): 1632-1634.
20. Mahgoub E.S., *et al.* "Cryptosporidiosis in children in a north Jordanian paediatric hospital". *Eastern Mediterranean Health Journal* 10.4-5 (2004) 494-501.
21. Latif B and Rossle NF. "Cryptosporidiosis among children with diarrhoea in three Asian Countries: A review". *Asian Pacific Journal of Tropical Medicine* 5.11 (2015): 885-888.
22. Munoz-Antoli C., *et al.* "Prevalence and molecular characterization of Cryptosporidium in school children from department of Rio San Juan (Nicaragua)". *Journal of Tropical Biomedicine* 28.1 (2011): 40-47.
23. Huang, *et al.* "Dietary Sodium Butyrate Decreases Postweaning Diarrhea by Modulating Intestinal Permeability and Changing the Bacterial Communities in Waned Piglets". *Journal of Nutrition* 145.12 (2015): 2774-2780.
24. Wang, *et al.* "Advance of interactions between exogenous natural bioactive peptides and intestinal barrier and immune responses". *Current Protein and Peptide Science* 16.7 (2015): 574-575.
25. Fan, *et al.* "Metabolites of dietary protein and peptides by intestinal microbes and their impacts on gut". *Current Protein and Peptide Science* 16.7 (2015): 646-654.

26. Chen., *et al.* "Interaction between microbes and host intestinal health: modulation by dietary nutrients and gut-brain-endocrine immune axis". *Current Protein and Peptide Science* 16.7 (2015): 592-603.
27. Liu., *et al.* "Oral administration of *Lactobacillus fermentum* I5007 favors intestinal development and alters the intestinal microbiota in formula-fed piglets". *Journal of Agricultural and Food Chemistry* 62.4 (2014): 860-866.

**Volume 4 Issue 1 October 2016**

**© All rights reserved by Samendra Sherchan., *et al.***