



Examination of Potential Causes of Gastroenteritis in Children with a Focus on Viruses and Parasites

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Abstract:

Gastroenteritis is a digestive system ailment that can be either acute or chronic. Its symptoms include diarrhea and symptoms of stomach irritation, including nausea, vomiting, and epi-gastric pain. The most typical sign of a lower gastrointestinal tract ailment is diarrhea. Diarrhea occurs when there are more than three bowel motions in a day and the stool is more watery or softer than normal. An increase in the volume, fluidity, or frequency of bowel movements in comparison to a person's typical habits is another definition of diarrhea. Diarrheal disease is a major public health concern for children under five in developing countries such as Nepal. In 2023–2024, there were 194 diarrheal deaths and 787,094 diarrheal visits globally. The incidence of diarrhea in children under five was 222/1000, and the case fatality rate was 0.2/1000. More than 12 bouts of diarrhea occur in children in certain poor countries each year, and diarrheal sickness accounts for 15–34% of all deaths. The purpose of this study was to identify the current state of affairs and additional elements that contribute to the spread of illness in our nation's socio-regional context. Dehydration, dysentery, malnutrition, and various illnesses are the primary causes of mortality from diarrhea. In this investigation, enteropathogens such as bacteria, *Cyclospora cayetanensis*, various parasites, and rotavirus were found in 201 (52.07%) of the diarrheal patients and 45 (39.47%) of the non-diarrheal cases. Massive micro biota are linked to gastroenteritis. They include bacteria like *Shigella*, enteropathogenic *E. coli*, *Salmonella*, *Vibrio cholera*, and others, as well as parasites like *Entamoeba histolytica* and *Giardia*. Certain bacteria, such as *Campylobacter jejuni*, enteropathogenic *E. coli*, *Helicobacter pylori*, etc., protozoans, such as *Cyclospora cayetanensis* and *Cryptosporidium parvum*, and other diarrheal viruses, such as rotavirus, Adeno virus, Norwalk agents, etc., have just recently been identified in the previous few decades. It is evident from the data that the Kathmandu valley and its surroundings are home to several entero pathogenic bacteria and parasites, including *Cyclospora*. It has been discovered that these infections are transmitted by food and water. The feco-oral pathway is how certain gastrointestinal disorders are spread. By putting into practice straightforward tactics like appropriate waste and water management and education on maintaining sanitary environments, the prevalence of gastrointestinal sickness may be considerably reduced.

Key words: gastroenteritis; diarrhea; fatality; dehydration; malnutrition

Introduction:

An illness of the digestive system, gastroenteritis can be either acute or persistent. Diarrhea and signs of stomach irritation, such as nausea, vomiting, and epi-gastric discomfort, are among its symptoms. The most typical sign of a lower gastrointestinal tract ailment is diarrhea (Babaei, Rafiee et al. 2022). Diarrhea occurs when there are more than three bowel motions per day and the stool is softer or more liquid than normal. An increase in the volume, fluidity, or frequency of bowel movements in comparison to an individual's typical habits is another definition of diarrhea (Florez, Nino-Serna et al. 2020). The three clinical syndromes of diarrhea that are differentiated by pathogenicity are acute watery diarrhea (lasting less than 14 days), dysentery (feces with blood and mucus), and persistent diarrhea (lasting at least 14 days) (Black, Perin et al. 2024). One of the main public health issues affecting children under five in poor nations like Nepal is diarrheal illness. There were 787,094 diarrheal visits and 194 diarrheal fatalities worldwide in 2023–2024. Children under five had a 222/1000 population incidence of diarrhea and a 0.2/1000 population case fatality rate (Ghosh, Chakraborty et al. 2021). Children in certain impoverished nations experience more than 12 episodes of diarrhea annually, and diarrheal illness is responsible for 15–34% of all fatalities. Massive micro biota is linked to gastroenteritis. They include bacteria like *Shigella*, enteropathogenic *E. coli*, *Salmonella*, *Vibrio cholera*, and

others, as well as parasites like *Entamoeba histolytica* and *Giardia*. Certain bacteria, such as *Campylobacter jejuni*, enteropathogenic *E. coli*, *Helicobacter pylori*, etc., protozoans, such as *Cyclospora cayetanensis* and *Cryptosporidium parvum*, and other diarrheal viruses, such as rotavirus, Adeno virus, Norwalk agents, etc., have just recently been identified in the previous few decades (Savarino, Zingone et al. 2022). According to conservative estimates, diarrheal illness claims 4 to 6 million lives annually (Khan and Amin 2024), with young children accounting for the majority of these deaths (Levine, Nasrin et al. 2020). Accurate surveillance and diagnosis are made more difficult by the variety of bacterial and viral illnesses that can cause diarrhea, particularly when access to contemporary laboratory techniques is limited or nonexistent (Smalley, Falck-Ytter et al. 2019). The first two years of life account for around 60% of diarrheal fatalities. Dehydration, dysentery, malnutrition, and various illnesses are the primary causes of mortality from diarrhea (Jain, Shah et al. 2020).

Particulars	Amoebic dysentery	Bacillary dysentery
Macroscopic		
Number	6-8 motions per day	Over 10 motions per day
Amount	Relatively copious	Small
Odour	Offensive	Odourless
Color	Dark red	Bright red
Nature	Blood and mucus mixed with feces	Blood and mucus; No feces
Reaction	Acid	Alkaline
Consistency	No adherent to the container	Adherent to the bottom of the container
Microscopic		
R.B.C	In clumps; reddish-yellow in color	Discrete or in rouleaux; bright red in color
Pus cells	Scanty	Numerous
Macrophages	Very few	Large and numerous; many of them contain R.B.C; hence mistaken for <i>E.histolytica</i>
Eosinophil	Present	Scarce
Pyknotic bodies	Very common	Nil
Ghost cells	Nil	Numerous
Parasite	Trophozoites of <i>E.histolytica</i>	Nil
Bacteria	Many motile bacteria	Nil
Charcot-Layden Crystals	Present	Nil

Table 1: Differences between Amoebic and Bacillary dysentery (Ifeoma and Kenneth 2024)

The purpose of this study was to identify the current state of affairs and additional elements that contribute to the spread of illness in our nation's socio-regional context. Additionally, it was anticipated that the current study on etiological agents would establish certain facts in one hand and be helpful for future research in this area.

Objectives:

General Goal

- To access potential causes of gastroenteritis in children, paying particular emphasis to viruses and parasites.

Specific Goal

- To find the prevalence of enteropathogens in children
- To assess the relationship between the prevalence of enteropathogens with age and gender.
- To determine the incidence of *Cyclospora cayetanensis* in relation to the patient's age, gender, month, and water-drinking habits.

Materials and Methods:

Between May and September of 2024, this study was conducted at T.U. IOM's Health Research Laboratory in Kathmandu. Children from Kanti Children's Hospital under the age of five who had gastroenteritis had their stool samples taken. A grease-free, wide-mouthed container with an airtight top was used to collect stool samples from children under five who were visiting Kanti Children's Hospital's oral rehydration therapy (ORT) and outpatient department (OPD) wards. Parents of the patients were interviewed side by side. To ascertain whether the disease was diarrheal or not, the doctor's prescription was closely monitored during sample collection. The stool samples were then promptly transported to the lab. Standard laboratory procedures were followed in the processing of the obtained stool samples (Camilleri 2025). The data was analyzed using descriptive statistics, such as the Chi square test, to demonstrate the relationship between enteropathogen infection and risk variables.

The existence of adult parasite larvae or blood mucus in the stool samples was investigated. Additionally, the feces samples' color and consistency were noted macroscopically. The cyst, eggs, or oocyst of the parasites were examined by direct microscopy of slide preparation. Observing the RBC, pus cells, and WBC in feces was also beneficial. Wet mount saline preparation and the iodine preparation method were used to identify oocyst, cysts, and trophozoites of protozoa, ova, or helminthes eggs at low power 10x magnification. Additionally, each sample was stained with Ziehl Neelsen to examine *Cyclospora* and *Cryptosporidium*.

Appearance	Possible Pathogens
Unformed containing pus and mucus mixed with pus	Shigellosis; Campylobacter enteritis
Unformed with blood and mucus (acid pH)	Amoebic dysentery
Unformed or semi formed often with blood and mucus	Schistosomiasis
Watery stool	ETEC infection, Rota virus enteritis
Rice watery stools with mucous flakes	Cholera
Unformed or watery and sometimes with blood, mucus and pus	Salmonellosis
Unformed, pale colored, frothy, unpleasant smelling stools that float on water (high fat content)	Giardiasis, other conditions that cause malabsorption e.g. Post infective tropical malabsorption
Fluid stools (containing lactose with pH below 6)	Lactase deficiency
Unformed or semi formed black stools (positive occult blood test)	Melena (gastrointestinal bleeding), Hookworm diseases, Iron therapy

Table 2: Clue for the identification of enteropathogens (Paruch and Paruch 2022)

Registration	Diarrheal cases		Non-Diarrheal cases	
	Frequency	%	Frequency	%
In patient	250	64.8	---	---
Out-patient	136	35.2	114	100.0
Total	386	100.0	114	100.0

Table 3: Frequency distribution according to hospital registration

According to the table, 250 (64.8%) of the 500 diarrheal children who were enrolled in the study were admitted to the ORT ward and 136 (35.2%) were seen in the outpatient department.

The research comprised 114 (100%) OPD patients who did not have diarrhea.

Age group in months	Diarrheal cases		Non-diarrheal cases		Total
	Frequency	%	Frequency	%	
0 – 6	40	10.4	12	10.5	52
7 – 24	190	49.2	52	45.6	242
25 –60	156	40.4	50	43.9	206
Total	386	100.0	114	100.0	500

Table 4: Age wise distribution of diarrheal and non-diarrheal cases

According to the table, of the 386 diarrheal children, 40 (10.4%) were in the 0–6 month age group, 190 (49.2%) were in the 7–24 month age group, and 156 (40.4%) were in the 25–60 month age group. Similarly, among the 114 children who were not diarrheal, 12 (10.5%) instances were from the 0–6 month age group, 52 (45.6%) from the 7–24 age group, and 50 (43.9%) from the 25–60 age group.

Gender	Diarrheal cases		Non –diarrheal cases	
	Frequency	%	Frequency	%
Male	245	63.5	66	57.9
Female	141	36.5	48	42.1
Total	386	100	114	100

Table 5: Gender-wise distribution of diarrheal and non-diarrheal cases

According to the table, of the 386 diarrheal patients who were recruited in the research, 245 (63.5%) were male and 141 (36.5%) were female. Of the 114 non-diarrheal patients, 66 (57.9%) were male and 48 (42.1%) were female

Frequency of stool/day	Frequency	%
3-5	167	43.3
5-10	129	33.4
>10	90	23.3
Total	386	100.0

Table 6: Frequency distribution based on number of stool/day in diarrheal cases

According to the table, of the 386 patients who were recruited in the research, 167 (43.3%) had stools three to five times a day, 129 (33.4%) had stools five to ten times a day, and 90 (23.3%) had stools more than ten times a day.

Stool type	Frequency				Total
	Yes	%	No	%	
Watery	266	68.9	120	31.0	386
Mucus	211	54.6	175	45.3	386
Blood	5	1.3	381	98.7	386

Table 7: Frequency distribution based on consistency of stool in diarrheal cases

According to the table, of the 386 cases of diarrhea, 266 (68.9%) had watery stool, 211 (54.6%) had mucus, and 5 (1.3%) had blood. In contrast, 120 (31.0%), 175 (45.3%), and 381 (98.7%) had no blood, mucus, or watery stool, respectively.

Intestinal parasites	Diarrheal cases		Non-diarrheal cases	
	Frequency	%	Frequency	%
Positive	119	30.8	30	26.3
Negative	267	69.2	84	73.7
Total	386	100.0	114	100.0

Table 8: Distribution of intestinal parasites in diarrheal and non-diarrheal cases

According to the table, 119 (30.8%) of the 386 total diarrheal stool samples tested positive for intestinal parasites, whereas 267 (69.2%) tested negative. Similarly, 30 (26.3%) of the 114 non-diarrheal stool samples tested positive for intestinal parasites, whereas 84 (73.7%) tested negative. These results were statistically inconsequential at the 5% level of significance, $p > 0.05$.

Enteropathogens	Diarrheal cases		Non-diarrheal cases	
	Frequency	% (n=386)	Frequency	% (n=114)
Bacteria	13	3.36	1	0.87
Shigella spp.	0	0.00	0	0.00
Salmonella spp.	1	0.25	0	0.00
Vibrio spp.	12	3.1	1	0.87
Protozoans	112	29.01	33	28.94
Entamoeba histolytica	36	9.32	8	7.01
Giardia lamblia	32	8.29	12	10.52
Cyclospora cayetanensis	30	7.77	7	6.14
Cryptosporidium parvum	3	0.77	1	0.87
Entamoeba coli	4	1.04	1	0.87
Blastocystis hominis	7	1.81	4	3.50
Helminthes	76	2.84	6	5.26
Ascaris lumbricoides	4	1.04	2	1.75
Trichuris trichuria	2	0.51	2	1.75
Hymenolepsis nana	4	1.04	1	0.87
Taenia spp.	1	0.03	1	0.87
Rotavirus	65	1.68	5	4.38

Table 9: Distribution of enteropathogens among total cases

According to the table, of all the diarrheal stool samples (n = 386), the most common bacteria were Vibrio spp. (number 12, 3.1%), followed by Salmonella spp. (number 1, 0.25%), and Shigella spp.

Protozoans 112 (29.01%) outnumbered helminthes 11 (2.84%) among parasites. Entamoeba histolytica ranked highest among protozoans at number 36 (9.32%), followed by Giardia lamblia at number 32 (8.29%), Cyclospora cayetanensis at number 30 (7.77%), Blastocystis hominis at number 7 (1.81%), Entamoeba coli at number 4 (1.04%), and Cryptosporidium parvum at number 3 (0.77%).

Ascaris lumbricoides 4 (1.04%), Hymenolepsis nana 4 (1.04%), Trichuris trichuria 2 (0.51%), and Taenia spp.1 (0.03%) are among the helminthes.

The only bacterial pathogen found in all of the non-diarrheal samples (n = 114) was Vibrio spp. 1 (0.87%). Protozoans 33 (28.94%) outnumbered helminthes 6 (5.26%) among parasites. Giardia lamblia ranked highest among protozoans at number 12 (10.52%), followed by Entamoeba histolytica at 8 (7.01%), Cyclospora cayetanensis at 7 (6.14%), Blastocystis hominis at 4 (3.50%), Entamoeba coli at 1 (0.87%), and Cryptosporidium parvum at 1 (0.87%).

Hymenolepsis nana 1 (0.87%), Ascaris lumbricoides 2 (1.75%), Trichuris trichuria 2 (1.75%), and Taenia spp. 1 (0.87%) are helminthes.

Age in month	Diarrheal cases			Non-diarrheal cases		
	Frequency	<i>Cyclospora</i> positive	%	Frequency	<i>Cyclospora</i> positive	%
0-6	40	5	12.5	12	0	0.00
7-24	190	12	6.31	52	1	1.92
25-60	156	13	8.33	50	6	12.00
Total	386	30	7.77	114	7	6.14

Table 10: Age wise distribution of *Cyclospora cayetanensis*

The age-wise comparison of *Cyclospora cayetanensis* infection in patients with and without diarrhea is displayed in the table. 5(12.5%) of the forty diarrheal cases in the 0–6 month age group tested positive for *C. cayetanensis*, while none of the twelve non-diarrheal cases tested positive. 12 (6.31%) of the 190 diarrheal cases in the 7–24 month age group tested positive for *C. cayetanensis*, whereas 1 (1.92%) of the 52 non-diarrheal cases tested positive. In the 25–60 month age range, 13 (8.33%) of the 156 patients with diarrhea tested positive for *C. cayetanensis*, whereas 6 (12.00%) of the 50 cases without diarrhea tested positive, which was judged to be statistically insignificant ($p>0.05$).

Sex	Diarrheal cases			Non-diarrheal cases		
	Frequency	<i>Cyclospora</i> positive	%	Frequency	<i>Cyclospora</i> positive	%
Male	245	22	8.97	66	4	6.06
Female	141	8	5.67	48	3	6.25
Total	386	30	7.77	114	7	6.14

Table 11: Sex wise distribution of *Cyclospora cayetanensis*

Cyclospora cayetanensis infections in diarrheal and non-diarrheal patients are compared by sex in the table. Out of 245 male patients with diarrhea, 22 (8.97%) tested positive for *C. cayetanensis*, whereas 4 (6.06%) of the 66 cases without diarrhea tested positive. 8 (5.67%) of the females in 141 diarrheal cases tested positive for *C. cayetanensis*, whereas 3 (6.25%) of the females in 48 non-diarrheal cases tested positive, which was deemed to be statistically insignificant ($p>0.05$).

Source of drinking water	Diarrheal cases			Non-diarrheal cases		
	Frequency	<i>Cyclospora</i> positive	%	Frequency	<i>Cyclospora</i> positive	%
Tap	290	25	8.62	62	4	6.45
Well	9	0	0.00	3	0	0.0
Boiled	52	4	7.69	36	2	5.55
Filtered	35	1	2.85	13	1	7.69
Total	386	30	7.77	114	7	6.14

Table 12: Distribution of *Cyclospora cayetanensis* according to source of drinking water

The distribution of *Cyclospora cayetanensis* by drinking water source in both diarrheal and non-diarrheal cases is displayed in the table. Out of 290 patients who used tap water, 25 (8.62%) tested positive for *C. cayetanensis*; in contrast, 4 (6.45%) of the 62 patients who did not have diarrhea tested positive. None of the nine diarrheal cases among patients drinking well water tested positive, whereas none of the three non-diarrheal cases tested positive. Four (7.69%) of the 52 diarrheal patients who used boiling water tested positive, whereas two (5.55%) of the 36 non-diarrheal patients tested positive. Similarly, among patients who used filtered water, 1 (2.85%) of the 35 diarrheal cases tested positive for *C. cayetanensis*, and 1 (7.69%) of the 13 non-diarrheal cases tested positive.

Symptoms	Yes			No		
	Frequency	<i>Cyclospora</i> positive	%	Frequency	<i>Cyclospora</i> positive	%
Fever	196	16	8.16	190	15	7.89
Abdominal pain	302	27	8.94	84	5	5.95
Vomiting	201	19	9.45	185	13	7.02
Nausea	195	19	9.74	191	13	6.80

Table 13: Distribution of *Cyclospora cayetanensis* according to clinical symptoms

The distribution of *Cyclospora cayetanensis* based on clinical signs is displayed in the table. 16 (8.16%) of the 196 individuals with fever were positive for *C. cayetanensis*, compared to 15 (7.89%) of the 190 cases without fever. Of the 302 individuals with stomach discomfort, 27 (8.94%) had *C. cayetanensis*; of the 84 cases without abdominal pain, 5 (5.95%) had *C. cayetanensis*. Thirteen (7.02%) of the 185 patients without vomiting tested positive for *C. cayetanensis*, compared to 19 (9.45%) of the 201 cases with vomiting. Out of 195 patients with nausea, 19 (9.74%) tested positive for *C. cayetanensis*; in contrast, out of 191 cases without nausea, 13 (6.80%) tested positive for *C. cayetanensis*; this difference was statistically significant, $p < 0.05$.

Conclusions:

One of the leading causes of childhood mortality, diarrhea continues to be a major public health issue in underdeveloped nations, particularly among children under five. In this investigation, enteropathogens such as bacteria, *Cyclospora cayetanensis*, various parasites, and rotavirus were found in 201 (52.07%) of the diarrheal patients and 45 (39.47%) of the non-diarrheal cases. These results were consistent with research conducted in Saudi Arabia by Sheikh and Assouli in 2021 (Alnezary, Alamri et al. 2024) (47.6%), Jordan by Youssuf et al., 2020 (66.4%) (Al-khlifeh and Hassanat 2023), India by and Howard et al., 2020 (39%).

It is evident from the data that the Kathmandu valley and its surroundings are home to several entero pathogenic bacteria and parasites, including *Cyclospora*. It has been discovered that these infections are transmitted by food and water. The feco-oral pathway is how certain gastrointestinal disorders are spread. By putting into practice straightforward tactics like appropriate waste and water management and education on maintaining sanitary environments, the prevalence of gastrointestinal sickness may be considerably reduced.

Ethics approval

We author hereby declare you that the article entitled examination of potential causes of gastroenteritis in children with a focus on viruses and parasites has no harm to anyone while preparing the manuscript.

Consent for Publication: Not applicable.

Data Availability: Data from Kanti Children Hospital for diarrheal and non-diarrheal cases were taken into consideration as a reference.

Competing Interest:

We declare that an article entitled Examination of potential causes of gastroenteritis in children with a focus on viruses and parasites has no competing interest among the authors.

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Authors Contribution:

- R. Kandel conceived and designed the study, led the research implementation, performed data analysis, and was the primary author of the manuscript. P. Bhattarai and P. Sharma coordinated all stages of the project and ensured the scientific integrity of the work. U. Ghimire contributed to field data collection, assisted in sample analysis, and supported the interpretation of results. She also participated in manuscript review and editing. R. Kandel provided guidance on methodology, assisted in laboratory procedures, and contributed to drafting and reviewing and editing the manuscript for intellectual content and supervised the work throughout the research period.

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