



Intestinal Parasitic Infections and Associated Risk Factors Among School-going Children of Age 1-5 years in Dharan, Eastern Nepal

^aDhiren Subba Limbu*, ^aSamana Shrestha, ^aKamana Bantawa, ^bRamesh Majhi, ^cMilan Kharel

^aDepartment of Microbiology, Central Campus of Technology, Tribhuvan University, Dharan, Nepal

^bForum for Health Research and Development (FHRD), Dharan, Nepal

^cDepartment of Zoology, Post Graduate Campus, Biratnagar, Nepal

*Corresponding email: dhiren.limbu@cct.tu.edu.np

Abstract

Intestinal parasitic infections have been a major public health burden of developing countries, especially in children. Higher prevalence has been reported among school children, mostly in rural areas of Nepal where water, toilets, hygiene, and sanitation facilities are insufficient. This cross-sectional study was carried out from April to September 2019 to determine the prevalence of intestinal parasitic infections among school-going children 1-5 years of Dharan, Nepal, and to assess the associated risk factors. Stool samples were collected in a clean, dry, screw-capped, and wide-mouthed plastic container, kept in an icebox, and transported to the laboratory. Data relating to different risk factors were collected from the parents of 116 participants using a structured questionnaire. The parasites were identified by using the direct wet mount method and formal-ether concentration method. Pearson's chi-square test was carried out to establish associations between dependent and independent variables using SPSS version 20, and the test considered a greater than $< 0.05\%$ as statistically significant with a 95% confidence level. Out of the 116 stool samples, 9 (7.75%) tested positive for the parasitic infections in which 5 (55.5%) were protozoa and 4 (44.45%) were helminths. The prevalent parasites, were *Entamoeba histolytica* (23%), *Hyamenolepis nana* (22%), *Giardia lamblia* (11%), Hookworm (11%), *Entamoeba coli* (11%), *Ascaris lumbricoides* (11%), and Intestinal parasitic infections had a significant association with drinking water, bowel syndrome, bathing habit, toilet facility, and washing hands after toilets ($p < 0.05$). Lack of toilets, poor hygiene, and unsafe drinking water were the main risk factors. Improved hygienic practices, safe drinking water, and the use of latrines could lower the rate of parasitic infections.

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1. Introduction

Parasitic infections have been with humankind since time immemorial (Okuy et al., 2004). They are classified as protozoa, helminths, and ectoparasites. They live and acquire their nourishment from the host (Haque, 2007). The parasitic infection is the major cause of human affliction and death globally (Okuy et al., 2004). An estimated 3.5 billion people are affected, and 450 million are ill as a result of these infections. People of all age groups and gender are affected by parasitosis, the majority being children (Upama et al., 2019).

They cause iron deficiency anemia with a high death rate in pregnant women and children, growth retardation in children, and other physical as well as mental health problems (Okuy et al., 2004; Upama et al., 2019). Like other developing nations, intestinal parasite infections (IPIs) are major health problems in Nepal (Shrestha et al., 2021). It is important to carry out epidemiological surveys on the IPIs to understand the sanitary conditions of the community and to formulate basic data for controlling parasitic infections in the future (Curtale et al., 1998). The World Health Organization estimated that approximately 270 million pre-school children and more than 600 million school-

going children live in those areas where parasites are common and need instant therapy, prevention, and control (Brooker et al., 2010).

The prevalence of IPIs reaches nearly 100% in some tropical areas of the world (Sharma et al., 2004). *Ascaris lumbricoides*, *Giardia lamblia*, *Entamoeba histolytica*, *Trichuris trichiura*, *Hymenolepis nana*, *Entamoeba coli*, and Hookworms are the most dominant intestinal parasites worldwide (Ashok et al., 2013). Intestinal worm infections rank fourth among the top ten diseases in Nepal (Jaiswal et al., 2014). Giardiasis, ascariasis, amoebiasis, ancylostomiasis, and taeniasis are common IPIs in Nepal (Jaiswal et al., 2014).

According to some previous studies, the prevalence of IPIs has been ranging from low to a hundred percent in Nepal (Rai, 1986, 2002). Among all global diseases, 12% of the disease outbreak is caused by intestinal parasites and is observed more in children under 14 years in developing countries (Hailegebriel, 2017).

Due to the lack of proper toilets and improper handwashing after defecation and before eating, the transmission of parasites is increasing (Pooja et al., 2014). In Nepal, one of the most important reasons behind the higher prevalence of parasitosis is open excretion. Poor hygiene, overpopulation, contaminated drinking water, poor sanitation, illiteracy, lack of awareness, contaminated food, farming occupation, socioeconomic condition, and cultural practices are considered to be important lagging factors responsible for the increasing rate of parasitic diseases in Nepal (Bertoncello et al., 2017; Dhakal & Subedi, 2019; Gupta et al., 2020; Yadav & Prakash, 2016).

Little studies are conducted for determining the prevalence of IPIs and assessing the associated risk factors among children in Dharan, Nepal. Hence, this study was designed to cope with the knowledge gap on the prevalence and associated risk factors of IPIs among school-going children of age group 1-5 years in Dharan, Nepal.

2. Materials and Method

Study type, location, population, and sample size determination

A school-based cross-sectional study was conducted from April to September 2019 in Dharan,

Province number 1, Nepal. The study population was school-going children of age group 1-5 years enrolled in 14 schools of Dharan, Nepal.

The sample size was calculated using a single population proportion formula for cross-sectional surveys ($n = z^2 pq / e^2$) where, n = sample size, z = level of confidence, p = estimated proportion/prevalence of the population, $q = 1 - p$ and e = tolerated margin of error. So, assuming a 95% level of confidence, 5% margin of error, taking prevalence of 8.17% from the study conducted in eastern Nepal (Baral et al., 2017) sample size was 116 $[(1.96)^2 \cdot 0.0817(1 - 0.0817) / (0.05)^2 = 115.28 \sim 116]$. Children who started anti-parasitic drug/s for the last 15 days and those who had fever were excluded from the study. Study participants were selected using convenient sampling and every participant and their guardian were interviewed.

Sample collection, processing, and quality control

About 2 grams of fresh stool samples were collected in the clean, dry, screw-capped, and wide-mouthed plastic containers given with applicator sticks at home. The collected samples were transported to the laboratory maintaining a cold chain. The children and guardians were instructed properly for the collection of the samples. During sample collection, the date, name, age, and gender of participants were noted. The samples were mixed in 10% formal saline and transported to the laboratory and processed on the same day. Samples that were not to be processed on the same day were preserved in the refrigerator. All the stool samples were examined microscopically using direct wet-mount and formal-ether concentration methods as mentioned in WHO guidelines (Isenberg, 1998; Kibru & Mekete, 2000; Organization, 1991). All the developmental stages exhibited by the parasites (egg, cyst, larvae, and adult) were registered. The slides were observed by more than one observer and consultation with experts (medical lab technologists) was taken to ensure the developmental stages of parasites. Laboratory equipment like a microscope, centrifuge, staining reagents, sample collection containers, and icebox were checked before use to ensure the correct functioning. Each sample was double-checked for correct labeling. To avoid examiner bias, each specimen was observed independently by two examiners and their results were compared for quality control of microscopy.

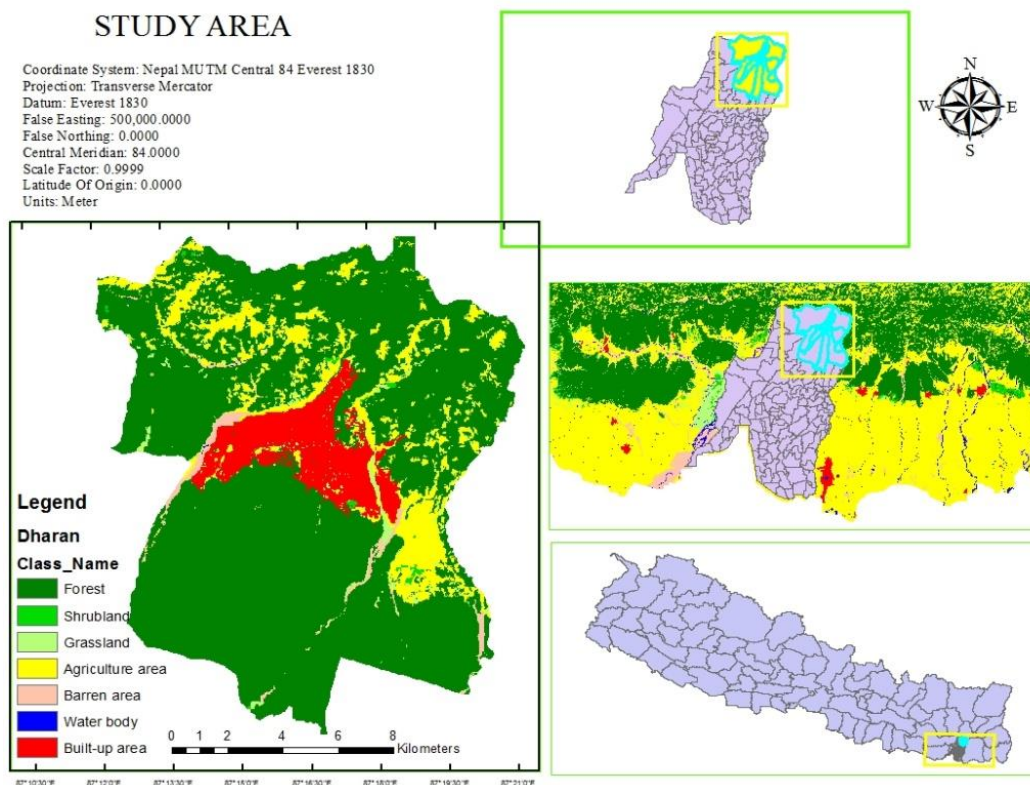


Figure 1: GIS map of the study site

Data collection and analysis

The structured questionnaires regarding the socio-demographic data, behavioral and hygienic practices were prepared by reviewing local and international literature in Nepali and translated into English after the interview. Data collectors were trained for 1 day about the questionnaire and data collection technique by researchers. The data was collected by a face-to-face interview with their parents/guardians at home by using a structured questionnaire. The collected data were tabulated with Microsoft Office Excel 2007 and analyzed by using Statistical Package for Social Science (SPSS) software version 20. Chi-square (χ^2) test was done to determine the association between the prevalence of (IPIs) and associated risk factors. At a 5% level of significance, the association was considered significant if $p < 0.05$.

3. Results and Discussion

Socio-demographic characteristics of the study participants

A total of 116 school children of age 1-5 years from the government as well as private schools were the study population consisting of 64 (55.17%) male and 52 (44.82%) female children (Table 1). All of the

questionnaires on associated risk factors were filled by data collectors. More than half of students' mothers were illiterate housewives and only about 68% of students' parents were literate. Furthermore, 83% of participants came from family sizes of 4 and above. Among the participants, only 70.68% were taking baths daily and 93.10% were using toilets, the rest of them were using open fields for defecation. After defecation, only 74.13% of participants wash their hands with soap and water.

Prevalence of Parasites

The sample containing any forms of parasites like cyst, egg, and ova was considered as positive for IPIs otherwise negative. Altogether 9 samples were found to be positive for any kind of IPIs. Out of 9 positive samples, 5 (54%) parasites were protozoa and 4 (46%) parasites were helminths. It shows that the presence of protozoan parasites was higher than helminths. Among the protozoans, 2 (22.22%) were *E. histolytica*, 2 (22.22%) were *G. lamblia*, and 1 (11.11%) were *Entamoeba coli*. Similarly, among the helminths, 2 (22%) *H. nana*, 1 (11.11%) *A. lumbricoides*, and 1 (11.11%) Hookworms were identified (Figure 1). Different forms of parasites like cyst, egg, and ova were detected in stool samples. Among the positive

cases, none of them was multi-parasitic (with more than one parasite) in this study.

Table 1: Socio-demographic characteristics and sanitation habits of the study participants

Characteristics	No. of participants (%)	Parasite		p-value
		Positive	Negative	
Gender				
Male	64 (55.17)	6	58	0.47
Female	52 (44.82)	3	49	
Residence				
Rented	49 (42.25)	3	46	0.573
Owned	67 (57.75)	6	61	
Family type				
Nuclear	88 (75.87)	8	80	0.342
Joint	28(24.13)	1	27	
Income (NPR/month)				
> 10,000	70 (60.34)	5	65	0.760
< 10,000	46 (39.66)	4	42	
Drinking water				
Municipal (Treated)	78 (67.24)	3	75	0.024
Non-municipal (Untreated)	38 (32.76)	6	32	
Bowel syndrome (Diarrhea and abdominal cramp)				
Yes	30 (25.87)	7	23	0.00
No	86 (74.13)	2	84	
Bathing habit				
Irregular	34 (29.14)	7	27	0.01
Regular (Daily)	82(70.68)	2	80	
Toilet facility				
Open Fields	8 (6.90)	4	4	0.00
Toilet	108 (93.10)	5	103	
Washing hands after toilets				
Soap and water	86 (74.13)	3	83	0.004
Water	30 (25.86)	6	24	

3.3 Association of socio-demographic factors with intestinal parasite infections

The prevalence of IPIs in school-going children of age up to 5 years was a dependent variable whereas different risk factors were independent variables. In this study, significant relationships were observed between IPIs and in some of the risk factors (Table 1). Of those positive for IPIs, no statistically significant relationships were observed between IPIs and residence ($p= 0.573$), family type ($p= 0.342$) and income ($p= 0.76$). However, significant associations were seen between IPIs and type of drinking water ($p= 0.024$), bowel syndrome ($p= 0.00$), bathing habit ($p= 0.01$), toilet facility ($p= 0.00$) and handwashing habit

after toilet ($p= 0.004$). The IPIs were found equally in males and females and the result was statistically insignificant ($p= 0.47$).

Participants who had an irregular bathing habit had a higher risk to be infected by IPIs than those who had regular bathing habits with a statistically significant association ($p= 0.01$). A higher number of IPIs was found in tap water users (15.78%) than municipal tap water users (3.84%). Participants who do not have a toilet facility were more likely to be infected with IPIs (50%) whereas only 4.63% of participants having toilet facilities had IPIs. The prevalence of IPIs was higher in children who live in an owned house (8.95%) than those who live in a rented house (6.12%).

Parasites

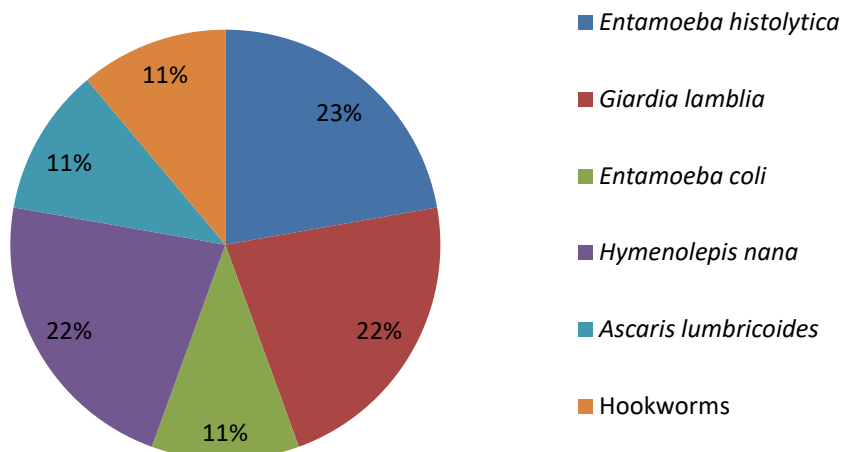


Figure 2: Prevalence of parasites

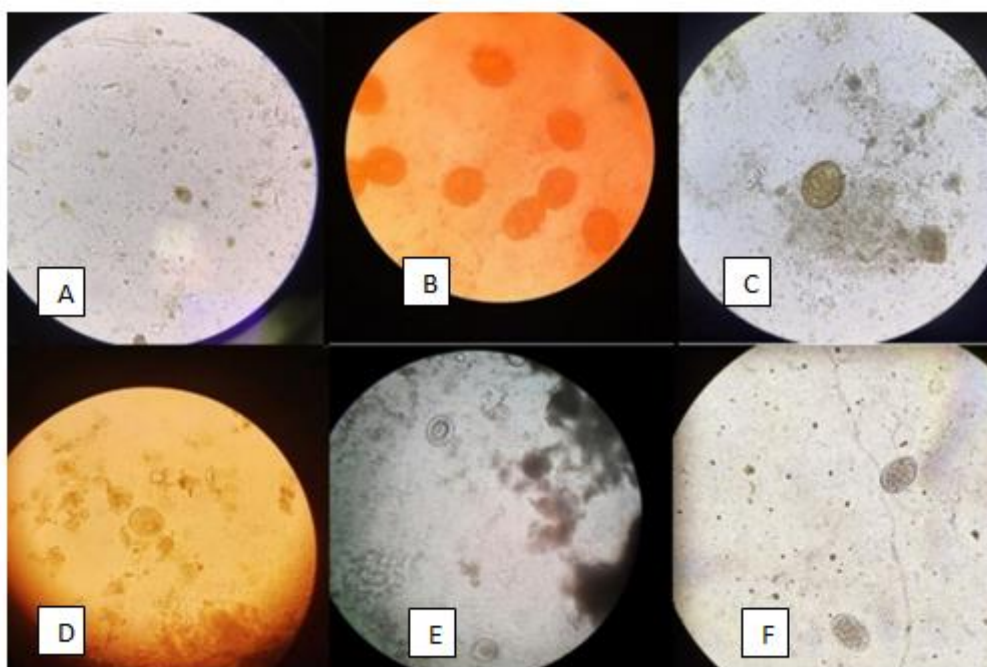


Figure 3: Microscopic observation of intestinal parasites: A. Cysts of *Giardia lamblia* B. Cyst of *Entamoeba coli* C. Cyst of *Entamoeba histolytica* D. Egg of *Hymenolepis nana* E. Egg of *Hymenolepis nana* F. Egg of Hookworm

Children who do not wash hands with any soap or detergent after defecation were more likely to have IPIs (20%) as compared to those washing hands with soap (3.48 %). In this study, 23.33% of participants with bowel syndrome tested positive for IPIs with a strong statistically significant association ($p=0.00$) while only 2.33 % of participants without bowel syndrome tested positive for IPIs. The transmission of IPIs depends on the sanitary condition and the socio-demographic status and behavioral factors in the community. Different factors of a community like hygiene, economic status, toilet facility, and drinking water determine the prevalence of IPIs (Abossie & Seid,

2014). In this study, the overall prevalence of IPIs was 7.76% (9/116) which was consistent with the findings of the previous studies done in Dharan, Saudi Arabia, Ethiopia, and Iran (Baral et al., 2017; Chala, 2013; Sah, Paudel, et al., 2013; Zagloul et al., 2011). Our finding is nearly together with a laboratory-based study in tertiary care hospital of the same study area 8.17% (Baral et al., 2017) indicating that the prevalence of IPIs is decreasing. Dharan is a small city in which only a few people work in the farmlands which could be the one reason for the lower prevalence of IPIs. However, this finding is lower than several studies conducted in Dharan, Nepal.

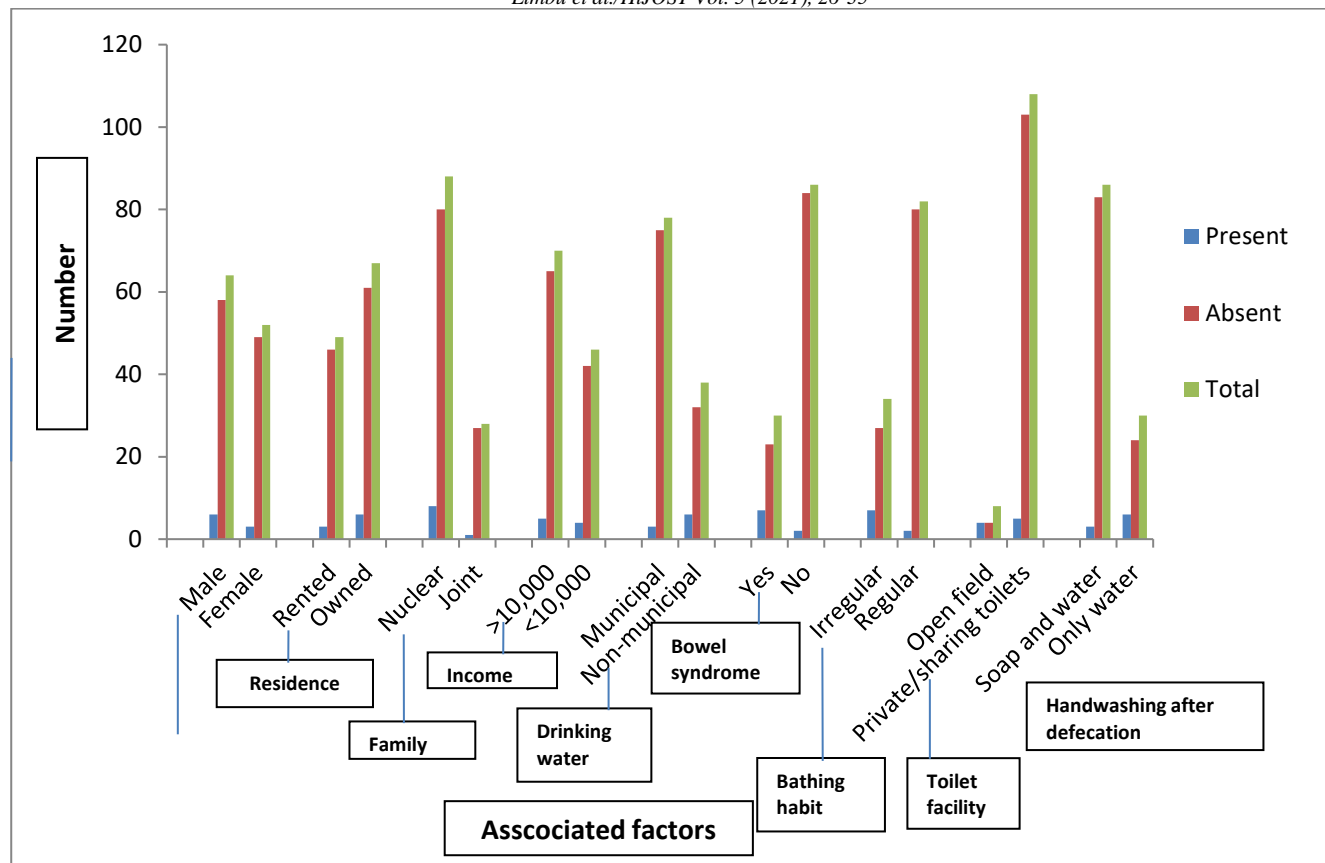


Figure 4: Association of IPIs and associated factors

For instance, the overall prevalence of parasites was, 22.5% in 2010 (Gyawali et al., 2010), 24.4% in 2013 (Sah, Bhattarai, et al., 2013), 41.4% in 2016 (Chongbang et al., 2016), and 32.1% in 2017 (Poudyal et al., 2017). Similarly, IPIs among school-going children of Kathmandu valley were reported higher (Thapa Magar et al., 2011). The reduction in prevalence could be attributed to the awareness program, improved living standards, personal and community hygiene, and regular intake of anti-parasitic drugs which are being distributed by the government especially for children for deworming (Eamsobhana et al., 2005; Kathmandu, 2012; Khanal et al., 2011; Rai, 1986, 2001; Rai et al., 2000). The lower prevalence of IPIs could also be due to the small sample size. There were no positive cases of multiparasitism which denied the findings of a previous study (Thapa Magar et al., 2011). Such disparity might be due to taking antiparasitic drugs regularly as scheduled by the Nepal government.

In this study, the prevalence of protozoan and helminthic parasites were 55.56% (5/9) and 44.44% (8/24) respectively which complies with other studies in Nepal (Pradhan et al., 2014; Tandukar et al., 2013). In contrast, a study in India reported a higher

prevalence of helminths than protozoa (Chandi & Lakhani, 2018). It might be relatable to the dispersal pattern of parasites, change in place, environmental conditions, and diversification of parasites. The most common parasites found in this study were *E. histolytica* (22.22%, 2/9), *G. lamblia* (22.22%, 2/9), and Hookworm (22.22%, 2/9) respectively which was in agreement with a previous study conducted in differently-abled people of Dharan in 2017 (Poudyal et al., 2017) but conflicts with the study of 2010 (Dahal et al., 2018; Gyawali et al., 2010). The similarity in the incidence of parasites could be due to the selection of the same study site, population, and age group. It also showed that the occurrence of intestinal parasites may vary with the time interval.

In a study of Kathmandu, Nepal had reported a 33.3% prevalence of *E. histolytica* (Dahal et al., 2018; Gyawali et al., 2010). A study performed in Ethiopia reported an average of 19.95% (Sitotaw et al., 2019) *G. lamblia* and which was nearly similar to our findings. In contrast, different study reports of Dharan, Nepal had shown 40% (Poudyal et al., 2017) and 11.5% (Gyawali et al., 2010) prevalence of *G. lamblia* which is dissimilar with our findings. In Kathmandu, the overall prevalence of Hookworm was found to be 23.7% (Sharma et al., 2004) which agreed with our

findings but different studies done in Dharan differed from these findings (Chongbang et al., 2016; Gyawali et al., 2010). We found a 1.5% prevalence of *A. lumbricoides*, 2.3% prevalence of *H. nana*, and 11.3% prevalence of *E. coli* which is comparable to a study performed in India (Ashok et al., 2013). The declined prevalence of different parasites indicates the enhancement of sanitation, education, and socio-economic status of Nepalese. The regular deworming program conducted by the government of Nepal might be helping to reduce IPIs in children.

In this study, male participants were found to be more likely to be infected with parasites with a 9.37% (6/64) prevalence than the female with a 5.76% (3/52) prevalence but the difference was not significant ($p=0.47$). The finding was consistent with the other studies in Dharan (Gyawali et al., 2010) and Kathmandu (Sharma et al., 2004), however, incompatible with previous studies done in Dhankuta (Shakya, 2003) and Itahari (Sah, Bhattarai, et al., 2013). It reflects that gender may not play the role in IPIs but may depend on sanitation conditions and hygiene practices. In the present study, participants who wash hands with soap after defecation had a lower prevalence of parasitic infection (3.84%) than those who wash hands only with water (20%), and the difference is statistically significant ($p=0.004$). The result is comparable to previous studies done in Ethiopia (Tadesse, 2005) and India (Bisht et al., 2011; Sah, Bhattarai, et al., 2013). The children who take regular baths had lower IPIs (2.43%) than irregular (20.58%) ones and the difference is significant ($p=0.01$). Washing hands with soap and regular bathing enhance personal hygiene and are maybe the reason for the low prevalence of parasites.

A significant difference between toilet facilities and IPIs was observed in this study ($p=0.00$). A similar finding was observed in the study conducted in Ethiopia (Tadesse, 2005). Lack of toilets and soap may be the main cause of the higher prevalence of parasites. Likewise, a significant association between bowel syndrome and parasitic infection was observed ($p=0.00$) which was comparable to a previous study done in Dharan (Gyawali et al., 2010). It shows that parasites may induce inflammation and pain. This association could be related to the mobile nature and unconsciousness of children which might help in getting pathogenic organisms including intestinal

parasites. In this study, children of slum areas who could not get treated municipal water were also included. The prevalence of parasitic infection was found to be higher in non-municipal water users than untreated non-municipal water users and the difference was statistically significant ($p=0.024$). A similar finding was recorded in a previous study done in Ethiopia (Sitotaw et al., 2019). Water is also considered an important vehicle for transmitting parasites as well as other water-borne pathogens. It showed that treatment of water before distribution is done by the municipality could lower a load of parasites along with other pathogens or remove them.

No significant association between the income of parents and the presence of IPIs was observed in this study ($p=0.76$). It shows that income level was not associated with the prevalence of parasites but other sanitation factors could do. In this study, the prevalence of IPIs in the nuclear and joint family was 9.09% and 8.75% respectively and the difference was not significant ($p=0.342$). The result was consistent with a study conducted in Ethiopia which reported that 42.85% and 40.14% prevalence in nuclear and joint family respectively (Sitotaw et al., 2019). The reason for the lower prevalence in the joint family could be related to the care provided to children by other family members of the joint family. Moreover, no significant difference between the type of residence (rental/owned) and the presence of parasite ($p=0.573$) had shown that type of residence of participants was not associated with the prevalence of IPIs.

4. Conclusion

The presence of IPIs in children indicates that the protozoa and helminths are commonly found in the environment of the study site. Lack of toilets, poor hygiene, and the use of unsafe drinking water is found to be the main risk factors. Improved hygienic practices, safe drinking water, and proper toilet facility could play role in the reduction of parasitic infections. Therefore, a more effective national deworming program along with awareness and good health practices should be employed to lower the burden of IPIs in children.

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Conflicts of Interest

The authors report no conflicts of interest for this work.

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