



## COMPARATIVE DIAGNOSIS OF *Entamoeba histolytica* And *Giardia lamblia* AMONG CHILDREN WITH DIARRHOEA AND THEIR ASSOCIATED RISK FACTORS IN SOME SELECTED HOSPITALS IN ADAMAWA STATE, NIGERIA

\*<sup>1</sup>Kadabiyu G. Jones, <sup>1</sup>Daniel Lucky, <sup>2</sup>Chessed Godly, and <sup>3</sup>Manu J. Maitandu

<sup>1</sup>Department of Public Health, State Ministry of Health, P. M. B. 2078, Yola, Adamawa State, Nigeria.

<sup>2</sup>Department of Public Health, Iconic Open University, P.M.B 02121, Sokoto State, Nigeria.

<sup>3</sup>Department of Zoology, Modibbo Adama University, P. M. B. 2076, Yola, Adamawa State, Nigeria.

<sup>4</sup>Department of Science Laboratory Technology, Modibbo Adama University, P. M. B.

\*Corresponding authors' email: [jonesbuto@gmail.com](mailto:jonesbuto@gmail.com)

### ABSTRACT

A cross-sectional hospital-based study was conducted to determine the prevalence of *intestinal Protozoans and other parasites* among diarrheal children in some selected hospitals from November 2024 to December 2025. Stool and blood samples were obtained and examined microscopically (saline wet mount, formol-ether concentration), and serologically using Enzyme-Linked Immunosorbent Assay (ELISA), to identify the antigens of *G. lamblia* and *E. histolytica*. Questionnaires were also administered to obtain socio-demographic information and risk factors. Data collected were analysed using R-statistical software, chi-square tests, and risk factors were identified through multivariate logistic regression, with statistical significance set at  $p < 0.05$ . The overall prevalence of intestinal parasite infections was 28.8%. where Specialist Hospital Yola had a higher prevalence of infection (32.5%), while the lowest was in General Hospital Numan (24.7%). Also, 5 – 12 years had a higher number of identified parasites, while the least was in under five years. The overall Sero-prevalence of the intestinal protozoan infections was 36.2%, where *Entamoeba histolytica* was more prevalent (22.4%) than *Giardia lamblia*, 69(13.8%). Sero-prevalence varied significantly by health facility ( $p < 0.0001$ ), *E. histolytica* being highest at General Hospital, Numan (30.0%), while *G. lamblia* was the lowest (9.0%) in Specialist Hospital, Yola. *Entamoeba histolytica* and *Giardia lamblia* are significant causes of diarrhoea among children in the study areas. The findings demonstrate that serological assays are a more sensitive diagnostic tool than conventional microscopy, revealing a higher true burden of infection. Key risk factors were identified as environmental, including unsafe water, poor sanitation, and household crowding, with school-aged children being particularly vulnerable.

**Keywords:** *Entamoeba histolytica*, *Giardia lamblia*, Seroprevalence, Diarrhoea, Children, Risk Factors, Nigeria

### INTRODUCTION

Diarrhoeal disease remains one of the leading causes of death among children under five globally. According to the World Health Organisation (WHO) and the United Nations International Children's Emergency Fund (UNICEF), diarrhoea was the third leading cause of death among children aged 1–59 months in 2024, resulting in approximately 443,832 deaths each year (WHO, 2024; UNICEF, 2024). The primary causes of diarrhoea are the ingestion of pathogenic bacteria, viruses, or protozoa originating from human or animal faeces. Transmission occurs through contaminated food and water, person-to-person contact, or zoonotic exposure, especially in areas with poor sanitation and hygiene (WHO, 2023). Diarrhoea is defined as the passage of three or more loose or watery stools within 24 hours, and a duration of 14 days or longer is considered persistent diarrhoea (WHO, 2023; UNICEF, 2024).

Intestinal parasitic infections are amongst the most common infections worldwide and have been identified as one of the most significant causes of morbidity and mortality among disadvantaged populations (WHO, 2017). According to the WHO, diarrhoeal disease is the world's second leading cause of paediatric deaths, killing around 525,000 children under five annually. Globally, there are approximately 1.7 billion cases of childhood diarrhoeal disease every year, and diarrhoea due to protozoan infection is widespread throughout the developing world (WHO, 2017). In low-income countries, many of the risk factors for contracting diarrhoeal illnesses are linked to poor socio-economic conditions, such as lack of

access to safe water and sanitation, poor hygiene practices, and unsafe human waste disposal (Boithias *et al.*, 2016). Diarrhoea can be related to a wide range of bacteria, viruses, and intestinal parasites (WHO, 2017).

Intestinal parasitic infections are among the most common infections worldwide and are a significant cause of morbidity, especially among disadvantaged populations. WHO estimates that about 3.5 billion people globally are affected by intestinal parasitic infections, with around 450 million people experiencing symptomatic illness; over 200,000 deaths are attributed annually to these infections. (WHO, 2021). According to the most recent WHO/UNICEF data, diarrhoeal disease is the third leading cause of death among children aged 1–59 months, killing about 443,832 children under five each year; there are roughly 1.7 billion cases of childhood diarrhoeal disease globally per annum. (WHO/UNICEF, 2024). In low-income countries, many risk factors for diarrhoeal illnesses are linked to poor socio-economic conditions, such as lack of access to safe water and sanitation, inadequate hygiene, and unsafe disposal of human waste (WHO, 2024). Diarrhoea can be caused by a wide range of pathogens: bacteria, viruses, and intestinal parasites (WHO, 2024; Franceville *et al.*, 2020).

Soil-transmitted helminth (STH) infections remain among the most common infections worldwide. The World Health Organization (WHO, 2023) estimates that about 1.5 billion people, or approximately 24% of the global population, are infected with one or more soil-transmitted helminths, with over 260 million preschool-age children and 654 million

school-age children living in areas where these infections are intensively transmitted (WHO, 2023).

More recent meta-analytic estimates break this down by species: in 2021, about 732 million people were estimated to harbour *Ascaris lumbricoides*, while *Trichuris trichiura* infections affected roughly 513 million, and hookworm infections (i.e., *Ancylostoma duodenale* + *Necator americanus*) about 113 million (Global Burden of STH Infections Study, 2024). STHs include nematodes such as roundworms (*A. lumbricoides*), whipworms (*T. trichiura*), and hookworms (*A. duodenale* and *N. americanus*), which are highly prevalent in tropical and subtropical regions where water, sanitation, and hygiene (WASH) infrastructure is inadequate (WHO, 2023; Global Burden of STH Infections Study, 2024).

Intestinal protozoa that are most commonly associated with diarrhoea in children include *Entamoeba histolytica*, *Giardia lamblia/duodenalis*, spore-forming protozoans such as *Cyclospora*, *Cryptosporidium*, and *Microsporidium*, as well as *Blastocystis* sp., *Balantidium coli*, and *Dientamoeba fragilis* (Burgess et al., 2017). The diseases caused by these intestinal protozoan parasites—amoebiasis, giardiasis, cyclosporiasis, blastocystosis, cryptosporidiosis, balantidiasis, and dientamoebiasis—are strongly associated with diarrhoeal illnesses (Ahmed et al., 2022). Globally, *Cryptosporidium* spp. remains the most frequently identified protozoan pathogen responsible for diarrhoea, while giardiasis continues to affect an estimated one-third of people in developing countries (Osman et al., 2016; CDC, 2022). Similar to *Cryptosporidium* and *Giardia*, intestinal protozoa such as *Blastocystis* sp. and *D. fragilis* are of increasing public health significance due to their widespread occurrence and potential socio-economic impact. Moreover, *D. fragilis* is often detected in co-infections with other protozoans, especially *Blastocystis* sp. (Stark et al., 2016; Ahmed et al., 2022). Symptoms of infection typically follow an incubation period of about one week and include watery diarrhoea, abdominal cramps, nausea, and loss of appetite, which in children may lead to undernutrition, as observed in multiple studies in sub-Saharan Africa and South America (Das et al., 2023).

Many research works indicated different factors had an association with the prevalence of intestinal protozoan infections among children; some of these factors are related to poverty, socio-demographic characteristics, and some others might be related to individuals' practices, such as age, gender, residence, and occupation. The protozoan parasitic infections are among the most prevalent infections in people living in the study area, which is a result of poor environmental sanitation and very poor personal hygiene and unclean habits practices by endemic community dwellers, compounded by public ignorance and illiteracy of the population. Baseline data on the burden, distribution, and trend of intestinal protozoan parasites can provide essential evidence for implementation of effective prevention strategies in combating these neglected protozoan infections (Sarkari et al., 2016). The degree of harm caused by intestinal parasitic infections to the health of individuals and communities depends largely on the parasite species involved, the presence of concurrent infections, and underlying nutritional and socio-economic conditions. Recent studies highlight that co-infections with multiple intestinal parasites can exacerbate anaemia, stunting, and impaired cognitive development, particularly in low-income populations with poor sanitation and inadequate nutrition (Opara et al., 2022; Adedaja et al., 2023; WHO, 2024). Generally, intestinal parasitic infections are dangerous disease-causing agents among school children,

ultimately resulting in malnutrition. Mohammed et al. (2019) stated that in Nigeria, parasitic infection is endemic and commonly reported among individuals in various communities. Conventional methods, such as microscopic examination of stool samples, have been the mainstay for the diagnosis of protozoan intestinal parasites. This method has its limitations, such as low specificity and sensitivity. This is related to the instability and rapid deterioration of some protozoan parasites once outside the host (Nino et al., 2017). Microscopic examination cannot identify between different species of *Cryptosporidium*, between different assemblages of *Giardia*, between different subtypes of *Blastocystis*, and between pathogenic and non-pathogenic species of *Entamoeba* (Gómez-Márquez et al., 2019; Morales et al., 2021). It is therefore imperative for the use of Serological techniques to diagnose intestinal protozoan parasites in vulnerable populations to accurately measure parasite prevalence in endemic settings. The unsanitary environment in the study areas and environs, where children are commonly vulnerable, and the consumption of contaminated foods and water, which results in many cases of diarrhoea, is rampant. Thus, the main aim of this study was to assess the prevalence of intestinal protozoan parasitic infection in some selected hospitals in Adamawa and to determine the associated risk factors.

## Methodology

### Study Area

This research study was conducted in Adamawa State, North-eastern Nigeria. Three (3) hospitals were selected across the 3 senatorial zones of the state, namely: Specialist Hospital Yola (Central Senatorial Zone), General Hospital Mubi (Northern Senatorial Zone), and General Hospital Numan (Southern Senatorial Zone), which serve patients in the respective zones and even neighbouring states.

Adamawa State is located between Longitude 12.3984° E and Latitude 9.3265° N. With an estimated population of 5,150,630 and an area of roughly 36,917 square kilometers, it is one of Nigeria's largest states (National Population Commission [NPC], 2023). The estimated total population includes 1,395,821 children aged 9 months to 14 years, of whom 875,607 are younger than 5 years. The state experiences both rainy and dry seasons, indicating its tropical climate. Beginning in May, the rainy season lasts until mid- or late-October. A single maximum rainfall and a mean total rainfall of 1,113.3 mm describe the rainfall. The North-Easterly trade wind, which sweeps from the Sahara Desert over West Africa into the Gulf of Guinea, is what defines it. It is dry and dusty. Most of the time, the weather is chilly, but depending on local conditions, it can also be scorching in some areas (Minka and Ayo, 2023). The region's relative humidity is roughly 26% in January and 0% in February. High relative humidity readings of 58, 69, 79, 77, and 66, respectively, can be found from May to October, with July and August showing the highest relative humidity levels at around 80% (Adebayo and Tukur, 2020). The majority of residents work as civil servants, traders, farmers, fishermen, and cattle ranchers. The Sudan savannah vegetation zone, which includes small trees and grasses, is where the study region is located (Yohanna et al., 2016). Many rivers, most of which are seasonal, adequately drain the state. According to Yohanna et al. (2016), the primary river is the River Benue, which has significant tributaries with flat sandy beds, steep rocky incised valley walls, and undulating terrain that is ideal for irrigational farming, fishing, and raising cattle. A prevalent occurrence that happens seasonally (during the rainy season) is flooding, which impacts the majority of the

state's LGAs, particularly the Central and Southern senatorial zones.

### Ethical Approval

The Adamawa State Ministry of Health Ethical Committee on Health Research approved and authorized this study. The Department of Zoology at Modibbo Adama University in Yola provided an introductory letter. Before the commencement of the study, the management of each hospital was informed, and their consent was obtained. To guarantee voluntary involvement, informed assent was obtained from the patients or their caregivers.

### Study Design

A descriptive cross-sectional hospital-based study was conducted at General Hospital Numan, General Hospital Mubi, and Specialist Hospital Yola. These three hospitals provide both inpatient and outpatient care to the people living in the region and the surrounding areas. The medical facilities were chosen due to their strategic roles, which include serving as referral centers for patients from other hospitals and offering specialist medical care to the state and communities in their respective geopolitical zones. The research was conducted from November 2024 to December 2025. Smith's (2013) formula ( $n = [(Zscore)^2 \times SD(1-SD)]/e^2$ ) for sample size determination was used to calculate the sample size. A total of 500 subjects were selected for this study.

Five hundred (500) stool and blood samples were collected in sterile and Ethylenediaminetetraacetic acid (EDTA) containers, respectively, from diarrhoeal subjects. Stool samples were transported to Specialist Hospital Laboratory, Yola, in ice boxes and stored at 15 °C, while the blood sample was transported to Biocursor Laboratory, Yola, for analysis. Prior to sample collection, written informed assent was obtained from caregivers in line with the Declaration of Helsinki (World Medical Association [WMA], 2024).

Each container for both blood and stool was labelled with a unique identification number, capturing the hospital name, age, and gender. Also, a structured questionnaire was administered to caregivers to assess risk factors associated with intestinal protozoan parasitosis and their level of understanding of these infections.

### Stool Macroscopic Examination

Stool samples were examined macroscopically for colour, odour, consistency, presence of mucus, blood, and segments of intestinal parasites (WHO, 2017).

### Stool Microscopic Examination

Intestinal protozoa were detected through microscopic examination of the stool samples. For this study, both the saline wet mount and the formal-ether concentration techniques were employed, following standard procedures recommended by the World Health Organization (WHO, 2017).

### Saline wet-Mount Microscopic Examination

With the aid of a dropper, a drop of physiological saline (0.9%) was placed on a clean, grease-free glass slide. Using an applicator stick, a small portion of stool was transferred onto the saline drop and mixed thoroughly until a homogenous suspension was obtained. A cover slip was gently placed over the preparation, and the slide was examined microscopically under  $\times 10$  and  $\times 40$  objective lenses, following the procedure described by Cheesbrough (2010).

### Formal-ether Concentration Technique

Faecal samples were also assayed using the formal-ether concentration technique as described by Cheesbrough (2010). The resultant sediment was examined microscopically under  $\times 10$  and  $\times 40$  objective lenses. Parasitic ova and cysts encountered were subsequently identified based on their morphological characteristics, with reference to standard identification keys (Cheesbrough, 2010).

### Serological Testing

Serological testing for intestinal protozoan infections was performed using enzyme-linked immunosorbent assay (ELISA) kits (e.g., from Diagnostic Automation, Inc., or equivalent), following the manufacturer's instructions. Approximately 3000 $\mu$ g of serum samples were collected using EDTA containers and centrifuged for 30 minutes at 3000g at 2-8°C within 30 minutes of collection and processed to detect protozoan antigens. Briefly, serum samples were homogenized in the provided dilution buffer to release soluble antigens, followed by centrifugation to remove particulate matter. The clarified supernatant was then transferred into antigen-coated microplate wells.

During incubation at room temperature (20–25°C), protozoan antigens present in the samples bound specifically to capture antibodies immobilized on the plate surface. After washing to remove unbound material, enzyme-conjugated secondary antibodies were added and allowed to bind to the antigen-antibody complexes. Subsequent washing steps ensured the removal of residual unbound conjugate.

A chromogenic substrate was then added, and colour development occurred in proportion to the quantity of antigen present. The reaction was stopped with a terminating solution, and optical densities (OD) were measured at 450 nm using a microplate reader. Positive and negative controls were included in each assay to ensure validity. Results were interpreted according to the manufacturer's cutoff values, with samples above the cutoff considered positive for infection.

### Data Analysis

Data were analysed using both descriptive and inferential statistical methods in R statistical software (R version 4.3.2; R Core Team, 2023). The Chi-square test ( $\chi^2$ ) assessed associations between infections and categorical variables (e.g., location, age, Gender). Binary and multivariate logistic regression models were used to identify risk factors and calculate odds ratios (ORs) and adjusted odds ratios (aORs), controlling for confounders. A p-value of less than 0.05 was considered statistically significant. Before analysis, data were checked for completeness and consistency; missing values were observed and updated.

## RESULTS AND DISCUSSION

A total of 500 school-age children presenting with watery stool or diarrhoea at the selected health facilities were considered for the study. Of this population, 200 children were considered from the Specialist Hospital, Yola, while 150 children each from the facilities for the study. Conventional and serological procedures were compared for the study population in relation to the health facility, Age-Gender distributions, stool consistency, and their risk factors were studied and analysed.

A total of 500 children who presented with diarrhea or watery stool at the health facilities were selected. Out of this population, 200 children from Specialist Hospital Yola, 150 children each from General Hospital Mubi, and General

Hospital Numan. Conventional and Serological methods were compared for the study population, and age-gender distributions, stool consistency, and risk factors were looked at and assessed in connection with the health facility.

The intensity and prevalence of intestinal parasite infections among children attending the selected hospitals in Adamawa State are summarized in Tables 1–4. A total of 500 children were examined, out of which 144 were found to be infected with intestinal parasites, with overall light, moderate, and heavy infection intensities recorded in 33 (22.9%), 98 (68.1%), and 13 (9.0%) participants, respectively.

Microscopical prevalence of intestinal parasites across the three hospitals (Table 1). At General Hospital Mubi,

*Entamoeba histolytica* was the most frequently detected parasite, affecting children (16.0%), followed by Hookworm (4.0%), *Taenia spp.* (2.7%), while the least was *H. nana*, *S. mansoni*, and *G. lamblia*, all having 1.3%. General Hospital Numan showed a similar pattern, with *E. histolytica* identified in children (12.6%), Hookworm (4.0%), and *A. lumbricoides* (3.3%), and the lowest was *Taenia spp.* (2.0%). While at Specialist Hospital Yola, the highest prevalence observed was *E. histolytica* (15.0%), followed by Hookworm (6.0%), and *Taenia spp.* (4.0%). Statistical analysis indicated no significant differences in parasite prevalence among hospitals ( $\chi^2 = 0.308$ ,  $p > 0.05$ ), suggesting a relatively uniform distribution across the health facilities.

**Table 1: Infection Intensity and Prevalence of Intestinal Parasites by Hospital (Microscopy)**

Hospitals	Parasite	No. Examined	Light (%)	Moderate (%)	Heavy (%)	Prevalence (%)	p-value
FMC Mubi	<i>Ascaris lumbricoides</i>	150	0 (0.0)	3 (2.0)	0 (0.0)	3 (2.0)	0.735
	<i>Entamoeba histolytica</i>	”	1 (0.7)	21 (14.0)	1 (0.7)	23 (16.0)	0.322
	<i>Giardia lamblia</i>	”	0 (0.0)	0 (0.0)	2 (1.3)	2 (1.3)	0.257
	Hookworm	”	5 (6.2)	1 (0.7)	0 (0.0)	6 (4.0)	0.185
	<i>Hymenolepis nana</i>	”	0 (0.0)	2 (1.3)	0 (0.0)	2 (1.3)	0.368
	<i>Schistosoma mansoni</i>	”	2 (1.3)	0 (0.0)	0 (0.0)	2 (1.3)	0.414
	<i>Taenia spp.</i>	”	3 (2.0)	1 (0.7)	0 (0.0)	4 (2.7)	0.323
	<b>Total</b>		<b>150</b>	<b>11(7.3)</b>	<b>28(18.7)</b>	<b>3(2.0)</b>	<b>42(28.0)</b>
GH Numan	<i>Ascaris lumbricoides</i>	150	0 (0.0)	5 (3.3)	0 (0.0)	5 (3.3)	0.735
	<i>Entamoeba histolytica</i>	”	2 (1.3)	15 (10.0)	2 (1.3)	19 (12.7)	0.322
	Hookworm	”	2 (1.3)	4 (2.7)	0 (0.0)	6 (4.0)	0.185
	<i>Hymenolepis nana</i>	”	0 (0.0)	4 (4.0)	0 (0.0)	4 (4.0)	0.368
	<i>Taenia spp.</i>	”	1 (0.7)	2 (1.3)	0 (0.0)	3 (2.0)	0.323
	<b>Total</b>		<b>150</b>	<b>5(3.3)</b>	<b>30(20.0)</b>	<b>2(1.3)</b>	<b>37(24.7)</b>
SH Yola	<i>Ascaris lumbricoides</i>	200	0 (0.0)	5 (2.5)	0 (0.0)	5 (2.5)	0.735
	<i>Entamoeba histolytica</i>	”	0 (0.0)	29 (14.5)	1 (0.5)	30 (15.0)	0.322
	<i>Giardia lamblia</i>	”	0 (0.0)	0 (0.0)	5 (2.5)	5 (2.5)	0.257
	Hookworm	”	8 (4.0)	4 (2.0)	0 (0.0)	12 (6.0)	0.185
	<i>Hymenolepis nana</i>	”	0 (0.0)	1 (0.5)	0 (0.0)	1 (0.5)	0.368
	<i>Schistosoma mansoni</i>	”	4 (2.0)	0 (0.0)	0 (0.0)	4 (2.0)	0.414
	<i>Taenia spp.</i>	”	5 (2.5)	1 (0.5%)	2 (1.0%)	8 (4.0)	0.323
	<b>Total</b>		<b>200</b>	<b>17(8.5)</b>	<b>30(15.0)</b>	<b>8(4.0)</b>	<b>65(32.5)</b>
<b>Grand Total</b>	—	<b>500</b>	<b>33 (6.6)</b>	<b>98 (19.6)</b>	<b>13(2.6)</b>	<b>144 (28.8)</b>	0.308

Note: General Hospital (GH), State Specialist Hospital (SH). Prevalence and intensity of intestinal parasites in children by Hospital;  $\chi^2$  analysis showed no significant differences ( $p > 0.05$ ).”

Microscopic examination of intestinal parasites among children with diarrhoea (Table 2). Children aged 5–12 years bore the greatest burden of infection. In this group, *E. histolytica* was most prevalent (13.6%), followed by Hookworm (4.6%) and *A. lumbricoides* (3.0%). While children 13–17 years had a lower prevalence, *E. histolytica*

having 18.5%, Hookworm (6.2%), while the youngest cohort (0–4 years) exhibited minimal infection, with only one case of *E. histolytica* (50.0%). No significant differences were observed in parasite prevalence among the age groups ( $p = 0.308$ ).

**Table 2: Infection Intensity and Prevalence of Intestinal Parasites by Age Group (Microscopy)**

Age (Years)	Parasite	No. Examined	Light (%)	Moderate (%)	Heavy (%)	Prevalence (%)	p-value
0–4	<i>E. histolytica</i>	2	0 (0.0)	1 (50.0)	0 (0.0)	1 (50.0)	-
	<b>Total</b>	<b>2</b>	<b>0(0.0)</b>	<b>1(50.0)</b>	<b>0(0.0)</b>	<b>1(50.0)</b>	
5–12	<i>Ascaris lumbricoides</i>	433	1 (0.2)	12 (2.8)	0 (0.0)	13 (3.0)	0.782

Age (Years)	Parasite	No. Examined	Light (%)	Moderate (%)	Heavy (%)	Prevalence (%)	p-value
13-17	<i>E. histolytica</i>	36	4 (0.9)	53 (12.2)	2 (0.5)	59 (13.6)	0.789
	<i>Giardia lamblia</i>	6	0 (0.0)	0 (0.0)	6 (1.4)	6 (1.4)	0.059
	Hookworm	14	4 (3.2)	6 (1.4)	0 (0.0)	20 (4.6)	1.000
	<i>Hymenolepis nana</i>	5	0 (0.0)	5 (1.2)	0 (0.0)	5 (1.2)	0.257
	<i>Schistosoma mansoni</i>	6	6 (1.4)	0 (0.0)	0 (0.0)	6 (1.4)	1.000
	<i>Taenia spp.</i>	8	3 (0.7)	8 (1.8)	2 (0.5)	13 (3.0)	0.042
	<b>Total</b>	<b>433</b>	<b>28(6.5)</b>	<b>84(19.4)</b>	<b>10(2.3)</b>	<b>122(28.2)</b>	
	<i>E. histolytica</i>	65	0 (0.0)	12 (18.5)	0 (0.0)	12 (18.5)	0.789
	<i>Giardia lamblia</i>	1	0 (0.0)	0 (0.0)	1 (5.4)	1 (5.4)	0.059
	Hookworm	4	4 (6.2)	0 (0.0)	0 (0.0)	4 (6.2)	1.000
	<i>Hymenolepis nana</i>	2	0 (0%)	2 (3.1)	0 (0.0)	2 (3.1)	0.257
<i>Taenia spp.</i>	2	2 (3.1)	0 (0.0)	0 (0.0)	2 (3.1)	0.042	
<b>Total</b>	<b>65</b>	<b>6(9.2)</b>	<b>14(21.5)</b>	<b>1(1.5)</b>	<b>21(32.3)</b>		
<b>Grand Total</b>	—	<b>500</b>	<b>33 (6.6%)</b>	<b>98 (19.2%)</b>	<b>13 (2.6%)</b>	<b>144 (28.8%)</b>	0.308

Microscopic intestinal parasite prevalence in relation to gender (Table 3). Parasite prevalence did not differ substantially between genders. Both females and males had *E. histolytica* as the most common infection, each with 36 cases (14.6%) and (14.2%), respectively. Among females, *Taenia spp.* (4.0%) and Hookworm (2.8%) were also common, where

males showed higher prevalence of Hookworm (6.8%) and *A. lumbricoides* (4.2%). Overall, light, moderate, and heavy infections in both males and females were 6.6%, 19.2%, and 2.6% children, respectively. Statistical comparisons confirmed no significant differences in prevalence between genders ( $\chi^2 = 0.308, p > 0.05$ ).

**Table 3: Infection Intensity and Prevalence of Intestinal Parasites by Gender (Microscopy)**

Gender	Parasite	No. Examined	Light (%)	Moderate (%)	Heavy (%)	Prevalence (%)	$\chi^2$ (p-value)	
Female	<i>Ascaris lumbricoides</i>	247	0 (0.0)	7 (2.8)	0 (0.0)	7 (2.8)	0.782	
	<i>Entamoeba histolytica</i>	36	3 (1.2)	32 (13.0)	1 (0.4)	36 (14.6)	0.134	
	<i>Giardia lamblia</i>	3	0 (0.0)	0 (0.0)	3 (1.2)	3 (1.2)	0.705	
	Hookworm	4	4 (1.6)	3 (1.2)	0 (0.0)	7 (2.8)	1.000	
	<i>Hymenolepis nana</i>	4	0 (0.0)	4 (1.6)	0 (0.0)	4 (1.6)	0.705	
	<i>Schistosoma mansoni</i>	3	3 (1.2)	0 (0.0)	0 (0.0)	3 (1.2)	1.000	
	<i>Taenia spp.</i>	10	7 (2.8)	3 (1.2)	0 (0.0)	10 (4.0)	0.099	
	<b>Total</b>	<b>247</b>	<b>17(6.9)</b>	<b>49(19.8)</b>	<b>4(1.6)</b>	<b>70(28.3)</b>		
	Male	<i>Ascaris lumbricoides</i>	253	0 (0.0)	6 (2.4)	0 (0.0)	6 (2.4)	0.782
		<i>Entamoeba histolytica</i>	36	0 (0.0)	33 (13.0)	3 (1.2)	36 (14.2)	0.134
<i>Giardia lamblia</i>		4	0 (0.0)	0 (0.0)	4 (1.6)	4 (1.6)	0.705	
Hookworm		11	11 (4.3)	6 (2.4)	0 (0.0)	17 (6.7)	1.000	
<i>Hymenolepis nana</i>		3	0 (0.0)	3 (1.2)	0 (0.0)	3 (1.2)	0.705	
<i>Schistosoma mansoni</i>		3	3 (1.2)	0 (0.0)	0 (0.0)	3 (1.2)	1.000	
<i>Taenia spp.</i>		5	2 (0.8)	1 (0.4)	2 (0.8)	5 (2.0)	0.099	
<b>Total</b>		<b>253</b>	<b>16(6.3)</b>	<b>49(19.4)</b>	<b>9(3.6)</b>	<b>74(29)</b>		
<b>Grand Total</b>		—	<b>500</b>	<b>33 (6.6%)</b>	<b>98 (19.2%)</b>	<b>13 (2.6%)</b>	<b>144 (28.8%)</b>	0.308

The comparative distribution of protozoan parasites by health facility, Gender, and age is presented in Table 5. The prevalence of the intestinal protozoan parasite showed a higher prevalence with serological diagnosis than with microscopic examination. *E. histolytica* was most prevalent at GH Numan, both microscopically (12.7%) and serologically (30.0%), followed by General Hospital Mubi (16.0% and 22.0%), while the least was recorded in SH Yola (15.0% & 17.0%). Similarly, *G. lamblia* Sero-prevalence was

highest in GH Numan (20.0%), with no parasite identified microscopically, compared with GH Mubi and SH Yola.

By gender, *E. histolytica* was slightly more common in males serologically (23.3%), while *G. lamblia* was more frequent among females (17.0%). However, females had slightly higher *E. histolytica* microscopic prevalence (14.6%) than their male counterparts (14.2%). *G. lamblia* microscopic prevalence indicates males had slightly higher prevalence (1.6%) than females (1.2%), though neither association reached significance.

Both parasites were largely confined to the 5–12 years group, while serological diagnosis identified 181 positive cases (36.2%), indicating a higher detection rate by the serological method. Overall, microscopy detected 79 positive cases (15.8%),

**Table 4: Comparative Analysis of Intestinal Protozoan Parasites Using Microscopy and Serological Diagnosis**

Variable	Category	Parasite	No. Examined	Microscopy		Serological Diagnosis	
				Prevalence (%)	$\chi^2$ (p-value)	Prevalence (%)	$\chi^2$ (p-value)
Health Facility	GH Mubi	<i>E. histolytica</i>	150	23 (15.3)	0.322	33 (22.0)	
		<i>G. lamblia</i>	”	2 (1.3)	0.257	21 (14.0)	
	GH Numan	<i>E. histolytica</i>	150	19 (12.7)	0.322	45 (30.0)	8.35 (0.0154)*
		<i>G. lamblia</i>	”	0 (0.0)		30 (20.0)	8.37 (0.0127)*
	SH Yola	<i>E. histolytica</i>	200	30 (15.0)	0.322	34 (17.0)	0.95 (0.876)
		<i>G. lamblia</i>	”	5 (2.5)	0.257	18 (9.0)	0.92 (0.823)
Total		500	79 (15.8)	0.308	181 (36.2)		
Gender	Female	<i>E. histolytica</i>	247	36 (14.6)	0.134	53 (21.5)	
		<i>G. lamblia</i>	”	3 (1.2)	0.705	42 (17.0)	
		Total	500	79 (15.8)	0.308	181 (36.2)	
	Male	<i>E. histolytica</i>	253	36 (14.2)	0.134	59 (23.3)	0.15 (0.695)
		<i>G. lamblia</i>	”	4 (1.6)	0.705	27 (10.7)	3.70 (0.0545)
		Total	500	79 (15.8)	0.308	181 (36.2)	
Age Group	0–4	<i>E. histolytica</i>	2	1 (50.0)	-	2 (100.0)	
		<i>G. lamblia</i>	”	0 (0.0)		0 (0.0)	
	5–12	<i>E. histolytica</i>	433	59 (13.6)	0.789	100 (23.1)	1.28 (0.528)
		<i>G. lamblia</i>	”	6 (1.4)	0.059	62 (14.3)	0.92 (0.631)
	13–17	<i>E. histolytica</i>	65	12 (18.5)	0.789	12 (18.5)	1.82 (0.118)
		<i>G. lamblia</i>	”	1 (5.4)	0.059	7 (10.7)	1.55 (0.278)
Total		500	79 (15.8)	0.308	181 (36.2)		

The serological prevalence of intestinal protozoan infections demonstrated a clear variation according to stool consistency (Figure 1). *Entamoeba histolytica* exhibited its highest prevalence in soft stool samples, with considerable prevalence also observed in loose and brown soft stools. Prevalence was lower in mucous, formed, and watery stools. In contrast, *Giardia lamblia* showed a distinct pattern, with the highest prevalence in brown soft stool and markedly lower

proportions in all other consistencies, including formed, mucous, watery, and soft stool. It is noteworthy that no infections were detected in the categories labelled simply as “soft” and “soft stool,” highlighting potential inconsistencies in classification or the absence of infection in those specific sub-groups. This establishes stool consistency as a strong visual indicator of infection severity.

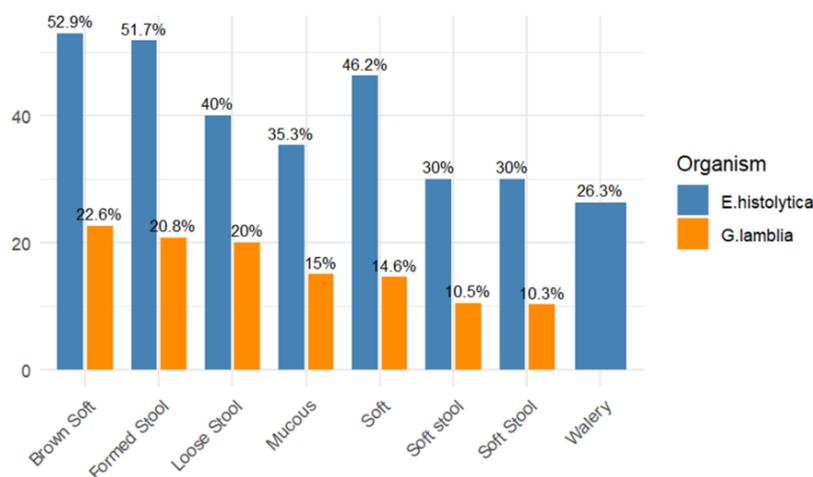


Figure 1: Serological Prevalence of *Giardia lamblia* and *Entamoeba histolytica* Across Diarrhoeal (Stool) Types

Risk Factors Associated with *Entamoeba histolytica* and *Giardia lamblia* Infections in Children with Diarrhoea (Table 5). A multivariate logistic regression model was used to identify independent predictors of *Entamoeba histolytica* and *Giardia lamblia* seropositivity among the study population Table 4.9. The results revealed that stool consistency and sex were significant predictors of infection, while other demographic and environmental variables were not statistically associated with protozoan prevalence. For *E. histolytica*, stool consistency was significantly associated with infection status. Participants with formed stools were less likely to test positive for *E. histolytica* compared to those with soft stools (AOR = 0.35, 95% CI: 0.13–0.92,  $p = 0.033$ ). Similarly, watery stool consistency was strongly associated with reduced odds of *E. histolytica* seropositivity (AOR = 0.10, 95% CI: 0.03–0.30,  $p < 0.001$ ). These findings suggest that infection was more frequently detected among individuals with soft stool, indicating a possible relationship between stool consistency and active intestinal infection. No statistically significant associations were observed between *E. histolytica* infection and sex, age group, health facility, source of drinking water, or household size. Males had slightly higher odds of infection than females (AOR = 1.14, 95% CI:

0.84–1.86,  $p = 0.600$ ), but this relationship was not significant. Similarly, participants aged 5–12 years and 13–17 years showed higher odds of infection compared to the 0–4-year group, although the associations were not statistically meaningful ( $p > 0.05$ ). For *G. lamblia*, both sex and stool consistency were significantly associated with infection. Males were significantly less likely to be infected than females (AOR = 0.42, 95% CI: 0.22–0.80,  $p = 0.009$ ). In contrast, stool consistency again showed a strong relationship with infection prevalence: individuals with formed stools (AOR = 0.26, 95% CI: 0.09–0.67,  $p = 0.006$ ) and those with watery stools (AOR = 0.19, 95% CI: 0.07–0.53,  $p = 0.002$ ) were significantly less likely to have *G. lamblia* seropositivity compared to those with soft stool. Other variables, including age group, health facility, source of drinking water, and household size, did not show statistically significant associations with *G. lamblia* infection ( $p > 0.05$ ). However, participants relying on unsafe water sources had slightly higher odds of infection than those using safe sources, although the association approached but did not reach statistical significance (AOR = 0.55, 95% CI: 0.30–1.00,  $p = 0.051$ ).

**Table 5: Multivariate Logistic Regression Analysis of Factors Associated with Serological Prevalence of Intestinal Protozoan Parasite Infections**

Variable	Category	<i>E. histolytica</i> AOR (95% CI)	p-value	<i>G. lamblia</i> AOR (95% CI)	p-value
Gender	Male	1.14 (0.84 - 1.86)	0.600	0.42 (0.22 - 0.80)	0.009*
	Female	Ref		Ref	
Age Group	0-4 years	Ref		Ref	
	5-12 years	1.45 (0.70 - 3.01)	0.316	1.62 (0.75 - 3.49)	0.218
	13-17 years	1.82 (0.86 - 3.85)	0.118	1.55 (0.70 - 3.41)	0.278
Stool Consistency	Formed	0.35 (0.13 - 0.92)	0.033	0.26 (0.09 - 0.67)	0.006*
	Watery	0.10 (0.03 - 0.30)	<0.001*	0.19 (0.07 - 0.53)	0.002*
	Soft	Ref		Ref	
Health Facility	GH Numan	1.08 (0.59 - 2.01)	0.800	1.15 (0.58 - 2.29)	0.690
	SHY Yola	0.95 (0.50 - 1.81)	0.876	0.92 (0.45 - 1.89)	0.823
	GH Mubi	Ref		Ref	
Source of Drinking Water	Safe (piped/borehole)	0.70 (0.40 - 1.23)	0.220	0.55 (0.30 - 1.00)	0.051
	Unsafe (surface/unprotected well)	Ref		Ref	
Household Size	≤5 members	Ref		Ref	
	>5 members	1.35 (0.80 - 2.28)	0.250	1.50 (0.90 - 2.50)	0.120

Note: GH=General Hospital, AOR = Adjusted Odds Ratio; CI = Confidence Interval; Ref = Reference category;  $p < 0.05$  indicates statistical significance.

## DISCUSSION

Diarrheal infections in children are largely caused by intestinal protozoan parasites, especially in underdeveloped nations where transmission is facilitated by inadequate sanitation and contaminated water sources. Serological diagnostic techniques are more sensitive than traditional microscopy, which makes it possible to identify risk variables and estimate prevalence more precisely. The serological and microscopical prevalence of intestinal protozoan parasites in children with diarrhea in the General Hospitals Mubi and Numan, Nigeria, as well as the Specialist Hospital Yola, was evaluated in this study.

The overall prevalence of intestinal parasitic infections observed in this study (28.8%) is consistent with reports from many African and global surveys, which commonly range from 20% to 40% depending on the study population, diagnostic techniques, and level of intervention coverage

(Sitotaw *et al.*, 2020; Hajissa *et al.*, 2022). This level of infection indicates that intestinal parasites remain a significant public health concern among children in Adamawa State, despite ongoing preventive measures. The predominance of moderate-intensity infections (19.6%) reflects persistent transmission, likely sustained by reinfection following mass drug administration (MDA) programs, as has been documented in similar endemic settings (Sitotaw *et al.*, 2020).

The most frequently detected parasite was *Entamoeba histolytica*, which was observed across all three health facilities. This finding is in line with previous clinic-based studies in Nigeria and other African countries where protozoan parasites are frequently reported among symptomatic populations (Roro *et al.*, 2022; Pukuma *et al.*, 2023). In contrast, community-based surveys in Nigeria often identify soil-transmitted helminths (STHs), particularly

*Ascaris lumbricoides* and hookworm, as the dominant species (Oyeyemi et al., 2022; Akinbo et al., 2019). These differences may be explained by variations in diagnostic methodology, setting (hospital-based versus community-based sampling), and the impact of periodic deworming campaigns, which substantially reduce helminth prevalence but have a limited effect on protozoan infections. The reliance on microscopy in the present study also limits differentiation between *E. histolytica* and non-pathogenic *E. dispar*, potentially leading to an overestimation of the burden of pathogenic *Entamoeba*. Gender-related differences were not significant, which concurs with findings from other African studies reporting broadly similar infection prevalence among males and females (Sitotaw et al., 2020; Tyoalumun et al., 2021). Where differences exist, they are generally marginal and attributed to behavioural rather than biological factors.

Age-stratified analysis demonstrated that children aged 5–12 years bore the highest infection burden, with significantly higher prevalence compared to younger and older cohorts. This pattern is consistent with global and African evidence, which indicates that school-age children are the most at-risk group due to frequent outdoor play, greater soil exposure, and poor hygiene practices (Hajissa et al., 2022; Hakizimana et al., 2023). The low prevalence among children under five years is likely related to closer parental supervision and reduced environmental exposure, while the decline in prevalence among older adolescents reflects improved hygiene awareness and behaviours. Regionally, the findings correspond with earlier studies from Adamawa and north-eastern Nigeria, where prevalence among children has been estimated between 25% and 35%, with mixed protozoan and helminth distributions (Pukuma et al., 2023; Nwaorgu et al., 2018). The dominance of *E. histolytica* in the current study may reflect both the diagnostic limitations of microscopy and ongoing challenges in water, sanitation, and hygiene (WASH) infrastructure, which facilitate faecal–oral protozoan transmission (Roro et al., 2022).

Serological Prevalence of Intestinal Protozoan Infections by Health Facility, Age, and Gender. In addition to microscopy-based estimates, the serological prevalence of intestinal protozoan infections of 36.2% observed in this study is higher than the microscopy-based prevalence (15.8%), reflecting the greater sensitivity of serological assays in detecting current or past exposure. This pattern has been consistently reported in global literature, where serological methods often yield higher prevalence estimates for *E. histolytica* and *Giardia lamblia* than conventional microscopy (Ali et al., 2019; Lebbad et al., 2021). Such findings underscore the limitations of stool microscopy and highlight the need for incorporating serological or Serological diagnostics in routine surveillance. *E. histolytica* was the most frequently detected parasite, followed by *G. lamblia* across all study parameters (Health Facility, Age, and Gender distributions). The predominance of *E. histolytica* aligns with earlier studies in Nigeria and other African settings where amoebiasis remains a leading cause of protozoan-associated diarrhoea (Roro et al., 2022; Pukuma et al., 2023). Globally, *Giardia* often emerges as the most prevalent intestinal protozoan, especially in community-based surveys of children in Asia and Latin America (Fletcher et al., 2020; Certad et al., 2017). The lower *Giardia* prevalence in our study compared to global estimates may reflect local transmission ecology and differences in water source contamination patterns. Moreover, hospital-based sampling may bias results toward *E. histolytica*, given its higher likelihood of symptomatic presentation. The highest Sero-prevalence for both *E. histolytica* and *G. lamblia* was observed in General Hospital Numan (30.0% and 20.0%),

significantly exceeding rates at the General Hospital Mubi (22.0% and 14.0%) and Specialist Hospital Yola (17.0% and 9.0%), respectively. This geographic heterogeneity suggests localized environmental or infrastructural factors, such as differences in water supply, sanitation, or food hygiene practices, that facilitate protozoan transmission. Similar intra-regional variations have been reported in Ethiopia, Sudan, and northern Nigeria, where hospital catchment areas with poor WASH conditions consistently record higher protozoan burdens (Hajissa et al., 2022; Oyeyemi et al., 2022). The significantly elevated rates in Numan may therefore reflect site-specific vulnerabilities that warrant targeted interventions. The overall Sero-prevalence for both *E. histolytica* and *G. lamblia* was slightly higher among females (21.5% and 17.0%) than their male counterparts (23.3% and 10.7%); the difference by gender was not statistically significant. This pattern is in agreement with most African and Nigerian studies reporting no consistent gender-associated differences in protozoan prevalence (Sitotaw et al., 2020; Tyoalumun et al., 2021). Where gender disparities are detected, they are typically modest and linked to behavioural factors, such as domestic water-related chores for girls or outdoor play for boys, rather than inherent susceptibility. Interestingly, our data suggested that *E. histolytica* was more frequent in males, whereas *G. lamblia* was more common in females, though neither reached significance. Sero-prevalence in relation to age distribution showed that infections were overwhelmingly concentrated among children aged 5–12 years for *E. histolytica* (23.1%) and *G. lamblia* (14.3%). This age profile is consistent with global and African evidence identifying school-age children as the group most at risk for protozoan infections due to increased exposure through contaminated water, outdoor play, and inadequate hygiene (Fletcher et al., 2022; Hakizimana et al., 2023). The near absence of infection in children under five years mirrors findings from Rwanda and Nigeria, where younger children are relatively protected by closer parental supervision and reduced independent exposure to contaminated environments (Oyeyemi et al., 2022; Hakizimana et al., 2023). Comparatively, this study demonstrated that serological assays were more effective than microscopy for detecting *Entamoeba histolytica* and *Giardia lamblia* infections. The consistent superiority of serological diagnosis across health facilities, genders, and age groups highlights the limitations of microscopy as a standalone diagnostic tool. The enhanced performance of serological methods reflects their ability to detect antigen–antibody reactions even in subclinical or low-intensity infections that may not be visible under light microscopy. At the facility level, serological methods identified a greater number of infections than microscopy, and in some locations, the difference was statistically significant. This supports previous evidence that microscopy underestimates protozoan infection rates due to its dependence on sample quality, the presence of viable cysts or trophozoites, and operator expertise (Fitri et al., 2022; Morán et al., 2023). In contrast, serological techniques can detect circulating parasite-specific antigens or antibodies regardless of parasite stage, offering improved diagnostic sensitivity and reproducibility. Gender-related patterns in infection were not statistically significant, although slight variations were observed between males and females. Such differences are often associated with behavioral and environmental factors, including water contact patterns, hygiene practices, and occupational exposure, as previously documented in endemic communities (Ali et al., 2020). The absence of significant gender-based differences in this study suggests that exposure to infection sources is likely similar among both groups

within the study area. Age-related analysis indicated that younger individuals, particularly school-aged children, were more commonly affected. This observation aligns with previous studies that have reported higher vulnerability to intestinal protozoan infections among children due to immature immunity, poor hygiene practices, and frequent exposure to contaminated environments (Nkrumah & Nguah, 2011; Zemene *et al.*, 2018). Although the association between age and infection was not statistically significant, the trend underscores the importance of targeted preventive measures among children. The findings of this study emphasize that microscopy, though cost-effective and widely accessible, lacks the diagnostic precision required for accurate epidemiological assessment. Serological assays, on the other hand, offer higher sensitivity and can detect both symptomatic and asymptomatic infections. However, serological techniques are not without limitations, including the potential for cross-reactivity with other protozoa and the persistence of antibodies following infection clearance (Morán *et al.*, 2023). Despite these limitations, serological and other immunodiagnostic approaches remain valuable tools for improving case detection and strengthening surveillance systems.

In order to understand the clinical significance of these infections, we analysed stool consistencies and socio-demographic factors (Figure 1). The present study demonstrates that stool consistency is a strong predictor of intestinal protozoan infection, with *Entamoeba histolytica* and *Giardia lamblia* showing distinct prevalence profiles across diarrhoeal types. The higher prevalence of *E. histolytica* in soft (46.2%), loose (40.0%), and brown soft (52.9%) stools aligns with the pathogen's invasive potential, which commonly manifests as dysentery or mucoid diarrhoea (Roro *et al.*, 2022). The positive association between watery or loose stool and infection intensity observed here is consistent with global reports describing diarrhoea severity as a clinical correlate of invasive amoebiasis, particularly in children (Shirley *et al.*, 2019). Conversely, the predominance of *G. lamblia* in brown soft stools (22.6%) and its lower occurrence in watery or mucous stools (15.0%) corresponds with its known non-invasive pathogenesis, which typically produces chronic, intermittent, and less severe diarrhoeal presentations (Cotton *et al.*, 2011). These patterns are broadly consistent with African studies, where stool form has been identified as an important diagnostic clue in differentiating protozoan infections (Hajissa *et al.*, 2022). For instance, Ethiopian and Rwandan surveys similarly reported *E. histolytica* predominating in children with dysenteric diarrhoea, while *Giardia* was more frequently isolated from children with semi-formed stool (Mengistie *et al.*, 2018; Hakizimana *et al.*, 2023). In Nigeria, clinic-based studies also confirm these distinctions, with *E. histolytica* often associated with bloody or mucous diarrhoea and *Giardia* with prolonged but less acute forms of diarrhoea (Akinbo *et al.*, 2019; Pukuma *et al.*, 2023). The current findings, therefore, reinforce the clinical relevance of stool consistency in guiding presumptive diagnosis in resource-limited settings where Serological tests are unavailable.

Multivariate logistic regression analysis of factors associated with serological prevalence of *Entamoeba histolytica* and *Giardia lamblia* indicated that socio-demographic factors were not significantly associated with *E. histolytica* infection in this study; the result is in agreement with global and African reports that age and gender exert minimal independent influence when environmental exposures and sanitation practices are the main drivers (Sitotaw *et al.*, 2020). Similarly, the absence of significant facility-level differences

supports the view that risk is shaped more by community-level transmission than by point-of-care characteristics, as documented in other Nigerian surveys (Nwaorgu *et al.*, 2018; Oyeyemi *et al.*, 2022). For *G. lamblia*, however, gender emerged as a significant predictor, with females exhibiting higher odds of infection. While some African studies have reported no Gender differences (Tyoalumun *et al.*, 2021), others suggest slightly higher female prevalence, potentially reflecting gendered roles such as water collection and domestic tasks that increase exposure to contaminated water (Hajissa *et al.*, 2022). The marked effect of stool consistency on *Giardia* infection risk also echoes global evidence, which emphasizes that semi-formed stools are more likely to yield cysts, while watery diarrhoea is less diagnostic given reduced cyst excretion during acute episodes (Cotton *et al.*, 2011). At the regional level, the findings are consistent with earlier work in Adamawa and north-eastern Nigeria, where protozoan infections, particularly *E. histolytica*, were linked to symptomatic diarrhoea among children (Pukuma *et al.*, 2023). The strong predictive value of stool form highlights a practical diagnostic marker in these resource-limited settings, where syndromic diagnosis remains common. Risk factors associated with *Entamoeba histolytica* and *Giardia lamblia* infections in children with diarrhoea explore key risk factors, including socio-demographic variables and clinical features that influence infection with *E. histolytica* and *G. lamblia*. This study identified stool consistency as the strongest predictor of protozoan infection, whereas socio-demographic factors, including gender, age, and health facility, were largely non-significant. For *Entamoeba histolytica*, the absence of significant demographic associations mirrors findings from global and African studies, which consistently report that environmental determinants such as poor sanitation, unsafe water, and hygiene practices outweigh individual demographic factors in shaping risk (Sitotaw *et al.*, 2020; Hajissa *et al.*, 2022). The markedly reduced odds of infection among children with formed and watery stools compared to those with soft stools confirm stool characteristics as a clinical correlate of amoebic infection, consistent with evidence that *E. histolytica* is most frequently associated with semi-formed or mucoid diarrhoea (Shirley *et al.*, 2019). In contrast, *Giardia lamblia* infection demonstrated significant associations with both gender and stool form. Females exhibited higher odds of infection, a pattern that has been reported in some African settings where gendered roles, such as water collection and food preparation, increase exposure to contaminated sources (Hakizimana *et al.*, 2023; Tyoalumun *et al.*, 2021). The protective effect observed among males in this study corresponds with Nigerian surveys that similarly documented higher giardiasis prevalence among girls, though findings across regions remain inconsistent (Akinbo *et al.*, 2019; Oyeyemi *et al.*, 2022). Stool consistency again emerged as a key determinant, with soft stools conferring the greatest risk, in line with global literature indicating that *Giardia* cyst excretion peaks in semi-formed stools and diminishes in watery diarrhoea, complicating detection during acute episodes (Cotton *et al.*, 2011). The non-significance of age in both infections diverges from several community-based surveys that often report higher *Giardia* prevalence in younger children and higher *E. histolytica* burden in school-aged groups (Hajissa *et al.*, 2022; Mengistie *et al.*, 2018). This discrepancy may be explained by the hospital-based sampling frame, where symptomatic cases across all ages present for care, potentially attenuating age-related patterns observed in community surveys. Similarly, the lack of variation by health facility suggests shared exposure risks across communities in Adamawa State,

consistent with prior reports attributing transmission primarily to environmental and household-level factors rather than healthcare access (Pukuma *et al.*, 2023). At the regional level, the findings complement earlier studies in north-eastern Nigeria that have identified unsafe water sources, poor sanitation, and diarrhoeal symptoms as stronger risk factors for protozoan infections than socio-demographic variables (Nwaorgu *et al.*, 2018; Pukuma *et al.*, 2023). By highlighting stool consistency and, in the case of *Giardia*, female Gender as significant predictors, this study underscores the importance of integrating clinical presentation with gender-sensitive public health strategies. Collectively, the results reinforce that behavioural and environmental determinants, rather than demographic characteristics, drive protozoan transmission in endemic settings. They further suggest that stool characteristics provide a low-cost, practical diagnostic marker to guide presumptive treatment, especially where laboratory confirmation is limited.

### CONCLUSION

This study confirms that intestinal protozoan infections, primarily *Entamoeba histolytica* and *Giardia lamblia*, are a significant cause of diarrhoea among children in the study areas. The findings demonstrate that serological assays are a more sensitive diagnostic tool than conventional microscopy, revealing a higher true burden of infection. Key risk factors were identified as environmental—including unsafe water, poor sanitation, and household crowding—with school-aged children being particularly vulnerable. These findings underscore the necessity of integrating improved serological diagnostics into routine practice and implementing robust water, sanitation, and hygiene (WASH) interventions to effectively reduce morbidity and control disease transmission in the region.

### REFERENCES

- Adebayo, A. A., Tukur, A. L., & Zemba, A. A. (2020). Adamawa State in maps (2nd ed.). Modibbo Adama University of Technology & Paraclete Publishers
- Adedoja, A. O., Adeyeba, O. A., & Akinyemi, K. (2023). Co-infection and nutritional implications of intestinal parasites among Nigerian children. *Journal of Tropical Medicine and Health*, 12(3), 145–153
- Ahmed, R., Ali, S., & Khan, M. A. (2022). Global burden of intestinal protozoan infections: Epidemiological insights and public health impact. *Parasitology International*, 87, 102490
- Akinbo, F. O., Okaka, C. E., & Omoregie, R. (2019). Intestinal parasitic infections among patients with diarrhoea in Benin City, Nigeria. *Annals of Tropical Medicine and Public Health*, 12(3), 87–93
- Akinbo, F. O., Okaka, C. E., Omoregie, R., & Dearen, T. (2019). Prevalence of intestinal parasites among school children in Nigeria: Implications for control. *Journal of Infection in Developing Countries*, 13(7), 601–607
- Ali, I. K. M., Clark, C. G., & Petri, W. A. (2019). Molecular and serological epidemiology of *Entamoeba histolytica* and *Giardia lamblia* infections. *International Journal for Parasitology*, 49(5), 341–351
- Ali, M., Hossain, M. I., & Karim, M. M. (2020). Relationship between stool consistency and detection of *Giardia lamblia*

and *Entamoeba histolytica* among school-aged children. *Journal of Tropical Medicine and Hygiene*, 23(4), 145–152

- BMC Infectious Diseases. (2024). Global estimates of intestinal parasitic infections. *BMC Infectious Diseases*, 24(1), 230–245
- Boithias, L., Choisy, M., Souliyaseng, N., Jourden, M., Quet, F., and Buisson, Y. (2016). Hydrological Regime and Water Shortage as Drivers of the Seasonal Incidence of Diarrhoeal Diseases in a Tropical Montane Environment. *PLoS Neglected Tropical Disease* 10(12): e0005195
- Burgess, S.L., Gilchrist, C.A., Lynn, T.C., and Petri, W.A. (Jr). (2017). Parasitic Protozoa and Interactions with the Host Intestinal Microbiota. *Infection and immunity*, 85(8): e00101-17
- Centers for Disease Control and Prevention (CDC). (2022). Cryptosporidiosis (Cryptosporidium): Epidemiology & Risk Factors. <https://www.cdc.gov/parasites/crypto/>
- Certad, G., Benamrouz, S., Guyot, K., Mouray, A., Chassat, T., Delaire, B., ... & Dei-Cas, E. (2020). *Giardia duodenalis* infection: Clinical relevance and pathophysiology. *International Journal for Parasitology*, 50(3), 167–181
- Certad, G., Benamrouz-Vanneste, S., Gantois, N., & Guyot, K. (2017). Cryptosporidium and *Giardia* in immunocompromised patients: Update on clinical and therapeutic management. *Current Clinical Microbiology Reports*, 4(2), 94–102
- Cheesbrough, M. (2010). *District Laboratory Practice in Tropical Countries*, 2nd Ed. Cambridge University Press, London, UK. 310-311.
- Cotton, J. A., Beatty, J. K., & Buret, A. G. (2011). Host-parasite interactions and pathophysiology in *Giardia* infections. *International Journal for Parasitology*, 41(9), 925–933.
- Das, S., Mondal, D., & Haque, R. (2023). Impact of intestinal protozoa on child growth and nutrition in developing countries. *Journal of Global Health*, 13, 04023
- Fitri, L., Wahyuni, S., & Siregar, A. (2022). Comparison of microscopy and serological methods in the detection of intestinal protozoan infections. *Asian Pacific Journal of Tropical Biomedicine*, 12(9), 401–408
- Fletcher, S. M., Stark, D. J., & Ellis, J. T. (2022). Global burden and epidemiology of intestinal protozoa. *Tropical Medicine & International Health*, 27(8), 723–740
- Fletcher, S. M., Stark, D., & Ellis, J. (2020). Prevalence and risk factors for *Giardia* infection in low- and middle-income countries: A systematic review and meta-analysis. *PLoS Neglected Tropical Diseases*, 14(7), e0008412
- Franceville, M., Pombi, M., & Nguema, P. (2020). Protozoan diarrheal infections in Sub-Saharan Africa: An epidemiological review. *African Journal of Infectious Diseases*, 14(1), 17–25
- Global Burden of Soil-Transmitted Helminth Infections Study. (2024). Global distribution and burden of soil-

- transmitted helminth infections, 1990–2021. *The Lancet Global Health*, 12(2), e178–e191
- Gómez-Márquez, C., Díaz, E., & López, P. (2019). Limitations of microscopy in diagnosing intestinal protozoan infections. *Diagnostic Parasitology*, 5(1), 12–19
- Hajissa, K., Islam, M. A., Sanyang, A. M., & Mohamed, Z. (2022). Prevalence of intestinal protozoan parasites among school children in Africa: A systematic review and meta-analysis. *PLoS Neglected Tropical Diseases*, 16(10), e0009971
- Hakizimana, E., Ntiringanya, L., Ndayisaba, R., & Habimana, J. (2023). Intestinal parasitic infections among children aged 12–59 months in Nyamasheke District, Rwanda. *Parasites & Vectors*, 16(1), 331. <https://doi.org/10.1186/s13071-023-05813-9>
- Lebbad, M., Winięcka-Krusnell, J., & Lebbad, A. (2021). Serological and molecular diagnosis of intestinal protozoa: Comparative evaluation of diagnostic tools. *Parasites & Vectors*, 14(1), 612.
- Mengistie, Z., Tadesse, G., & Endris, M. (2018). Prevalence and associated risk factors of intestinal protozoan infections among children in Ethiopia. *Ethiopian Journal of Health Sciences*, 28(5), 589–598
- Minka, N.S. and Ayo, J.O. (2023). "Influence of cold–dry (harmattan) season on colonic temperature and the development of pulmonary hypertension in broiler chickens, and the modulating effect of ascorbic acid". *Dove Press Journal*, 6:1 – 11
- Mohammed, K., Gulma, M.K., Yahaya, M., Spencer, T., Nataala, S. and Garba, M. (2019). Prevalence of Intestinal Parasitic Infections among Patients Attending Usmanu Danfodiyo University Teaching Hospital, Sokoto, Nigeria. *Asian Journal of Research in Infectious Diseases*, 2019, 1–9.
- Morales, L., Villavicencio, J., & Rivera, R. (2021). Advances in molecular diagnostics of intestinal protozoa. *Frontiers in Parasitology*, 2: 57.
- Morán, P., Ramos, F., & Gómez, A. (2023). Serological and molecular diagnosis of *Entamoeba histolytica*: Advances and limitations. *Acta Tropica*, 245, 106882
- National Population Commission (NPC). (2023). Population projection report for Nigeria and its sub-national entities: 2023 revision. Abuja: National Population Commission
- Nino, A., Torres, M., & Delgado, R. (2017). Comparative performance of microscopy and serological methods in detecting intestinal protozoa. *Parasite Epidemiology and Control*, 2(3), 158–165
- Nkrumah, B., & Nguah, S. B. (2011). *Giardia lamblia*: A major cause of childhood diarrhoea in Ghana. *BMC Infectious Diseases*, 11, 307
- Nwaorgu, O. C., Ohaeri, C. C., & Ekejindu, I. M. (2018). Prevalence and risk factors for intestinal parasites among schoolchildren in north-eastern Nigeria. *Journal of Parasitology Research*, 2018, 1–8. <https://doi.org/10.1155/2018/7681058>
- Opara, K. N., Udoidung, N. I., & Arong, G. A. (2022). Intestinal parasitic infections and their nutritional impacts on Nigerian school children. *Journal of Parasitology Research*, 2022, 1–10
- Osman, M., El Safadi, D., Cian, A., Benamrouz, S., Nourrisson, C., Poirier, P. (2016). Prevalence and Risk Factors for Intestinal Protozoan Infections with *Cryptosporidium*, *Giardia*, *Blastocystis* and *Dientamoeba* among Schoolchildren in Tripoli, Lebanon. *PLoS Neglected Tropical Diseases*, 10(3):27-31
- Oyeyemi, O. T., Afolabi, O. J., & Olaleye, O. (2022). Epidemiology of soil-transmitted helminths in Nigeria: A systematic review and meta-analysis. *Transactions of the Royal Society of Tropical Medicine and Hygiene*, 116(1), 3–13
- Pukuma, M. S., Augustine, M. L., & Enoch, N. (2023). Prevalence of intestinal parasitic infections among internally displaced persons and host communities in Mubi North LGA, Adamawa State, Nigeria. *Journal of Applied Life Sciences International*, 26(3), 23–33
- R Core Team. (2023). R: A language and environment for statistical computing (Version 4.3.2). R Foundation for Statistical Computing. <https://www.R-project.org/>
- Roro, A. G., Kebede, F., & Teshome, H. (2022). Clinic-based study on the prevalence of *Entamoeba histolytica* and *Giardia lamblia* infections in children with diarrhoea. *African Health Sciences*, 22(1), 56–64
- Roro, G. B., Alemayehu, G., & Dires, A. (2022). Prevalence and associated risk factors of *Entamoeba histolytica* infection among schoolchildren. *Frontiers in Public Health*, 10, 976823
- Sarkari, B., Hosseini, G., Motazedian, M.H., Fararouei, M. and Moshfe, A. (2016). Prevalence and risk factors of intestinal protozoan infections: a population-based study in rural areas of Boyer-Ahmad district, Southwestern Iran. *BMC Infectious Diseases*, 16(1):1–5
- Shirley, D. A., Moonah, S. N., & Kotloff, K. L. (2024). Epidemiology and management of intestinal spore-forming protozoan infections. *Current Opinion in Infectious Diseases*, 37(1), 55–63
- Shirley, D.-A. T., Farr, L., & Watanabe, K. (2019). *Entamoeba histolytica* and amoebic liver abscess. *Seminars in Liver Disease*, 39(1), 35–46. <https://doi.org/10.1055/s-0039-1677739>
- Sitotaw, B., Seid, M., & Tigabu, A. (2020). Prevalence and risk factors associated with intestinal parasitic infections: A systematic review. *Journal of Parasitology Research*, 2020, 8681247
- Smith, P. (2013). Sample size determination for large population studies: A practical guide for researchers. *Journal of Statistical Applications and Methods*, 2(1), 1–10
- Stark, D., Barratt, J., Chan, D., and Ellis, J. T. (2016). *Dientamoeba fragilis*, the neglected trichomonad of the human bowel. *Clinical Microbiology Reviews*, 29:553–580

Tyoalumun, K., Abubakar, I., & Okoh, G. R. (2021). Gender-related patterns in intestinal parasitic infections among Nigerian schoolchildren. *African Journal of Infectious Diseases*, 15(1), 23–31

UNICEF. (2024). State of the world's children 2024: For every child, health. United Nations Children's Fund

WHO Diarrheal disease (2017). Available <https://www.who.int/news-room/fact-sheets/detail/diarrheal-disease> Accessed 21 January 2026

World Health Organization & United Nations International Children's Emergency Fund (WHO/UNICEF). (2024). Progress on household drinking water, sanitation and hygiene 2024 update. Geneva: WHO and UNICEF

World Health Organization (WHO). (2023). Intestinal parasitic infections: Global epidemiological update and control strategies. Geneva: WHO Press

World Medical Association (WMA). (2024). Declaration of Helsinki: Ethical principles for medical research involving human subjects (2024 revision). Geneva: World Medical Association

Yohanna, P., Enosh, S., and Bello, A.G. (2016). Temporal change detection of vegetation cover in Mubi metropolis and environs, Adamawa State, Nigeria. *Sky Journal of Soil Science and Environmental Management*, 5(3):59-65

Zemene, E., Shiferaw, M. B., & Tewabe, T. (2018). Prevalence and associated factors of intestinal parasitic infections among school-aged children in Ethiopia. *BMC Infectious Diseases*, 18: 673



©2026 This is an Open Access article distributed under the terms of the Creative Commons Attribution 4.0 International license viewed via <https://creativecommons.org/licenses/by/4.0/> which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is cited appropriately.