

Prevalence, Socio-Demographic Factors and the Relationship Between CD4 Count and Infection with *Cryptosporidium parvum* Among HIV/AIDS Infected Patients with Diarrhea in Jos University Teaching Hospital, Nigeria

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Abstract

Cryptosporidium parvum is a clinically important opportunistic protozoan parasite that causes chronic and severe diarrheal illness in immunocompromised patients, particularly people living with HIV/AIDS. This infection contributes substantially to morbidity and mortality in patients with diminished immune function. The present study investigated the prevalence of *C. parvum* infection, its association with selected socio-demographic factors, and its relationship with CD4 cell counts among HIV/AIDS patients presenting with diarrhea at the Jos University Teaching Hospital (JUTH), Nigeria. A hospital-based cross-sectional study was conducted among 200 HIV-positive patients presenting with diarrhea. Stool samples were collected and examined for *C. parvum* oocysts using the modified Ziehl-Neelsen staining technique, while CD4 counts were retrieved from patient medical records. Socio-demographic and clinical information were gathered through structured questionnaires. Statistical analyses were performed to evaluate associations between *C. parvum* infection, socio-demographic variables, and CD4 counts. Of the 200 samples examined, 22 (11.0%) tested positive for *C. parvum*. Males accounted for 15 (68.0%) positive cases and females for 7 (32.0%), yielding a male-to-female ratio of 2:1. The highest proportion of positive cases (19 cases, 86.4%) occurred among patients with CD4 counts below 200 cells/ μ L. The source of drinking water was a significant determinant, with 81.8% of infected participants consuming untreated water. No statistically significant association was observed between infection and age, place of residence, marital status, frequency of eating outside, or frequency of diarrhea. *Microsporidium* was also detected with a prevalence of 4.0%, and a co-infection prevalence with *C. parvum* of 3.0%. These findings underscore that *C. parvum* remains a clinically relevant opportunistic pathogen among HIV/AIDS patients with diarrhea in Jos, driven largely by immunosuppression and inadequate water treatment. Routine screening for intestinal parasites, improved sanitation, safe water access, and health education are essential strategies to reduce the burden of cryptosporidiosis in this vulnerable population.

Keywords: *Cryptosporidium parvum*; HIV/AIDS; CD4 count; diarrhea; prevalence; socio-demographic factors; opportunistic infection

1. Introduction

Cryptosporidium parvum is an obligate intracellular protozoan parasite that forms environmentally resistant oocysts and colonizes the epithelial lining of the gastrointestinal tract in humans and a broad range of animal hosts (Moore *et al.*, 2007; Ryan *et al.*, 2014). Although the organism produces self-limiting gastroenteritis in immunocompetent individuals, infection in immunocompromised patients, especially people living with HIV/AIDS, can become chronic, debilitating, and potentially fatal (Sharma *et al.*, 2023; Checkley *et al.*, 2015).

The primary mode of transmission follows the fecal-oral pathway, with infection most commonly acquired through ingestion of water or food contaminated by oocysts shed in the feces of infected human or animal hosts. A particularly concerning feature of *C. parvum* oocysts is their remarkable resistance to standard water treatment measures such as chlorination, and the fact that a single oocyst is sufficient to initiate clinical disease (Delling *et al.*, 2017; Efstratiou *et al.*, 2017). Symptomatic infection typically presents as profuse, watery diarrhea accompanied by abdominal pain, nausea, vomiting, and low-grade fever (Mendonça *et al.*, 2025; Adamu *et al.*, 2014).

Clinical and epidemiological evidence has firmly established that the severity of cryptosporidiosis, the risk of prolonged fecal shedding, and the likelihood of complications are inversely correlated with CD4 cell counts (Utami *et al.*, 2020; Ahmadpour *et al.*, 2020). Patients whose CD4 counts fall below 200 cells/ μ L experience the most severe and protracted illness, whereas individuals with CD4 counts above 500 cells/ μ L tend to experience self-limiting disease or remain entirely asymptomatic. Despite universal recognition that *Cryptosporidium* poses a serious threat to immunocompromised populations, reliable prevalence data from resource-limited settings remain scarce (Hunter and Nichols, 2002; Squire and Ryan, 2017).

Among HIV-positive individuals, *Cryptosporidium* ranks as one of the most frequently identified microbial causes of chronic, watery, non-bloody diarrhea, frequently associated with progressive wasting and malabsorption of essential nutrients (Agarwal, 2013; Nsagha *et al.*, 2016). Global surveillance data indicate that cryptosporidiosis has been documented in more than 69 countries (Dong *et al.*, 2020), with substantially higher infection rates in

developing nations where poverty, poor sanitation, unsafe water, and the HIV/AIDS pandemic converge (Semenza and Nichols, 2007; Aldeyarbi *et al.*, 2016). In these environments, opportunistic cryptosporidiosis accelerates HIV disease progression, promotes malabsorption of vitamins, fluids, and electrolytes, and contributes to wasting and death.

Despite isolated reports from various parts of Nigeria, the epidemiology of *C. parvum* among HIV-positive individuals in the country remains insufficiently characterized. The present study was therefore designed to determine the prevalence of *C. parvum* among HIV-positive patients presenting with diarrhea at JUTH, to evaluate the association between selected socio-demographic factors and infection status, and to examine the relationship between CD4 cell counts and the presence of *C. parvum* oocysts in stool samples.

2. Materials and Methods

2.1 Study Area

This study was conducted at the Jos University Teaching Hospital (JUTH), a federal tertiary health institution located in Jos, Plateau State. The hospital serves as a major referral center for the entire North-Central geopolitical zone of Nigeria and provides specialized clinical services for a wide spectrum of medical conditions, including comprehensive HIV/AIDS care.

2.2 Study Population and Design

A hospital-based cross-sectional design was adopted. The study enrolled 200 HIV/AIDS patients aged 18 years and above who presented with diarrhea at JUTH. Randomized sampling was employed to ensure that every eligible participant had an equal probability of selection and that the sample was broadly representative of the patient population attending the facility. HIV-negative patients, HIV-positive patients without diarrhea, and HIV-positive patients with diarrhea below 18 years of age were excluded from the study.

2.3 Ethical Considerations

Ethical clearance was obtained from the Institutional Health Research Ethics Committee of JUTH. Written informed consent was obtained from each participant prior to enrollment. Participants were informed of their right to withdraw from the study at any stage without any consequences to their ongoing medical care. All records and specimens were handled with strict confidentiality.

2.4 Sample Collection

A total of 200 stool samples were collected from the enrolled participants between November and December 2018. Each participant was provided with a clean, wide-mouthed, screw-capped plastic container labeled with unique identification codes. Applicator sticks were supplied for sample collection. A single stool sample was obtained per participant. Structured questionnaires were administered to collect socio-demographic and clinical data, and CD4 count records were retrieved from the hospital laboratory.

2.5 Modified Ziehl-Neelsen Staining Technique

The modified Ziehl-Neelsen staining technique was employed for the laboratory detection of *C. parvum* oocysts. This method exploits the acid-fast property of the parasite's oocyst wall, which retains the carbol fuchsin dye even after treatment with acid alcohol. Fecal smears were prepared directly from each sample, allowed to air dry, and then fixed in methanol for three minutes. The smears were stained with strong carbol fuchsin for 15 to 20 minutes, rinsed thoroughly with tap water, decolorized in acid alcohol (1% hydrochloric acid in methanol) for 15 to 20 seconds, rinsed again, and counterstained with 0.4% malachite green. After final rinsing and air drying, stained smears were examined under oil immersion at 100x magnification for the presence of *C. parvum* oocysts, which appeared as round, pinkish to purple structures measuring approximately 4 to 6 µm in diameter (Cheesbrough, 2006; Garcia, 2016).

2.6 Statistical Analysis

Data were analyzed using SPSS version 22. Chi-square tests were employed to assess the associations between *C. parvum* infection and the socio-demographic and clinical variables of the study participants. A *p*-value of less than 0.05 was considered statistically significant. Results were summarized in tables and figures.

3. Results

Of the 200 stool samples analyzed, 22 (11.0%) tested positive for *C. parvum* while 178 (89.0%) were negative. Males accounted for 15 (68.0%) positive cases and females for 7 (32.0%), yielding a male-to-female infection ratio of approximately 2:1 (Table 1; Figure 1).

Table 1. Prevalence of *C. parvum* among study subjects in relation to sex

Sex	No. Tested	No. Positive (%)
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Male	137	15 (10.95)
Female	63	7 (11.11)
Total	200	22 (11.00)

$p = 0.001$; $\chi^2 = 0.97$; $df = 1$

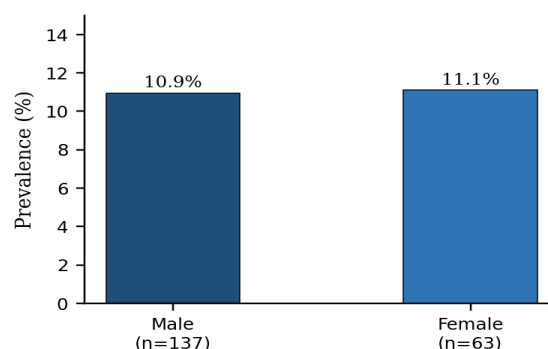


Figure 1. Prevalence of *C. parvum* infection by sex among HIV/AIDS patients at JUTH

The distribution of *C. parvum* infection across socio-demographic categories is presented in Table 2. Among age groups, participants aged 31 to 35 years and those older than 40 years recorded the highest proportions of positive cases (36.4% each), whereas those younger than 30 years had the lowest (4.5%). Urban residents accounted for 86.4% of positive cases, compared to 13.6% among rural residents. Married participants constituted 90.9% of positive cases. Source of drinking water emerged as a statistically significant factor ($p = 0.003$), with 81.8% of infected participants reporting consumption of untreated water (Figure 3).

Table 2. Distribution of *C. parvum* by socio-demographic factors

Characteristic	Tested	Pos.	%	X ²	p
Age (Years)					
<30	9	1	4.50		
31–35	82	8	36.40		
36–40	44	5	22.70	3.443	0.66
>40	65	8	36.40		
Residence					
Rural	30	3	13.60		
Urban	170	19	86.40	0.036	0.85
Marital Status					
Single	36	2	9.10		
Married	164	20	90.90	1.329	0.25
Source of Water					
Treated	10	4	18.20		
Untreated	190	18	81.80	9.049	0.003

Eating Outside					
Often	21	5	22.70		
Sometimes	151	13	59.10	4.710	0.10
Rarely	28	4	18.20		

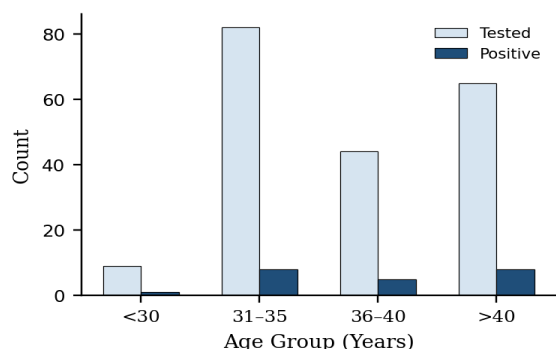


Figure 2. Distribution of *C. parvum* infection by age group

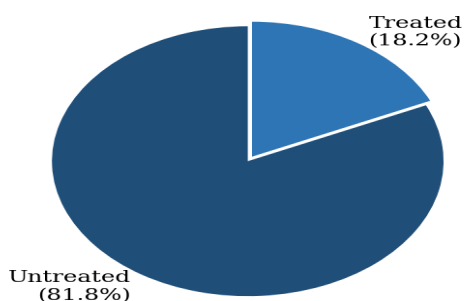


Figure 3. Proportion of *C. parvum* positive cases by source of drinking water

Table 3. Distribution of *C. parvum* in relation to frequency of diarrhea per day

Frequency/day	Tested	Positive	%
One	34	3	13.60
Three	61	7	33.30
Seven	105	12	54.50
Total	200	22	100

$\chi^2 = 0.198$; $df = 2$; $p = 0.906$

Table 4. Distribution of *C. parvum* in relation to CD4 count (CDC classification)

CD4 (cells/ μ L)	Tested	Positive	%
<200	171	19	86.40
200–499	29	3	13.60
>500	0	0	0.00
Total	200	22	100

$\chi^2 = 0.477$; $df = 2$; $p = 0.63$

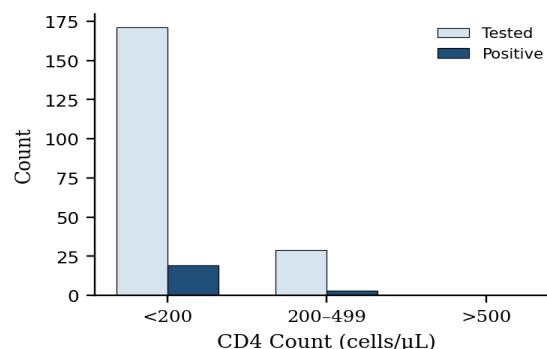


Figure 4. Distribution of *C. parvum* infection by CD4 count category

Table 5. Co-infection of *C. parvum* and *Microsporidium* at JUTH

Parasite	Tested	Positive	%
Microsporidium	200	8	4.00
Microsporidium + <i>C. parvum</i>	200	6	3.00

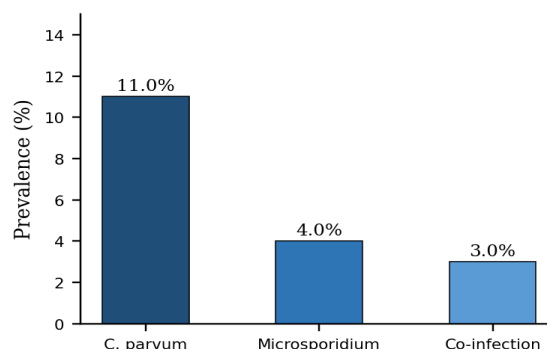


Figure 5. Comparative prevalence of *C. parvum*, *Microsporidium*, and co-infection

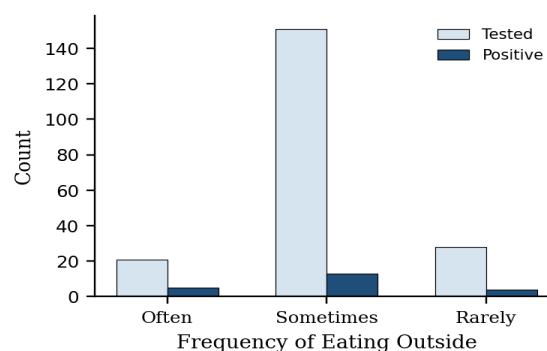


Figure 6. Distribution of *C. parvum* infection by frequency of eating outside the home

4. Discussion

Cryptosporidium parvum is widely acknowledged as one of the most common causes of acute and chronic diarrheal disease among people living with HIV/AIDS, and the clinical severity of the infection intensifies as the patient's

immune function deteriorates (Checkley *et al.*, 2015; Ahmadpour *et al.*, 2020). In low- and middle-income countries, cryptosporidiosis persists as an opportunistic infection of considerable public health significance because of its strong association with dehydration, progressive weight loss, and elevated morbidity among both children and adults who are immunocompromised (Mohammed *et al.*, 2017; Kotloff *et al.*, 2013). Clinical guidelines governing the management of opportunistic infections in HIV-infected populations consistently emphasize cryptosporidiosis as a recurrent clinical challenge and highlight the critical importance of immune restoration through effective antiretroviral therapy as a primary strategy for disease control (Laksemi *et al.*, 2020; Ghoshal *et al.*, 2018).

The 11.0% prevalence of *C. parvum* infection documented in this study is broadly consistent with findings from other parts of Nigeria and sub-Saharan Africa. Akinbo *et al.* (2010) reported a prevalence of 11.6% among HIV-positive patients in Benin City using a similar diagnostic approach, a figure that closely mirrors the present observation. Lower prevalence rates have been documented elsewhere in Nigeria: Anejo-Okopi *et al.* (2014) reported 4.7% at the same JUTH facility using formol-ether concentration combined with modified Ziehl-Neelsen staining; Erhabor *et al.* (2011) documented 2.9% in the Niger Delta; and Kumurya and Gwarzo (2013) recorded 4.0% in Kano State. In contrast, considerably higher rates have been reported in select settings, with Adesiji *et al.* (2007) documenting 79.3% in Oyo State and Akujobi *et al.* (2005) reporting 18.7% in Lagos.

These substantial variations in reported prevalence across studies likely reflect differences in diagnostic methodology, geographical location, seasonal factors, sample size, clinical staging of HIV disease, and access to antiretroviral therapy. When more sensitive diagnostic tools such as enzyme-linked immunosorbent assays or polymerase chain reaction-based methods are employed, higher prevalence figures are typically observed (Squire and Ryan, 2017; Wumba *et al.*, 2015). The reliance on microscopy-based detection in the present study, while practical and widely available, may therefore underestimate the true burden of infection in this population. The fluctuation of *C. parvum* prevalence over time and across different locations can also be attributed to variable levels of water and food contamination,

differential exposure to animal reservoirs, and differences in public health infrastructure (Arnone and Walling, 2006; Efstratiou *et al.*, 2017).

As Figure 1 illustrates, the prevalence rates between males (10.95%) and females (11.11%) were nearly equivalent when considered as a proportion of the number tested within each sex group, though males constituted a larger absolute number of positive cases (15 versus 7) owing to the skewed sex distribution of the study sample. This pattern is consistent with the findings of Akujobi and Ogunsola (2008), who reported a higher absolute prevalence in males in their Lagos-based study. The higher absolute number of infections among males may be explained by greater occupational exposure to environmental sources of contamination, including contact with farm animals, swimming in public pools, and consumption of untreated water from diverse sources.

The observation that *C. parvum* prevalence was higher among urban residents (86.4% of positive cases) compared to those from rural areas (13.6%) aligns with findings from Nepal (Amatya *et al.*, 2011) and Kenya (Nyamwange, 2013). Although rural areas are often associated with closer contact with livestock and poorer sanitation, urban environments in developing countries present their own distinctive challenges. These include overcrowded living conditions, reliance on shared and potentially contaminated water distribution systems, and frequent consumption of food from street vendors whose hygiene practices may be inadequate. Notably, Anejo-Okopi *et al.* (2014) documented higher rural prevalence (8.3%) than urban prevalence (3.0%), suggesting that the urban-rural distribution of cryptosporidiosis is context-dependent and shaped by local water supply conditions, sanitation infrastructure, and population density (Samie *et al.*, 2006; Efstratiou *et al.*, 2017).

No statistically significant association was identified between age and *C. parvum* infection ($\chi^2 = 3.443$, $p = 0.66$), which is consistent with reports from Uganda (Salyer *et al.*, 2012) and Venezuela (Certad *et al.*, 2007). As Figure 2 demonstrates, participants in the 31 to 35 year and above 40 year age brackets accounted for the highest proportions of positive cases, which may reflect occupational exposures, cumulative immunosuppression, or compromised personal hygiene in these groups (Nsagha *et al.*, 2016). Likewise, no significant association was found between marital status and infection, although

married individuals comprised the overwhelming majority of positive cases (90.9%). This likely reflects the demographic composition of the study sample rather than a genuine risk difference. Nonetheless, it bears noting that intimate behaviors involving oral-anal contact can facilitate direct person-to-person transmission of *C. parvum* in certain populations (Jamil *et al.*, 2023; Wumba *et al.*, 2015).

Source of drinking water emerged as the only statistically significant socio-demographic determinant of *C. parvum* infection in this study ($\chi^2 = 9.049$, $p = 0.003$). As depicted in Figure 3, an overwhelming 81.8% of infected participants consumed untreated water, compared to just 18.2% among those who treated their water before drinking. This finding aligns with the well-established role of contaminated water in the transmission of *Cryptosporidium* oocysts, which withstand chlorination but can be inactivated by boiling, ultraviolet irradiation, and ultrasonic treatment (Olvera *et al.*, 2008; Efstratiou *et al.*, 2017). Sucilathangam *et al.* (2012) similarly documented a strong relationship between untreated water consumption and *C. parvum* infection. The persistence of *Cryptosporidium* oocysts in chlorinated drinking water and recreational pools further underscores the need for water treatment strategies that go beyond chlorination alone (Schets *et al.*, 2007; Semenza and Nichols, 2007).

No significant association was detected between eating outside the home and *C. parvum* infection ($\chi^2 = 4.710$, $p = 0.10$), which is consistent with findings by Ajjampur *et al.* (2007), who identified animal contact and improper food storage as the principal risk factors for infection rather than restaurant patronage. However, as Figure 6 shows, participants who ate outside occasionally constituted the largest proportion of positive cases (59.1%), a pattern that warrants further investigation with larger sample sizes and more precise dietary exposure assessments.

No statistically significant relationship was observed between diarrhea frequency and *C. parvum* infection ($\chi^2 = 0.198$, $p = 0.906$). This contrasts with Girma *et al.* (2014), who reported a strong association between diarrhea duration and *C. parvum* detection in an Ethiopian cohort. The discrepancy may stem from differences in diarrhea definitions, HIV disease staging, availability of antiretroviral therapy, and the co-occurrence of other enteric pathogens in the respective study populations.

The relationship between CD4 cell counts and *C. parvum* infection in this study reveals a clear pattern of heightened susceptibility at lower immune thresholds. As Table 4 and Figure 4 demonstrate, the vast majority of positive cases (86.4%) were concentrated among patients with CD4 counts below 200 cells/ μ L, while only 13.6% occurred among those with counts between 200 and 499 cells/ μ L. No infections were detected in patients with CD4 counts exceeding 500 cells/ μ L. These observations are consistent with established immunological principles and corroborate the findings of Askira *et al.* (2022) and Ahmadpour *et al.* (2020), who demonstrated that advanced immunosuppression, as indicated by severely depleted CD4 cell counts, constitutes the foremost risk factor for opportunistic *Cryptosporidium* infection in HIV-positive populations. The strong inverse relationship between CD4 counts and susceptibility to cryptosporidiosis reinforces the critical importance of early initiation and sustained adherence to antiretroviral therapy, which has been shown to restore immune function and dramatically reduce the incidence of opportunistic parasitic infections (Laksemi *et al.*, 2020; Ghoshal *et al.*, 2018).

The identification of *Microsporidium* alongside *C. parvum*, although incidental to the primary study objective, is an observation of considerable clinical importance (Figure 5). The prevalence of *Microsporidium* (4.0%) and the co-infection rate with *C. parvum* (3.0%) reflect the well-documented phenomenon of overlapping opportunistic parasitic infections in immunocompromised hosts. Both organisms share common fecal-oral transmission pathways and preferentially colonize individuals with advanced HIV disease. Tumwine *et al.* (2005) reported substantially higher rates of both parasites among Ugandan children, and Nkenfou *et al.* (2013) documented a co-infection rate of 1.9% in Cameroon. The co-occurrence of these parasites in the same clinical population highlights the necessity for comprehensive parasitological screening protocols that target multiple opportunistic agents rather than focusing on a single pathogen (Akoolo *et al.*, 2022; Ghoshal *et al.*, 2018).

Several limitations of this study warrant acknowledgment. The reliance on microscopy alone for parasite detection, while practical and widely accessible, may have underestimated the true prevalence compared to molecular diagnostic approaches. The cross-sectional design precludes the establishment of causal relationships

between the identified variables and infection status. Furthermore, the sample was drawn from a single tertiary health facility, which may limit the generalizability of these findings to other settings and populations. Future investigations should incorporate molecular diagnostic tools, adopt longitudinal study designs, and recruit participants from multiple clinical centers to provide a more thorough understanding of the epidemiology of *C. parvum* in the Nigerian context (Squire and Ryan, 2017; Aldeyarbi *et al.*, 2016).

5. Conclusion

The findings of this study demonstrate that *C. parvum* remains a clinically important opportunistic parasite contributing to chronic diarrheal illness among HIV/AIDS patients at JUTH. The overall prevalence of 11.0% confirms that cryptosporidiosis is not uncommon in this clinical setting. The statistically significant association between untreated water consumption and infection highlights the central role of water, sanitation, and hygiene interventions in reducing transmission. The pronounced inverse relationship between CD4 cell counts and infection underscores the critical importance of immune restoration through antiretroviral therapy as a frontline strategy for preventing opportunistic parasitic infections.

Routine screening for intestinal parasites, particularly among patients with CD4 counts below 200 cells/ μ L and those presenting with persistent diarrhea, should be incorporated into standard HIV management protocols. Health education initiatives promoting safe water practices, proper food hygiene, and regular handwashing are essential for reducing community-level exposure to *C. parvum* and other fecal-oral pathogens. Strengthening adherence to antiretroviral therapy, improving access to clean drinking water, and implementing comprehensive parasitological surveillance will collectively contribute to reducing the burden of cryptosporidiosis and improving the quality of life of affected individuals.

Acknowledgments

The authors gratefully acknowledge the management and staff of the Jos University Teaching Hospital for their cooperation and logistical support throughout the conduct of this study. The voluntary participation and informed consent of all study subjects are deeply appreciated.

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