

## Prevalence of Intestinal Coccidiosis in HIV-Infected Patients

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**Received:** April 30, 2026; **Published:** June 03, 2026

### Abstract

**Introduction:** Intestinal coccidiosis is a common opportunistic parasitic infection among patients living with HIV and represents an important cause of digestive morbidity, particularly in low-resource settings. It is mainly caused by *Cryptosporidium* spp., *Cystoisospora belli* and *Cyclospora cayetanensis*, which are transmitted through the fecal-oral route. In HIV-infected patients, these infections may present with acute or chronic diarrhea, sometimes severe and prolonged, leading to dehydration, malabsorption and weight loss. Despite advances in HIV management, intestinal coccidiosis remains frequent and often underdiagnosed. Stool parasitological examination remains essential for its diagnosis.

**Materials and Methods:** A descriptive study was conducted from January 2022 to November 2025 in the Parasitology Laboratory of Hassan II University Hospital in Fez. HIV-positive patients who underwent stool parasitological examination, either in screening context or because of diarrhea, were included. Diagnosis relied on direct microscopic examination, concentration techniques (Ritchie method) and modified Ziehl-Neelsen staining for identification of intestinal coccidia.

**Results:** A total of 31 HIV-positive patients were included, with a mean age of 38.5 years and a female predominance (58%). Stool examination was mainly performed as part of screening (74%). Intestinal coccidia were detected in 25 patients, corresponding to an overall prevalence of 80.6%, with comparable positivity rates between diarrheic patients and those investigated for screening.

**Conclusion:** Intestinal coccidiosis remains frequent among HIV-infected patients and constitutes a significant cause of digestive morbidity. Stool parasitological examination remains indispensable for diagnosis, particularly in low-resource settings. These findings highlight the crucial role of parasitology laboratories in screening and managing HIV patients.

**Keywords:** *Cryptosporidiosis; Cyclosporiasis; HIV; Intestinal coccidiosis; Stool Parasitological Examination*

### Introduction

Intestinal coccidiosis, primarily caused by *Cryptosporidium* spp., *Cystoisospora belli*, and *Cyclospora cayetanensis*, represents a common opportunistic parasitic infection in patients infected with the human immunodeficiency virus (HIV) and constitutes a significant cause of gastrointestinal morbidity [1-3]. These infections are responsible for acute or chronic diarrhea, which may progress to severe forms associated with dehydration, malabsorption, and deterioration of general health status, particularly in immunocompromised individuals [4,5].

Despite improved access to antiretroviral therapy, opportunistic intestinal parasitic infections persist in many resource-limited settings due to environmental, socioeconomic, and diagnostic factors [6,7]. Several studies have reported a high prevalence of intestinal coccidiosis among HIV-infected patients, with variations depending on geographic region, clinical context, and diagnostic methods used [8-10].

The diagnosis of these infections relies primarily on stool parasitological examination, the performance of which is enhanced by the use of concentration techniques and specific staining methods, particularly the Ritchie technique and modified Ziehl-Neelsen staining [11-13]. In Morocco, recent hospital-based data on the prevalence of intestinal coccidiosis in HIV-infected patients remain limited.

### Aim of the Study

The aim of this study was to evaluate the prevalence of intestinal coccidiosis among HIV-positive patients referred to the Parasitology Laboratory of Hassan II University Hospital in Fez and to describe their clinical characteristics.

### Materials and Methods

#### Study design and period

This was a descriptive cross-sectional study conducted from January 2022 to November 2025 at the Parasitology Laboratory of Hassan II University Hospital in Fez.

#### Study population

The study included 31 HIV-positive patients who underwent stool parasitological examination, prescribed either as part of systematic screening or in the context of diarrhea.

#### Data collection

The variables analyzed included sex, age, the indication for stool examination (screening or diarrhea), and the presence or absence of intestinal coccidian oocysts in stool samples. Additional data included the distribution according to the identified coccidian species, possible co-infection with other intestinal parasites, and immunological parameters such as CD4 lymphocyte count, viral load, stage of HIV infection, and current antiretroviral therapy.

#### Parasitological methods

Stool parasitological examination was based on:

- Direct microscopic examination of fresh stool samples, the Ritchie concentration technique, and modified Ziehl-Neelsen staining.
- Direct examination allowed the detection of associated intestinal parasitic infections, although it does not permit identification of *Cryptosporidium* and *Cyclospora* oocysts, which are morphologically similar to yeasts.
- The Ritchie technique, a widely used physicochemical method, was employed to concentrate parasites and improve detection sensitivity.
- Modified Ziehl-Neelsen staining was the reference method for detecting coccidian oocysts on stool smears, with or without prior concentration. Observed oocysts were round to oval and stained red or pink against a blue-green background.

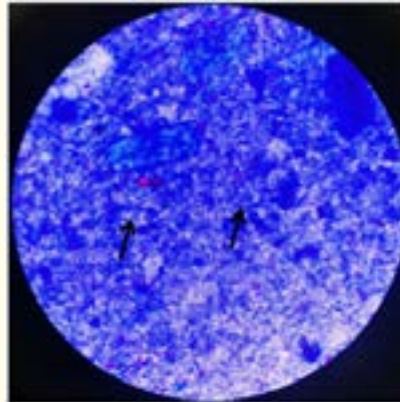
#### Statistical analysis

Data were analyzed descriptively. Qualitative variables were expressed as frequencies and percentages, and age as a mean value. Prevalence was calculated by dividing the number of positive cases by the total number of patients included.

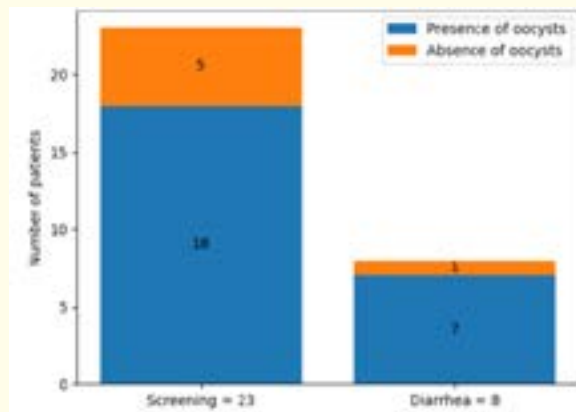
**Results**

Among the 31 patients included, 18 were female (58%) and 13 male (42%), with a mean age of 38.5 years. Stool parasitological examination was requested as part of screening in 23 patients (74%) and for the investigation of diarrhea in 8 patients (26%) (Table 1).

Intestinal coccidian oocysts were detected in 25 patients, corresponding to an overall prevalence of 81%. Positivity was observed in 87.5% of patients presenting with diarrhea (7/8) and in 78.5% of those screened systematically (18/23) (Table 1).



**Figure 1:** Microscopic appearance of stool samples stained using the modified Ziehl-Neelsen technique, showing coccidian oocysts (*Cryptosporidium* spp. and *Cyclospora* spp.) (×100).



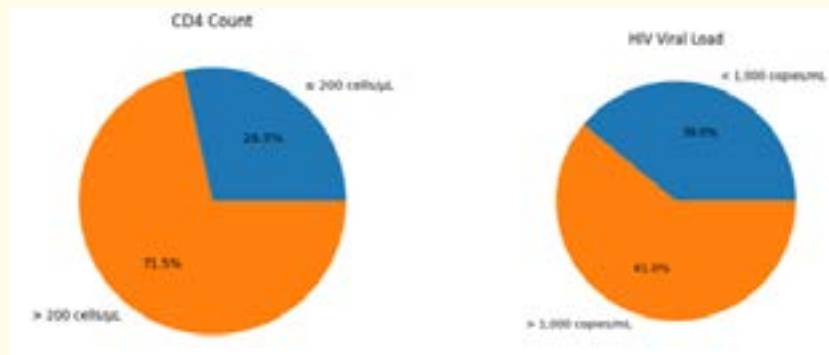
**Figure 2:** Distribution of patients according to indication and presence of oocyst.

In our series of 31 HIV-infected patients who underwent stool parasitological examination, the identified intestinal coccidia were predominantly *Cryptosporidium* spp. and *Cyclospora* spp. Co-infection with both parasites was observed in 24 patients (77%). *Cryptosporidium* spp. alone was detected in 5 patients (16%), whereas *Cyclospora* spp. alone was identified in 2 patients (7%). In addition, co-infection with non-opportunistic protozoa was observed in nearly one-third of patients, mainly involving cysts of *Entamoeba histolytica/dispar* and *Endolimax nana* (Table 1).

Regarding immunovirological parameters, three patients included in the study (n = 31) were transferred to another healthcare facility for further management, reducing the immunological and virological analysis to 28 patients. Among these, 8 had a CD4 count  $\leq$  200 cells/ $\mu$ L, while 20 had a CD4 count  $>$  200 cells/ $\mu$ L. Concerning HIV viral load, a high viral load (defined as  $>$  1,000 copies/mL) was observed in 19 patients, whereas the remaining patients had a controlled viral load ( $<$  1,000 copies/mL). Among the 28 patients followed in our center, five deaths were reported. In these five patients, the CD4 count was  $\leq$  200 cells/ $\mu$ L and the viral load was  $>$  1,000 copies/mL. It should be noted that all patients included in the study were receiving antiretroviral therapy at the time of investigation (Table 1).

Variables	Number (n)	Percentage (%)
<b>Sex</b>		
Female	18	58
Male	13	42
<b>Indication for stool examination</b>		
Screening	23	74
Diarrhea	8	26
<b>Type of coccidiosis</b>		
Co-infection (Cryptosporidium + Cyclospora)	24	77
Cryptosporidium alone	5	16
Cyclospora alone	2	7
<b>Parasitic association</b>		
Present (Entamoeba histolytica/dyspar, Endolimax nana)	10	33.3
Absent	21	66.7
<b>Immuno-virological parameters</b>		
<b>CD4 count</b>		
$\leq$ 200 cells/ $\mu$ L	8	28.5
$>$ 200 cells/ $\mu$ L	20	71.5
<b>HIV viral load</b>		
$<$ 1,000 copies/mL	12	39
$>$ 1,000 copies/mL	19	61

**Table 1:** Clinical, parasitological, and immunovirological characteristics of the HIV-positive patients included in the study.



**Figure 3:** Distribution of immunovirological parameters.

### Discussion

The present study highlights a very high prevalence of intestinal coccidiosis (81.5%) among HIV-seropositive patients referred to the Parasitology Laboratory of the Hassan II University Hospital in Fez between 2022 and 2025. This finding confirms that intestinal coccidiosis remains a major opportunistic parasitic infection in people living with HIV, as reported in several African and international studies [1-3,8]. This high frequency is consistent with trends observed in low-resource countries, where opportunistic gastrointestinal parasitic infections remain common among patients with advanced immunosuppression.

In Morocco, Badaoui, *et al.* reported that opportunistic digestive parasitic infections were the presenting feature of HIV infection in 77% of cases in a hospital-based cohort at Ibn Rochd University Hospital in Casablanca, among severely immunocompromised patients [23]. Similarly, in Tunisia, intestinal coccidiosis accounts for 70% of diarrheal episodes in HIV-infected patients [19], while prevalences of 86% and 79% have been reported in Nigeria and Uganda, respectively, among symptomatic or diarrheic HIV patients [20,21]. In contrast, lower rates are generally observed in industrialized countries; in France, cryptosporidiosis prevalence has been estimated at 17.7% among cases reported in recent hospital series of high-risk immunocompromised patients [22]. Despite advances in the overall management of HIV infection, these gastrointestinal infections persist and remain a major public health concern, particularly in resource-limited hospital settings.

A key contribution of this study is the high detection rate of intestinal coccidia among patients investigated in a screening context, with a positivity rate of 80.6%, closely approaching that observed in patients presenting with diarrhea (87%). These findings are consistent with the literature showing that intestinal coccidiosis is not exclusively associated with diarrheal syndromes and may occur in asymptomatic or paucisymptomatic forms in HIV-infected patients [6,10]. Therefore, the absence of diarrhea does not exclude these infections, reinforcing the need for systematic stool parasitological screening in this at-risk population.

In the context of diarrhea, the high proportion of positive cases observed in our series confirms the major etiological role of intestinal coccidiosis in HIV-associated diarrhea, as widely documented in the literature [4,5,9]. These infections should be systematically investigated in cases of persistent or unexplained diarrhea in HIV-infected patients to avoid diagnostic delays that may worsen digestive morbidity and overall patient condition.

Our results particularly emphasize the central role of stool parasitological examination in the diagnosis and screening of intestinal coccidiosis. The combined use of direct microscopic examination, concentration techniques-particularly the Ritchie method-and modified Ziehl-Neelsen staining optimized the detection of intestinal coccidian oocysts, in agreement with methodological data reported in the literature [11-14]. These techniques represent first-line diagnostic tools that are accessible, reproducible, and suitable for routine use in hospital laboratories.

Concentration methods are essential, as direct examination alone may have limited sensitivity, particularly when parasite load is low or when oocyst excretion is intermittent, a situation frequently observed in asymptomatic or paucisymptomatic patients [11,12]. The Ritchie technique significantly increases detection rates by concentrating parasitic elements and reducing fecal debris. Similarly, modified Ziehl-Neelsen staining remains crucial for reliable identification of intestinal coccidia, whose oocysts may be missed without specific staining [13,14].

Several studies have demonstrated a strong association between intestinal coccidiosis in HIV-infected patients and impaired immunovirological control. Indeed, profound immunosuppression characterized by CD4 counts below 200 cells/mm<sup>3</sup> is a major risk factor for cryptosporidiosis and other intestinal opportunistic infections. In addition, uncontrolled viral load reflects active viral replication and worsens immune dysfunction, thereby facilitating the emergence and persistence of intestinal coccidia. Sarfo, *et al.* observed an inverse correlation between HIV viral load and *Cryptosporidium* spp. burden, suggesting that lack of viral suppression contributes to

higher parasite loads [15]. Dereb, *et al.* reported that the combination of low CD4 counts and detectable viral load was significantly associated with intestinal opportunistic parasitic infections [16], while Otero-Rodríguez, *et al.* confirmed a high frequency of intestinal protozoa in patients with poor virological control [17]. These findings are consistent with our results. Collectively, these data highlight the importance of maintaining adequate CD4 levels and achieving viral suppression to reduce susceptibility to intestinal coccidiosis in HIV-infected patients and reinforce the central role of antiretroviral therapy in preventing opportunistic infections.

In resource-limited settings, where access to immunological or molecular techniques remains restricted, stool parasitological examination retains a central role in the diagnostic strategy for gastrointestinal opportunistic infections in HIV patients [14,18]. Optimization of conventional methods through a rigorous and standardized parasitological approach represents a practical and effective strategy to improve detection of intestinal coccidiosis for both diagnostic and screening purposes.

Although this is a single-center study with a limited sample size, it provides recent local data and highlights the strategic importance of the parasitology laboratory in the overall management of HIV-infected patients. The high proportion of cases detected outside a diarrheal context underscores the value of incorporating routine stool parasitological examination into HIV patient follow-up, regardless of digestive symptoms.

### Conclusion

Intestinal coccidiosis remains highly prevalent among HIV-seropositive patients followed at Hassan II University Hospital in Fez, with a high prevalence observed both in symptomatic patients and in those investigated in a screening setting. These findings show that the absence of diarrhea does not exclude coccidial infection and emphasize the importance of systematic parasitological screening in HIV-infected patients. Stool parasitological examination, based on the combination of direct microscopy, concentration methods, and specific staining techniques, remains an essential, accessible, and routine diagnostic tool. Its systematic integration into HIV patient management is essential to improve early diagnosis and reduce the digestive morbidity associated with intestinal coccidiosis.

### Funding Sources

No funding was received for this study.

### Acknowledgments

The authors would like to thank the medical and technical staff of the Parasitology and Mycology Department for their support in the microbiological analyses.

### Conflicts of Interest

The authors declare no conflicts of interest.

### Authorship Statement

All authors have read and approved the final version of the manuscript. They all meet the authorship criteria as per the journal's guidelines and confirm that this manuscript represents honest and original work.

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**Volume 22 Issue 6 June 2026**

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