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# Prevalence of Intestinal Parasites Among HIV Infected and HIV Uninfected Patients Treated at the 1° De Maio Health Centre in Maputo, Mozambique

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## Abstract

**Introduction—**Increased evidence suggests intestinal parasite infections, one of the major causes of morbidity and mortality in sub-Saharan Africa, increase the acquisition and progression of AIDS.

**Objective—**The aim of this study was to determine the prevalence of HIV and intestinal parasite co-infections, the relationship to the degree of immunosuppression and the effect of antiretroviral treatment (ART) and trimethoprim-sulfamethoxazole (TS) on patients treated at 1° de Maio Health Centre in Maputo, Mozambique.

**Methods—**A cross sectional study was conducted from December 2015 to August 2016. A total of 517 stool samples from 371 (71.8%) HIV infected and 146 (28.2%) HIV uninfected patients were examined for the presence of parasites using direct wet mount, Ritchie and modified Ziehl Neelsen techniques. A subsample of 201 stools from HIV infected patients was processed for coproantigens for the detection of *Cryptosporidium* spp.

**Results—**Overall, 148 (28.6%) of the individuals were infected with at least one parasite. The prevalence of intestinal parasites was 98 (26.4%) and 50 (34.2%) in HIV infected and uninfected patients, respectively. This difference was not statistically significant. We identified 10 different parasites including (most frequently) *Trichuris trichiura* 67 (12.9%), *Ascaris lumbricoides* 27 (5.2%) and *Entamoeba coli* 40 (7.7%). *Giardia intestinalis* prevalence was significantly higher in HIV infected patients 12 (3.2%), p = 0.02. Parasitic intensity was higher in HIV infected patients

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**Conflict of Interest**

The authors declare no conflict of interests.

than in HIV uninfected patients. *Cryptosporidium* spp. prevalence by coproantigen detection was 6% and was associated with degree of immune suppression. A $CD4^+$ T-cell count of < 200 cells/μL was significantly associated with higher prevalence and intensity of parasitism, while ART and TS prophylaxis was associated with lower parasitic prevalence.

**Conclusions—**Our study revealed that the prevalence and intensity of intestinal parasites in HIV infected patients was related to the degree of immune suppression as assessed by $CD4^+$ cell count, while ART and TS seemed to reduce the parasitic infection.

### Keywords

Co-Infection HIV intestinal parasites; Helminthes; Protozoan; Coccidiae

## Introduction

Intestinal parasites, including helminthes and protozoa are neglected tropical diseases (NTDs), targeted by the sustainable development goals (SDGs) to be eliminated by 2030. They constitute a major cause of morbidity and mortality throughout the world, particularly in resource limited tropical and subtropical regions, including sub-Saharan Africa (SSA) [1,2]. The most important intestinal helminthes are the soil transmitted helminthes (STHs), such as *Ascaris lumbricoides, Trichuris Trichiura, Strongyloides stercoralis* and hookworms. It is estimated that approximately 2 billion (24%) of the world's population is infected with intestinal helminthes [2,3]. Less is known about the burden of protozoal infections such as *Entamoeba histolytica, Giardia intestinalis, Cryptosporidium* spp. and *Cystoisospora belli* because of the sensitivity of available diagnostic techniques, the paucity of microscopic skills and a general lack of awareness of protozoan infection [4].

In SSA, the parasitic infection rate is remarkably high with some areas reporting 95% incidence as compared to developed countries were the incidence is 50%. In addition, parasitic infections overlap with the regions of high prevalence of human immune deficiency virus (HIV). In the WHO African Region including Mozambique, 11.4 million are infected with intestinal parasites [1,3,5–7]. The distribution and prevalence of STHs is closely associated with a lack of personal and environmental hygiene; infection can be asymptomatic or symptomatic. When symptomatic and depending on the parasitic agent involved, in addition to degree of immune suppression, individuals may present with chronic diarrhea, malabsorption syndrome, dehydration and anemia [4,8,9].

It has been hypothesized that helminthic infections are an important driver of HIV infection in Africa, due to their effect on the immune system, as demonstrated by the increased susceptibility to HIV infection and the clinical progression to AIDS [9–12]. Thus, control of intestinal parasites is an important tool to combat the HIV pandemic and the vicious cycle of poverty, to reduce inequity in alignment with Universal Health Coverage and, ultimately, to achieve the SDGs [2].

Studies aiming to address the prevalence of intestinal parasite and HIV co-infection and its relationship to antiretroviral treatment (ART) are controversial [7,13,14]. Some indicate that parasitic infections are reduced in HIV infected persons on ART, but the underlying

mechanisms are not known. Recovery of the immune system on ART may explain this phenomena, especially in protozoan infection, as noted in the case of *Cryptosporidium* spp. and *Cystoisospora belli* infection [15–17]. Other authors speculate that the reduction in prevalence of intestinal parasites in HIV patients on ART might be due to the anti-parasitic effects of ART drugs themselves or of trimethoprimsulfamethoxazole (TS) used as a prophylactic agent for several opportunistic infections such as *Toxoplasma gondii, Pneumocystis jirovecii, C. belli*, and *Cyclospora cayetanensis*, [5,15,17].

Mozambique represents one of the most affected countries by HIV with an adult prevalence of 11.5%. No recent or systematic data are available that pertain to co-infection with intestinal parasites and HIV [18].

Of the few published studies, there is a wide range of prevalence and distribution of helminthes and protozoa within the country [4,19–22]. *T. trichiura* prevalence varied from 36.06% to 93%, *A. lumbricoides* prevalence varied from 35.69% to 56%, *S. stercoralis* varied from 5.5% to 48%, and hookworm prevalence varied from 1.86% to 38%. The reported prevalence of some protozoan also varies greatly according to the studied population, geographical region, environmental factors and clinical status of the patient. Microscopic and molecular studies done in Mozambique reported *Giardia intestinalis* prevalence varying from 5.6% to 37%, *E. histolytica/E.dispar* from 4.83% to 10%, *Entamoeba coli* prevalence varying from 10.41% to 34%, and for *Cryptosporidium* spp. prevalence from 2.5% to and 9%.

The present study was undertaken to determine the prevalence of intestinal parasites in HIV infected and uninfected individuals treated at the 1° de Maio Health Center in Maputo city, Mozambique. In addition, this study aims to explore the relationship between the degree of immune suppression as measured by $CD4^+$ cell counts, as well as the effect of TS and ART on the prevalence of intestinal parasites in the HIV-infected patient population.

## Materials and Methods

### Study Design and Population

We conducted a cross sectional study at the 1° de Maio Health Centre, located in Maputo city from December 2015 to August 2016. The National Bioethics Committee of Mozambique approved the study.

Patients that sought or were in care for HIV infection and other illnesses were approached consecutively about their participation in the study. Prior to enrollment, either the principal investigator, or a research nurse explained the aim of the study and invited their participation.

Volunteers who agreed to participate and provided a written informed consent were enrolled into the study. In the case of children, a consent to include them in the study was obtained from their parents or guardians.

Demographic and clinical data including gender, age, source of drinking water, type of sewage, anorexia, diarrhea, HIV status, $CD4^+$ cell counts and ART (in the case of HIV

infected patients) were recorded in a questionnaire designed for this study. The use of selected concomitant medications such as albendazole, mebendazole, metronidazole and TS were also recorded.

Consenting patients were asked to provide one stool sample in a coded sterile bottle that was send to Parasitology Laboratory at the Faculty of Medicine - UEM, in Maputo city, for further processing. Once in the Parasitology laboratory the stool was divided into two aliquots for microscopic examination and *Cryptosporidium* spp. coproantigen detection.

### Parasitological Analysis Methods

One aliquot for each stool sample was examined the same day using light microscopy by simple wet preparation to search for trophozoites of protozoan and within 48 hours for the detection of helminthes and protozoan using formalin-ether sedimentation (10%) concentration technique procedures and iodine staining [23]. For the detection of *Cryptosporidium* spp., *C.belli* and *Cyclospora cayetanensis*. oocysts, the stool samples were also analyzed by modified Ziehl-Neelsen staining [24] (Figure 1).

Parasitological intensity was assessed using a semi-quantitative method according to Kato-Katzo technique [25].

A second aliquot comprising a subsample of 201 stools drawn from the HIV infected patients with $CD4^+$ cell counts less than 500 cells/μl was kept refrigerated at −20°C and processed within 3 months to detect coproantigens of *Cryptosporidium* spp. using the commercial Kit Ridascreen® *Cryptosporidium* – R-Biopharm AG (Darmstadt, Germany) according manufacturer's instructions (Figure 1).

Each stool sample was examined both by two laboratory technicians as well as by one of the authors (BZC), reaching 94% concordance among the three observations.

### Data Analysis

Epi-info™ version 7.2.1 was used to establish a database and for double-entry data input by two different individuals. After validation of the database, two identical datasets were obtained, of which one was used for all subsequent analyses with Statistical Package for Social Science (SPSS) version 20.0 statistical software. Differences in proportion of HIV positive patients with $CD4^+$ values within specific ranges and stool examination results showing characteristic findings were tested using chi-square test. Multivariate logistic regression modeling was employed to analyze the relationship of socio-demographic, and immunological variables with parasite infection and HIV sero status. We also tested the parasitic infection intensity, according to the HIV sero-status using the Mann-Whitney test. A p-value $< 0.05$ was considered statistically significant.

## Results and Discussion

### Socio- Demographic and general characteristics of the study population

We recruited 517 patients over 2 years old of whom 371 (71.8%) were HIV infected and 146 (28.2%) were HIV uninfected. The mean age was 40.8 (range 5 – 70 years) for the HIV

infected patients and 37.6 (range 3 – 76 years) for the HIV uninfected population. The majority of study population were female 273 (65%) and only 20 (3.9%) were children up to 14 years of age (see Table 1).

In total, 360 (97%) of the HIV infected patients were receiving antiretroviral therapy (ART) for a median duration of 48 (IQR: 1–149) months. Their median overall $CD4^+$ cell count at study enrollment was 437.4 cells/μl [IQR: 10 – 1810], and 72 (19.4%) were taking TS. The majority of the study participants were literate 418 (93%), 503 (97.3%) had piped water, 258 (49.9%) had animals at home and 515 (99.6%) reported washing hands after defecation. In our study, we found no association between risk factors such as education level, sources of drinking water, hand washing after defecation, pets in the peridomicile and parasite infection. These findings are similar to those of other studies conducted both within and outside of Mozambique, which suggests that all patients are equally exposed to the risk of infection regardless of their literacy, water source, hygienic behavior and HIV sero status [16,19]. This is further supported by the fact that in our study *Entamoeba coli*, although not pathogenic, was the second most frequently detected parasite, with 40 (7.7%) prevalence. This is an indication of oral fecal contamination and therefore supports the existence of poor environment and sanitation [19] (see Table 1).

### Prevalence of intestinal parasites in the HIV infected and uninfected patients

The microscopic examination of the stools demonstrated that 148 (28.6%) of all patients were infected with at least one parasitic species, and overall, 10 different species comprising helminthes and protozoa, including coccidia were identified in the stool samples (Table 2 and Figure 2).

This study demonstrated that the prevalence of intestinal parasites is high in both HIV infected patients 98 (26.4%) and HIV uninfected persons 50 (34.2%), although there was not a statistically significant difference. These findings are similar to other studies conducted in Mozambique and other sub-Saharan countries including Nigeria, Ethiopia and Uganda [5,7,10,16,19,22,26]. One possible explanation for having higher prevalence of parasites in uninfected population when compared with HIV infected population may result from the fact that HIV infected patients are more often likely to be in contact with the health care system and thus more exposed to anti-parasitic drugs. In adition, since syndromic treatment of fever or diarrhea with TS is common in those with AIDS and other conditions such as malaria in Mozambique. Given the effect of TS on folic acid metabolism of the worms, we were not surprised to find a lower prevalence of parasitic infection in HIV infected patients when compared to HIV uninfected patients [15,16]. Other studies document similar results and it is thought that the lower prevalence of intestinal parasites in HIV infected patients compared with HIV uninfected is associated with the use of ART. It is thought that ART drugs may have an effect on the parasite clearance, even when the patients respond poorly to ART in terms of the $CD4^+$ T-cell count, possibly due to the mitochondrial toxicity of the ART on the worms [16,17]. In our study, almost all HIV infected patients 360 (97%) were on ART.

Paradoxically, the overall parasitic intensity in HIV infected patients was 2.4 times that of HIV uninfected. Although these differences were not statistically significant, it might be

explained based on the impairment of the immune system in those patients, which is not allowing the clearance of the infection. HIV infection induces cellular depletion and early abnormalities of $CD4^+$ T cells, decreases $CD8^+$ T-cell and function, causes deterioration of specific antigen responses and leads to alteration of innate immunity through impairment of cytolytic activity and cytokine production by natural killer cells, all important pathways to necessary resolve infection [17].

The most frequently detected helminthes in both populations were *Trichuris trichiura* 67 (12.9%), *Ascaris lumbricoides* 27 (5.2%), while the most prevalent protozoa was *Entamoeba coli* 40 (7.7%). *G. intestinalis* prevalence was significantly higher in HIV infected patients 12 (3.2%) compared with HIV uninfected patients 1 (0.7%) for $p = 0.02$. This finding is consistent with other studies, such as in Ethiopia, though it is not considered opportunistic parasite [27].

*Cryptosporidum* spp. and *C. belli* were detected each in one stool sample of HIV infected (0.3%) and HIV uninfected patients (0.2%), respectively (see Table 2). The relatively low prevalence of *Cryptosporidium* spp. detected by microscopy is difficult to interpret when compared with other studies conducted within the country and other countries such as in Ethiopia, Cameron, Nigeria and India [4,6,20–22,26,28]. Among HIV infected patients, 360 (97%) were on ART. These findings could also explain the low parasitic prevalence in this group of patients when compared to HIV uninfected patients, if we consider improvement of the immune system function due to ART or to the drugs activity directly against parasites, as observed in other studies [16,27]. In addition, our HIV infected study population was not severally immunosuppressed as shown by the median $CD4^+$ cell count. This could be possibly another reason to explain the lower prevalence of *Cryptosporidium* spp. or *C. belli* detected in our study. Further controlled randomized studies with HIV infected patients on ART and without ART need to be carried out.

A single parasite was present in 119 (80.4%) of the study population while the remaining 29 (19,6%) were noted to have two parasite species. Unlike ours, other studies have reported mixed parasite infections of up to 5 parasites [4,19]. This could be related to the better living conditions of our study population or to the possibility that our HIV-1 infected patient population was less immunologically advanced. HIV infected patients had fewer mixed infections, 15 (15.3%) than HIV uninfected patients 14 (28%), but this difference was not statistically significant. The most frequent mixed parasites association was between *T. trichiura* + *A. lumbricoides* with 12 (8.1%), followed by the association between *T. trichiura* + *E. coli* with 7 (4.7%). Other combination comprised *T. trichiura* + *E. histolytica* (1.4%), *T. trichiura* + *S. mansoni* (0.7%), *T. trichiura* + *S. stercoralis* (1.4%), *G. intestinalis* + *E. coli* (1.4%). *A. duodenalis +E. coli, A. duodenalis +A. lumbricoides, T. trichiura +G. intestinalis* were present in 0.7% each (see Table 3).

Given that HIV infected patients are the group most exposed to the health system where anti-helminthic drugs and TS are routinely used for almost all patients seeking care, these results are not surprising. In addition, TS is known to have anthelminthic effect and is used for HIV positive patients for prophylaxis of opportunistic infections such as *C. belli*. Although *C. belli* is an opportunistic parasite, it was not identified in stool from any HIV

infected patients, but we did identify it in 1 (0.2%) HIV uninfected patient. Similar results were found in other studies where HIV infected patients did not have or presented with a lower prevalence of *C. belli*. This could be also attributed to the use of TS in those patients as a prophylaxis for opportunistic infections, in which *C. belli* is included [16]

Overall *Cryptosporidium* spp. coproantigen prevalence was 8 (6%) in patients with $CD4^+$ cell counts of less than 500. There was no statistically significant differences between patients with $CD4^+$ Cell count less than 200 and $CD4^+$ cell count between 200 – 500 cells/μL. Prior studies have demonstrated that the prevalence of *Cryptosporidium* spp. may be under-estimated by microscopy and by coproantigens compared to PCR that is much more sensitive [29]. Other studies revealed relatively higher sensitivity of immune enzymatic assays for detection of *Cryptosporidium* spp. when compared with microscopic detection [30]. In our study, the prevalence of *Cryptosporidium* spp. determined by coproantigen detection, an immune enzymatic assay (EIA), was higher (6%) than that detected by microscopy (0.3%). Though there is no known effective treatment to this parasite, given the life-threatening diarrhea and associated vomiting in patients with immunologically advanced HIV disease and in malnourished children, the ability to detect, prevent and treat this pathogen must be prioritized [22,27,29].

## Parasitic infection in HIV infected patients stratified according to $CD4^+$ cell counts and TS intake

In our study $CD4^+$ T-cell counts of < 200 cells/μl were associated with higher parasitic prevalence and parasitic intensity. We found that patients with $CD4^+$ T-cell counts of < 200 cells/μl had a higher prevalence of parasite infection (16,8%) than those with $CD4^+$ cell counts of more than 200 cells/μl. ($p = 0.05$). Similarly the mean parasitic intensity was significantly higher in HIV infected patients with $CD4^+$ cell counts below 200 cells/μl ($p = 0.04$). This difference may also reflect the impairment of both cell-mediated and humoral immunity by HIV with consequent inability of the host to completely clear up the infection [17,29].

HIV infected patients who reported treatment with TS demonstrated 3.5 times less prevalent intestinal parasite co-infection 6 (22.2%) vs 56 (77.8%) than patients not taking TS. Although these differences were not statistically significant, TS might be advised whenever possible if we take into consideration the effect of parasitism on the progression of AIDS and detrimental impact on the nutritional state of the patients [10,12,16,27]. Similarly, parasitic intensity was less in patients taking TS versus patients not on TS.

Several limitations in our study should be highlighted. First, our population was recruited by convenience in a time limited frame from patients attending a single health facility who were willing to informed consent. Thus, the data obtained from HIV uninfected patients cannot be assumed to be representative of the general population not actively receiving health care. Secondly, we used only one stool sample for each patient and it is possible that the parasite prevalence we determined is under-estimated due to the intermittent release of eggs and cysts from helminthes and protozoa and the low sensitivity of microscopic techniques [31]. It is usually recommended to perform serial stool tests with at least 3 samples per patient to increase the sensitivity of the techniques employed [27,32]. Finally, the results for

coproantigen detection of *Cryptosporidium* spp. may be under-estimated due to limitations of the sensitivity of our assay, which is around 92%. Thus, a negative result in this assay does not rule out the possibility of *Cryptosporidium* spp. infection. Such a result may be due to intermittent excretion of the parasite, or the amount of antigen in the sample may be below the level of detection of our assay.

## Conclusion

Given the high prevalence of intestinal parasites detected in HIV infected and HIV uninfected patients and their negative impact on the progression of AIDS and nutritional status of the patients, routine examinations of stool samples for parasite diagnosis and treatment is highly recommended in settings like ours. This might result in significant reductions in morbidity, improvement of the efficacy of antiretroviral therapy as well as the nutritional status of patients and might contribute to the SDGs achievement. In our study a $CD4^+$ T-cell count of < 200 cells/μL was a risk factor for parasitic prevalence and parasitic intensity, while TS intake seemed to have a mitigating effect on prevalence and parasitic intensity.

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## Abbreviations

| | |
|---|---|
| **AIDS** | Acquired Immunodeficiency Syndrome |
| **ART** | Anti-Retroviral Therapy |
| **$CD4^+$** | Cluster of Differentiation 4 |
| **EIA** | Enzymatic Immuno-Assay |
| **HIV** | Human Immunodeficiency Virus |
| **NTDs** | Neglected Tropical Diseases |
| **SSA** | Sub-Saharan Africa |
| **SDGs** | Sustainable Development Goals |
| **STHs** | Soil Transmitted Helminthes |

| | |
|---|---|
| **TS** | Trimethoprim-Sulphamethoxazole |
| **UEM** | University Eduardo Mondlane |
| **WHO** | World Health Organization |
| **ZNM** | Ziehl Neelsen |

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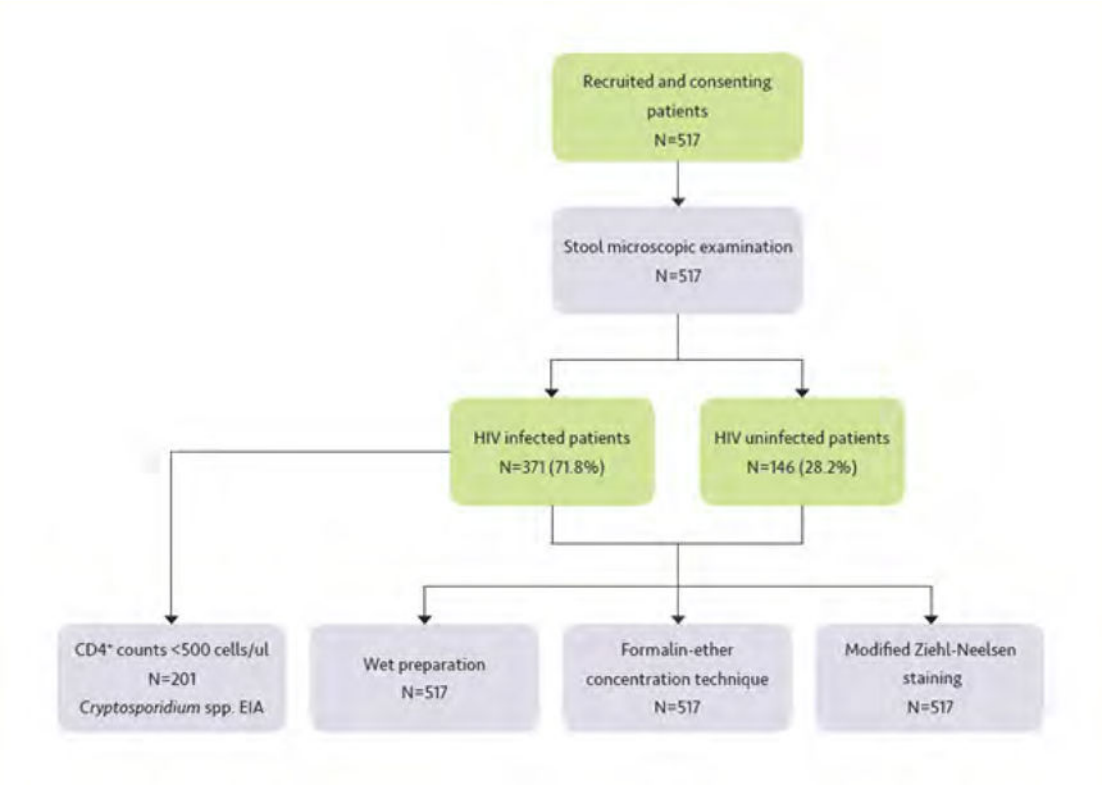


**Figure 1.**
Workflow chart of participants recruitment, enrolment strategy and laboratory diagnostics.

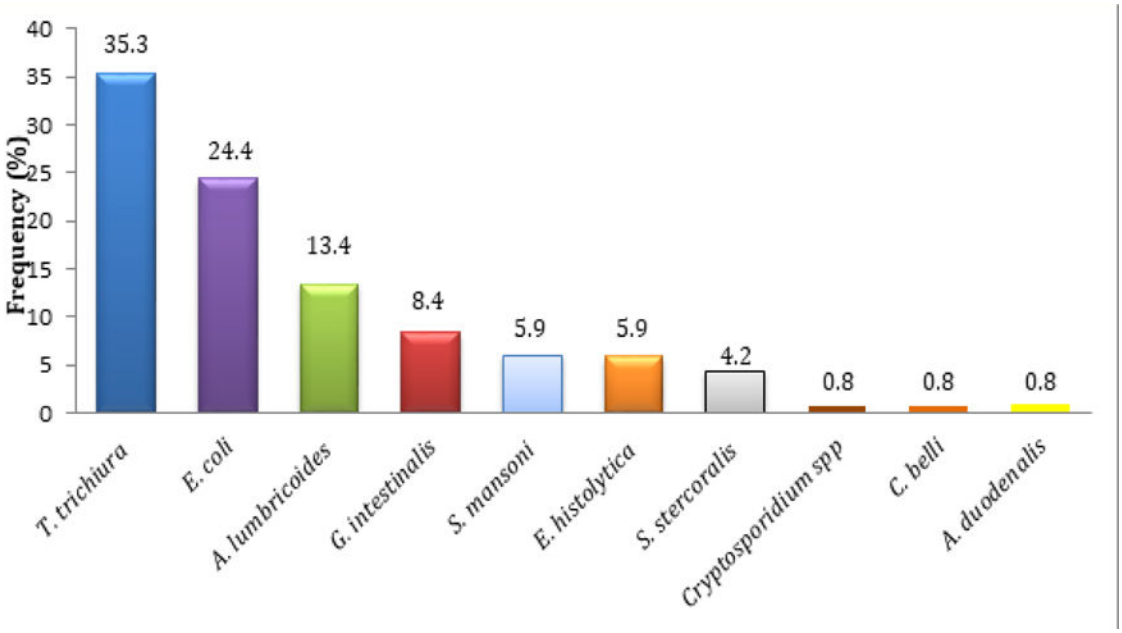


**Figure 2.**
Profile of parasites detected in the study population.

## Table 1

Socio-demographic profile of the study population.

| Demographic data | | HIV infected n = 371 (71.8%) | HIV uninfected n = 146 (28.2%) | Total n = 517 (100%) |
|---|---|---|---|---|
| **Sex** | Male | 134 (34.9%) | 51 (36.1%) | 185 (35.8%) |
| | Female | 237 (65.1%) | 95 (63.9%) | 332 (64.2%) |
| **Age** | Children ≤ 14 years | 5 (1.3%) | 15 (10.3%) | 20 (3.9%) |
| | Adults | 366 (98.7%) | 131 (89.7%) | 497 (96.1%) |
| **Education** | Illiterate | 17 (4.6%) | 19 (13.0%) | 36 (7.0%) |
| | Primary school | 243 (65.5%) | 77 (52.7%) | 320 (61.9%) |
| | Secondary school | 86 (23.2%) | 28 (19.2%) | 114 (22.1%) |
| | Pre-University | 18 (4.9%) | 16 (11.0%) | 34 (6.6%) |
| | University | 7 (1.9%) | 6 (4.1%) | 13 (2.5%) |
| **Water source** | | | | |
| | Tap | 362 (97.6%) | 141 (96.6%) | 503 (97.3%) |
| | Mineral | 5 (1.3%) | 1 (0.7%) | 6 (1.2%) |
| | Well | 4 (1.1%) | 4 (2.7%) | 8 (1.5%) |
| **Wash hands after:** | | | | |
| Defecation | Yes | 369 (99.5%) | 146 (100.0%) | 515 (99.6%) |
| | No | 2 (0.5%) | 0 (0.0%) | 2 (0.4%) |
| Contact with people | Yes | 369 (95.5%) | 143 (97.9%) | 512 (99.0%) |
| | No | 2 (0.5%) | 3 (2.1%) | 5 (1.0%) |
| Contact with animals | Yes | 370 (99.7%) | 146 (100.0%) | 516 (99.8%) |
| | No | 1 (0.3%) | 0 (0.0%) | 1 (0.2%) |
| Animals at home | Yes | 181 (48.8%) | 77 (52.7%) | 258 (49.9%) |
| | No | 190 (51.2%) | 69 (47.3%) | 259 (50.1%) |

## Table 2

Prevalence of intestinal parasites in the study population.

| **Parasites** | **HIV infected N = 98/371 (26.4 %)** | **HIV uninfected N = 50/146 (34.2 %)** | **Total N = 148/517(28.6%)** |
|---|---|---|---|
| Helminthes | | | |
| *T. trichiura* | 41 (11.1%) | 26 (17.8%) | 67 (12.9%) |
| *A. lumbricoides* | 15 (4.0%) | 12 (8.2%) | 27 (5.2%) |
| *S. mansoni* | 5 (1.3%) | 2 (1.4%) | 7 (1.3%) |
| *S. stercoralis* | 5 (1.3%) | 3 (2.1%) | 8 (1.5%) |
| *A. duodenalis* | 1 (0.3%) | 2 (1.4%) | 3 (0.6%) |
| Protozoan | | | |
| *E. coli* | 24 (6.5%) | 16 (11%) | 40 (7.7%) |
| **G. intestinalis* | 12 (3.2%) | 1 (0.7%) | 13 (2.5%) |
| *E. histolytica* | 7 (1.9%) | 2 (1.4%) | 9 (1.7%) |
| Coccidian | | | |
| *Cryptosporidium* spp | 1 (0.3%) | 0 (0.0%) | 1 (0.2%) |
| *C. belli* | 0 (0.0%) | 1 (0.7%) | 1 (0.2%) |

* p = 0.02

**Table 3**

Mixed Parasitism in HIV infected and HIV uninfected patients.

| **Parasitism** | **HIV infected patients N = 98/371 (26.4 %)** | **HIV uninfected patients N = 50/146 (34.2 %)** | **Total N = 148/517(28.6%)** |
|---|---|---|---|
| **One Parasite** | 83 (84,7%) | 36 (72%) | 119 (80,4%) |
| *T. trichiura + A. lumbricoides* | 5 (5.1%) | 7 (14.0%) | 12 (8.1%) |
| *T. trichiura + E. coli* | 3 (3.1%) | 4 (8.0%) | 7 (4.7%) |
| *T. trichiura + E. hystolitica* | 2 (2.0%) | 0 (0.0%) | 2 (1.4%) |
| *T. trichiura + S. mansoni* | 0 (0.0%) | 1 (2.0%) | 1 (0.7%) |
| *T. trichiura + S. stercolaris* | 1 (1.0%) | 1 (2.0%) | 2 (1.4%) |
| *G. Intestinalis + E. coli* | 2 (2.0%) | 0 (0.0%) | 2 (1.4%) |
| *A. lumbricoides + A. duodenalis* | 1 (1.0%) | 0 (0.0%) | 1 (0.7%) |
| *E. coli + A. duodenalis* | 0 (0.0%) | 1 (2.0%) | 1 (0.7%) |
| *T. trichiura + G. Intestinalis* | 1(1.0%) | 0 (0.0%) | 1 (0.7%) |
| Total mixed parasitism | 15 (15.3) | 14 (28%) | 29 (19.6%) |