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Original Research Article

Intestinal parasitic infections in HIV-1 sero-positive individuals and its correlation with CD4 cell count in a tertiary care teaching hospital, Gujarat

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Article History:*Received:** 17/04/2017**Revised:** 26/04/2017**Accepted:** 26/04/2017**DOI:** <https://dx.doi.org/10.7439/ijbar.v8i4.4114>**Abstract**

Background: Opportunistic infections constitute a major health problem in patients infected with HIV. Among these, intestinal parasitic infections are the commonest and are a major cause of morbidity and mortality in HIV positive individuals worldwide. The present study was carried out to identify various intestinal parasites in HIV-1 sero-positive individuals and to correlate these intestinal parasitic infections with CD4 count.

Methods and Material: In a cross-sectional prospective study, stool samples of a total of 27 patients were examined for the presence of the enteric parasites using direct saline & iodine wet mount, formal ether concentration technique, modified acid fast staining & Giemsa staining. Data of recent CD4 cell count was obtained from the patients.

Results: Out of 58 patients given written consent for participating in the study, only 27 (47%) patients provided stool samples. The prevalence of enteric parasites in HIV-1 sero-positive patients was 7.4%. The parasites detected in our study population were *Entamoeba coli* (3.7%) and *Ancylostoma duodenale* (3.7%). *Entamoeba coli* (3.7%) was detected in the patient with CD4 count less than 200 cells/ μ L. 96.3% patients were on ART. Intestinal parasites were detected from ART (3.8%) and naïve ART (3.8%) patient.

Conclusion: Routine examination of stool samples for parasites could significantly benefit the HIV-infected individuals by contributing to reduce morbidity and improve quality of life. However; considering to limited sample size and a single stool specimen received in the present study, a large study is required.

Key Messages: Early detection and treatment of intestinal parasitic infections are important to improve the quality of life of HIV sero-positive patients.

Keywords: HIV, intestinal parasitic infection, CD4 count

1. Introduction

Human immunodeficiency virus (HIV)/ Acquired Immunodeficiency Syndrome (AIDS) ranks among the most dreaded diseases afflicting mankind, causing dysfunction of both limbs of the immune system, resulting in overwhelming and fatal opportunistic infections. [1] The magnitude of intestinal parasitic infection in Acquired Immunodeficiency Syndrome patients requires careful consideration in the developing world where poor nutrition

is associated with poor hygiene and several tropical diseases. [2] These parasites have an undeniable influence on the patient's general condition and may lead to fatal complications in the immune-suppressed individuals. [2, 3] India is now facing the HIV epidemic, a good number of HIV sero-positive patients presents with diarrhea. [3] Such opportunistic parasitic gastrointestinal infections causing severe diarrhea profoundly compromise the absorptive

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function of the small intestine, and result in significant mortality. [4] Opportunistic infections caused by intestinal parasites also vary according to the geographical area and the endemic levels in each location. [5] Such co-infections present with more severe clinical symptoms compared to parasite infections of otherwise healthy people, and are more difficult to treat. The high prevalence of certain opportunistic parasites among HIV positives is well known. [4] Opportunistic parasitic infections are a common feature in HIV/AIDS infections where almost 80% of AIDS patients die of AIDS-related infections including intestinal parasites rather than of the HIV infection itself which usually occur late in the course of HIV infection when Cluster of Differentiation (CD4) + T-cell count has been severely depleted mostly below 200 cells/ μ L.[6] Diarrhea causing opportunistic parasites like *Cryptosporidium parvum* and *Isospora belli* is common among HIV positive persons with CD4 count less than 200 cells/ μ L. [7] As many of these infections are amenable to treatment, an early and accurate diagnosis is important. Screening of the intestinal parasite from the stool sample was still not carried out as a routine test in HIV sero-positive patients in most of the laboratories and information available among clinician about the prevalence of opportunistic infections is scarce. [3] There were few studies showing correlations of CD4 levels with rate of opportunistic intestinal parasites in India. Therefore, the present study was planned to identify various intestinal parasites in HIV-1 sero-positive individuals and to correlate these intestinal parasitic infections with CD4 count.

2. Materials and methods

This cross-sectional prospective study was carried out at Shree Krishna Hospital, Karamsad during May 2016 to February 2017. The study was conducted after the approval of Institutional Ethics Committee. HIV-1 sero-positive individuals of any age and sex, symptomatic or asymptomatic for GIT symptoms who visited Integrated Counselling & Testing Centre (ICTC) & Link-ART (Antiretroviral therapy) centres of Shree Krishna Hospital Karamsad, Civil Hospital Petlad and People Living with HIV/AIDS (PLHA) who met regularly at Emery Hospital, Anand were included in the study. All the patients were counselled and explained the objectives of the study & were encouraged to participate in the study. Written informed consent was obtained from the patients who volunteered for the study. In case of Paediatric patients, written consent was obtained from the parents or guardians. After the written consent of patient, details such as name, age, sex, occupation, marital status, history of ART, duration of ART, symptoms, date of collection of stool sample and other relevant information were recorded in the prescribed

proforma. Details of recent CD4 cell count were also recorded.

A single stool sample was collected from each patient in a clean screw capped, disposable plastic container and immediately transported to Microbiology laboratory at the Central Diagnostic Laboratory, Shree Krishna Hospital and Pramukhswami Medical College Karamsad, Anand, Gujarat, an NABL accredited laboratory. Stool specimen was examined immediately for presence of parasites as per standard operating procedures of the laboratory. In case of delay, samples were preserved by adding 10% formalin and examined maximum within 72 hrs. of collection. All stool samples were examined for the presence of cyst, trophozoites, eggs and larvae of parasites using direct (saline and iodine wet mount) microscopy and after formal ether concentration technique as per the standard protocol. Modified acid fast staining was done for detecting oocysts of *Cryptosporidium*, *Isospora*, and *Cyclospora*. Giemsa staining was done especially for detecting Microsporidial spores. [21]

3. Results

A number of patients were counselled and a total of 58 consent forms were obtained from the HIV-1 sero-positive patients who volunteered for the study. All the patients were asked to collect stool sample. A total of 27 (47%) patients provided stool samples for the study and 31 (53%) patients did not comply for various reasons. Thus a total of 27 stool samples from HIV-1 seropositive patients were examined for the enteric parasitic infections. In the present study, majority of the patients (44.4%) were in the 31-40 years age group. Male constitutes 62.9 % and females 37% of the total patients (Table 1).

Table 1: Demographic profile and prevalence of intestinal parasites in HIV-1 sero-positive patients (N=27)

Characteristics	Parasites Positive (%)	Parasites Negative (%)
Age (years)		
<10	00	01 (3.7)
11 – 20	01 (3.7)	00
21 – 30	00	01 (3.7)
31 – 40	00	13 (44.4)
41 – 50	01 (3.7)	09 (33.4)
> 50	00	01 (3.7)
Total	02 (7.4)	25 (92.6)
Gender		
Male	00	17 (62.9)
Female	02 (7.4)	08 (29.6)
Total	02 (7.4)	25 (92.6)
Diarrhoeal status		
Positive	02 (7.4)	02 (7.4)
Negative	00	23 (85.1)
Total	02 (7.4)	25 (92.6)

Table 2: Prevalence of parasitic infections in HIV-1 sero-positive patients (N=27)

Parasite species	No. positive/ No. examined	Positive rate (%)
<i>Entamoeba coli</i>	1 / 27	3.7
<i>Ancylostoma duodenale</i>	1 / 27	3.7
Total	2 / 27	7.4

Out of 27 HIV-1 sero-positive patients, only 2 (7.4%) cases were positive for intestinal parasites. In one case, cyst of *Entamoeba coli* was observed and in other case eggs of *Ancylostoma duodenale* was seen (Table 2).

Table 3: Parasite detection of HIV positive patients in relation to CD4 count (N=27)

CD4 count (cells/ μ l)	Parasite positive	Parasite negative	Total (%)
<200	01	01	02 (7.4)
200-349	00	04	04 (14.8)
350-499	00	09	09 (33.3)
\geq 500	00	11	11 (40.7)
Not done	01	00	01 (3.7)
Total	02	25	27

Out of two patients with parasitic infections, 1(3.7%) patient had CD4 count <200 cells/ μ l. However; CD4 count was not done in another patient at the time of enrolment in the study (Table 3).

Table 4: Associating intestinal parasite with ART status among HIV positive patients (N=27)

ART status	Parasite positive	Parasite negative	Total (%)
On ART	01	25	26 (96.3)
Naïve ART	01	00	01 (3.7)
Total	02	25	27

A total of 26 patients were on ART and 1 patient was found to be positive for intestinal parasitic infection. However; 1 naïve ART patient was also positive for intestinal parasitic infection (Table 4).

4. Discussion

HIV infection is a major threat to public health in India. [3] Since the first AIDS case was described, a high prevalence of gastrointestinal disorder has been reported, especially diarrhoea due to parasite infection. [5] Due to deterioration of the immune system because of HIV infection, numerous opportunistic infections occur, of which gastrointestinal parasitic infection is a universally recognized problem. This largely presents with diarrhoeal symptoms leading to life threatening complications. [1] The detection of such common intestinal parasites in both patients and controls could be a reflection of the poor environmental sanitation and personal hygienic practices,

which emphasize the need for interventional measures at the community level to reduce the risk factors of acquiring intestinal parasites in immunocompromised patients.[5] The high prevalence of parasitic diseases in HIV infected patients also draws attention to the need to include routine stool examination in HIV/AIDS management. [2]

In the present study, only 27 patients out of total 58 submitted the stool specimen. However 31 patients did not submit the specimen even after explaining the significance of stool examination and benefit to the patient. The main barriers for not submitting the specimen were mainly hesitation, embarrassment during transportation, why to give specimen if they are asymptomatic and fear of getting the bad result. Similar findings have been observed in other studies which include the embarrassment, lack of understanding, fear of the possibility of receiving bad result and personal attitude. [22]

Most of the studies performed in India and other countries reported a higher prevalence of enteric parasites ranging from 30% to 60%. [3] The prevalence rate of intestinal parasites in HIV-1 sero-positive patients in our study was 7.4%. This is in accordance with the studies done in other parts of India (New Delhi, Chennai, Maharashtra) with prevalence ranging from 3.7-11.8%. [3]

In the present study, only two parasites were detected in the stool sample of two patients suffering from diarrhoea out of 27-HIV 1 seropositive patients. One was the nematode *Ancylostoma duodenale* (3.7%) considered to be potentially pathogenic and other was protozoa *Entamoeba coli* (3.7%) considered to be non-pathogenic. In the study by Gupta *et al* [3] *Entamoeba coli* was found to be the most common non-pathogenic parasite. In contrast to our study, however predominance of *E. histolytica*/*E.dispar*, *Cryptosporidium* species, *S. stercoralis*, *A. lumbricoides* and *G. lamblia* were detected among HIV positive individuals. [19]

In other studies, [15] the most common parasite detected was *I. belli* in 11.5% of the total population followed by *E. histolytica*/*E. dispar* (4.7%) and *Cryptosporidium* sp. (3.6%). Within helminths, *S. stercoralis* and *A. lumbricoides* were the most common (3.1%).

Our study showed the presence of *Entamoeba coli* in the patient with CD4 count less than 200 cells/ μ L. This is in agreement with studies [23] that parasites associated with HIV are more likely encountered as the CD4 T-cell count falls below 200 cells/ μ L. This suggests that although it is considered to be non-pathogenic in immunocompetent person but may act as opportunistic pathogen in HIV seropositive patients. A study by Gupta *et al* [11] reported that the presence of the coccidian parasites was associated with a lower CD4+ T-cell count in the HIV positive

patients. Multiple infections with *Isoospora belli* and *Cryptosporidium* spp. were seen in two patients, *Isoospora belli*, *Cryptosporidium* and *Strongyloides stercoralis* in one case and *Isoospora belli* and *Giardia lamblia* in another. In the present study, CD4 count was not done in other patient with recent diagnosis of HIV infection but eggs of *Ancylostoma duodenale* were detected in the stool.

In the present study among the total patients, 96.3% were on ART. Out of two patients which were positive for intestinal parasites, one patient was on ART since 11 years and other was fresh new case recently detected so not on ART. This however suggests that prevalence of opportunistic enteric parasites was considerably low in patients on ART as compared to those not taking ART. A study by Ahmed *et al* [15] reported about 54.2% of the HIV sero-positive patients in the study population were on HAART. Out of total 57 parasites detected in HIV sero-positive patients, only 24.6% were detected in patients who were on HAART, and 75.4% were detected in patients who were not on HAART.

The low prevalence of intestinal parasites in our study might be due to that most of the patients were on the ART. The other reason might be due to difference in immunity, diarrheic status, environmental and personal hygiene of the patients.

There were some limitations in this study, which include a limited sample size, availability of single stool specimen; resource constrains to perform sensitive tests like polymerase chain reaction (PCR) and immunofluorescence for confirmation of parasites like *Cryptosporidium* and *Microsporidia*. In addition, the patients were unwilling to participate in the study.

With better knowledge and diagnosis of opportunistic parasitic infections in HIV-seropositive individuals and specific antimicrobial prophylaxis by itself or in conjunction with antiretroviral therapy can reduce substantial morbidity and mortality caused by opportunistic infections in HIV-infected individuals. The CD4 counts must also be evaluated in HIV-confirmed cases to follow up the cases.

Although the limited sample size, our findings highlight the importance of screening HIV-seropositive individuals. It also suggests that the intestinal parasitic infections vary with the geographical areas and further large prospective studies should be carried out in respective regions to evaluate the frequent causes of intestinal parasitic infections in HIV-seropositive individuals.

5. Conclusion

In the cross sectional study carried out at Shree Krishna Hospital, Karamsad, the overall prevalence of intestinal parasitic infection was 7.4%. The present study

highlights the importance of testing for intestinal parasites in patients who are HIV sero-positive. Routine examination of stool samples for parasitic infections could significantly benefit the HIV-infected individuals for treatment and preventive measures. However; considering to limited sample size and a single stool specimen received in the present study, a large study is required.

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