Opportunistic Intestinal Protozoan Infections in HIV/AIDS Patients on Antiretroviral Therapy in the North West Region of Cameroon

Ntonifor Helen Ngum¹, Asanji Nelly Ngum² and Shei Stanley Jini³

¹Department of Biological Sciences, University of Bamenda, North West Region, Cameroon.  
²Department of Biomedical Sciences, Troy University, Troy, Alabama, United States.  
³Department of Zoology and Animal Physiology, University of Buea, South West Region, Cameroon.

Authors’ contributions

This work was carried out in collaboration between all authors. Authors NHN, ANN and SSJ designed the study, performed the statistical analysis, wrote the protocol, and wrote the first draft of the manuscript and managed literature searches. Authors NHN and ANN managed the analyses of the study and literature searches. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/BMRJ/2015/15737

ABSTRACT

Background: In most developing countries, intestinal parasites are the major cause of morbidity and mortality killing millions of patients each year. Hence, this study was undertaken to determine the prevalence of intestinal protozoans in HIV/AIDS patients on ART in three hospitals in the North West region of Cameroon.

Methodology: A cross sectional study was carried out in three Hospitals in the North West Region of Cameroon, focusing on HIV/AIDS patients who were enrolled for ART in these Hospitals. A total of 320 HIV/AIDS patients aged 1-70 years, of both sexes (230 females and 90 males) participated in the study. Stool samples were collected and processed using direct wet mount, formol-ether concentration technique and modified Ziehl-Neelson staining techniques.
1. INTRODUCTION

AIDS has killed more than 25 million people since 1981, hence making it one of the most destructive pandemics in history [1]. According to UNAID/WHO, an estimated 35.3 million people were living with HIV globally at the end of 2012 with 69% of them in Sub-Saharan Africa [2].

One of the major health problems among HIV sero-positive patients are superimposed infections due to the deficient immunity. Opportunistic infections pose major health problems among these patients particularly in the late stage of the disease when immune suppression is severe. Furthermore, intestinal parasitic (IP) infections, which are also one of the basic health problems in tropical regions, are common in these patients. It is estimated that about 60% of the world’s population is infected with intestinal parasites which may play a significant role in morbidity due to intestinal infections [3]. Intestinal parasitic infections have been a major source of morbidity in tropical countries especially among HIV patients [4].

The magnitude of these intestinal parasitic infections in HIV/AIDS patients requires careful consideration in the developing world where poor nutrition is associated with poor hygiene and several tropical diseases. Diarrhoea due to intestinal parasites and microbial infections is a frequent manifestation among HIV infected patients. During the evolution of HIV infection, the gastrointestinal involvement is frequent and 90% of the patients consult for gastrointestinal disorders [5]. Reports indicate that diarrhoea occurs in 30-60% of AIDS patients in developed countries, whereas it reaches up to 90% in developing countries [5]. It has been speculated that HIV infected patients may have unique types of intestinal infections, and that activation from such parasites may affect the progression of HIV disease [6].

Cameroon, with a population exceeding 16 million, has one of the highest infection rates of HIV in the world; approximately 1 million adults (5.5%) are HIV positive [7]. Patients in resource limited settings typically start ART programmes with advanced symptomatic disease and very low blood CD4 cell counts which predisposes them to high rates of both clinical and subclinical opportunistic infections [8]. Therefore, this study was aimed to assess the prevalence of intestinal parasites and associated risk factors in among HIV/AIDS patients in 3 hospitals in the North West Region of Cameroon where little or no data on these opportunistic infections exists.

2. MATERIALS AND METHODS

2.1 Study Population, Design and Sample Size

The study was carried out between March 2012 and September 2012 in the North-West Region, located in the western highlands of Cameroon. The study population was divided into urban (Bamenda town) and rural (Ndop) groups. Samples for the urban area were collected from Mezam Polyclinic and St. Louis Clinic Nkwen and the District Hospital Ndop was the collection centre for the rural area. Participants consulting at the hospitals were given informed consent forms by the study team to fill prior to sample collection. This was a cross sectional comparative study where random sampling technique was used to select 320 consenting HIV positive subjects who were on ART.

2.2 Ethical Considerations

This study was carried out with the approval of the Ethical Review Committee on Health Research, regional delegation of health for the North West Region, Cameroon. Equally, Ethical clearance was obtained from the University of...
Buea ethical clearance committee. Informed written consent was obtained from each study participant. Each participant was free to withdraw consent at any time. All personal and medical information of the participants was treated strictly confidential.

2.3 Sample Size and Sampling Techniques

Stool samples were collected from 320 HIV positive individuals on antiretroviral therapy (ART), in the ART unit of the hospitals during the study period using sterile dry, clean, air tight, wide mouth screw cap labelled containers and analyzed within 24 hours of collection, following standard procedures. Each stool specimen was initially assessed for consistency. The samples were analyzed by direct wet mount [9], formal-ether concentration method and modified ZiehlNeelsen staining technique and weber’s modified trichrome stain [10,11] to detect cysts, oocysts, ova and trophozoites of parasites. Blood samples of all participants were screened for anti-HIV antibodies using Determine HIV 1/2 HIV rapid test and the reactive samples were subjected to confirmation using Genie III 1/2 HIV rapid test. This rapid test was done on the patients to confirm their HIV/AIDS status even though they were already on ART.

2.4 Statistical Analysis

Data generated was entered into Microsoft excel 2010 cleaned and imported into Statistical Package for Social Science (SPSS) software version 11 for analysis. The prevalence of intestinal parasites was determined in relation to different variables. Pearson’s chi square test was used to assess statistical significance difference between proportions. Multivariate logistic model was used to evaluate the risk of parasitic infection according to HIV status, socio demographic characteristics and hygiene condition. A given statistical test was reported significant when its P value was less than or equal to 0.05.

3. RESULTS

3.1 Characteristics of the Study Population

In this study, a total of 320 HIV/AIDS patients were examined, comprising of 230 females (71.9%) and 90 males (28.1%). Equally, 205 (64.1%) lived in the rural area and 115 (35.9%) lived in the urban area. The ages were grouped as shown in Table 1.

Six different species or groups of intestinal protozoan were recovered from the study including; Cryptosporidium parvum, Entamoeba coli, Entamoeba histolytica, Cystoisospora belli, Microsporidia spp and Iodamoeba butschlii. The prevalence of these different species of intestinal protozoa is shown on Fig. 1.

![Fig. 1. Prevalence of intestinal protozoan in the study population](image-url)
Table 1. Prevalence of the different species of protozoans in relation to sex, age and locality

| Group of patient | Examined patient | Infected patient (%) | Species infected (prevalence %) |
|------------------|------------------|----------------------|--------------------------------|
|                  |                  |                      | E. coli | E. h | I. b | C. parvum | C. belli | Mic |
| **Sex**          |                  |                      |         |     |     |           |         |     |
| Females          | 230              | 83 (36.09)           | 32 (13.9) | 26 (11.3) | 4 (1.7) | 31 (13.5) | 5 (2.2) | 19 (8.3) |
| Males            | 90               | 37 (41.1)            | 11 (12.2) | 10 (11.1) | 3 (3.3) | 15 (16.7) | 1 (1.1) | 6 (6.7)  |
| **Total**        | 320              | 120 (37.5)           | 43 (13.4) | 36 (11.3) | 7 (2.2) | 46 (14.4) | 6 (1.9) | 25 (7.8) |
| **Age (yrs)**    |                  |                      |         |     |     |           |         |     |
| 1-10             | 4                | 1 (25)               | 1 (66.7) | 0   | 0   | 0         | 0       | 0       |
| 11-20            | 12               | 3 (25)               | 1 (8.3)  | 1 (8.3) | 0   | 2 (16.7) | 0       | 1 (8.3) |
| 21-30            | 89               | 35 (39.3)            | 11 (12.4) | 13 (14.6) | 0   | 16 (18)  | 3 (3.4) | 3 (3.4) |
| 31-40            | 106              | 43 (40.6)            | 11 (10.4) | 7 (6.6) | 1 (0.9) | 12 (11.3) | 3 (2.8) | 12 (11.3) |
| 41-50            | 70               | 27 (38.6)            | 12 (17.1) | 10 (14.3) | 3 (4.3) | 10 (14.3) | 0       | 6 (8.6) |
| 51-60            | 33               | 10 (30.3)            | 5 (14.7)  | 4 (11.8) | 3 (8.8) | 4 (11.8) | 0       | 2 (5.9) |
| 61-70            | 6                | 1 (16.7)             | 1 (16.7)  | 1 (16.7) | 0   | 2 (33.3) | 0       | 1 (16.7) |
| **Total**        | 320              | 120                  | 43 (13.4) | 38 (11.3) | 7 (2.2) | 46 (14.4) | 6 (1.9) | 25 (7.8) |
| **Location**     |                  |                      |         |     |     |           |         |     |
| Rural            | 205              | 87 (42.4)            | 25 (12.2) | 19 (9.3) | 5 (2.4) | 38 (18.5) | 5 (2.4) | 20 (9.8) |
| Urban            | 115              | 33 (28.7)            | 18 (15.7) | 17 (14.8) | 2 (1.7) | 8 (7)    | 1 (0.9) | 5 (4.2) |
| **Total**        | 320              | 120                  | 43 (13.4) | 36 (11.3) | 7 (2.2) | 46 (14.4) | 6 (1.9) | 25 (7.8) |

*E. h, E. histolytica; I. b, I. butschlii; Mic., Microsporidia spp*
Based on parasitological examination of the stool specimens, out of the 320 HIV patients examined, 120 (37.5%) of them were infected with intestinal protozoan. Out of this lot, 83 (36.09%) were females and 37 (41.1%) were males. Also 87 (42.4%) of the infected patients lived in the rural area while 33 (28.7%) lived in the urban area. The most prevalent parasitic species was *C. parvum* as shown in Table 1.

The sex distribution results (Table 1) showed that prevalence was higher in males (41.1%) than in the females (36.09) and the result was statistically significant at \( P \leq 0.05 \). Equally, the age distribution results (Table 1) showed that the most infected age group was the 31 - 40 (40.6%) years age group, closely followed by the 21–30 (39.3%) years age group. The entire study however showed that most of the parasites were identified in the young and middle age groups of patients, i.e., between 21 years and 50 years, Table 1. Equally most of the patients who harboured intestinal protozoan were found in the rural areas (42.4%) as compared to the urban areas (28.7%) (Table 1).

The prevalence of diarrhoea was 21.9% (71). However, the patients with parasitic infections had a prevalence of 13.13% (42), while 24.4% of the sampled population with parasitic infections were asymptomatic (Tables 2 and 3). Patients with intestinal parasites more often had diarrhoea, in which *C. parvum* (32) was the most common parasite, followed by *E. histolytica* (28) (Table 3).

### 4. DISCUSSION

Intestinal parasitic infections are a major cause of morbidity and mortality in HIV infected patients in Cameroon. In the present study, about 37.5% of the patients were infected with intestinal parasites. These results are almost similar to those reported in Ethiopia [12], but lower than those from other parts of Cameroon and Africa [13-15]. The difference in prevalence may be due to personal hygienic and sanitary habits of the patients. Also all the patients were partially or totally on ART which signified that they may have been conscious of their low immune conditions.

Males had a higher prevalence as compared to females. This is in line with the study conducted in Kano [15]. This difference in infection rate may be due to the fact that most of the men in the rural areas spent the better part of the day in palm wine joints drinking and the palm wine in the study area is always adulterated right from the palm wine bushes with dirty water that might be contaminated. Equally most of the men in the rural area are polygamous. Also it might be due to the fact that both sexes are involved in farming activities where they are likely to ingest cysts of protozoan from the soil, and unwashed fruits and vegetables while working. The prevalence of infection was higher in the rural area than in the urban area. This is consistent with the research carried out in Ethiopia [16]. This increase prevalence in rural areas could be related to poverty, overcrowding, low levels of hygiene, lack of pipe borne water, promiscuous defecation and un-plastered floors in most of the houses. Most of the patients in the rural areas are farmers who are likely to come in contact with dust containing the protozoan cysts. High rate of parasitic infection among farmers in rural areas is due to increased occupational exposure to contaminated soil and water. The prevalence of infection was highest in the age group 31-40 years. These results are in line with the findings obtained in Senegal who reported that prevalence was highest in age group 31-50 years [17]. Also this is probably because the prevalence of HIV/AIDS is high in this age group [18-19]. Equally, prevalence might be high in this age group because it is the most sexually active age group. The least prevalence was in age groups 61–70 years, 1–10 and 11-20 years. In the study, we had few children involved which could be a reason for the low infection rate with protozoan. With the age group of 61–70 years it could be build up of resistance due to previous infections.

### Table 2. Prevalence of diarrhoea among HIV/AIDS patients on ART

| Number of HIV positive patients | Number of patients | Number with Parasitic infections | P Value |
|-------------------------------|-------------------|----------------------------------|---------|
| Symptomatic patients with diarrhoea | 71(21.9) | 42(13.13) |        |
| Asymptomatic patients | 249(77.8) | 78(24.4) | ≤ 0.05 |
| Total | 320 | 120(37.5) |        |
CONCLUSION

The overall prevalence of intestinal parasitic infections in HIV-infected patients in this study was high despite the availability of ART. Public health measures should continue to emphasize the importance of environmental and personal hygiene as well as provide and monitor the quality of drinking water aimed at obtaining a better quality of life for patients. Stool examination should be routinely performed in the follow-up of patients with HIV/AIDS attending ART clinics in order to optimize treatment of infected patients and other preventive measures.

ACKNOWLEDGEMENTS

The authors appreciate the support and cooperation of the staff of Mezam Polyclinic, St. Louis Clinic Nkwen and the District Hospital Ndop. Laboratory space and equipment was provided by these three hospitals, for which the authors are most grateful.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Awuba J, Macassa G. HIV/AIDS in Cameroon: Rising gender issue in policymaking matters. Afr J Hlth Scs. 2007;14(3-4):118-128.
2. UNAID/WHO. UNAIDS report on the global AIDS epidemic. Geneva: Switzerland. 2013;147.
3. WHO. Prevention and control of intestinal parasitic infections. WHO Technical report. 1997;749:1-86.
4. Okoduwa M, Adeyeba OA, Tatfeng YM, Okpala H. Age and sex distribution of intestinal parasitic infections among HIV infected subjects in Abeokuta, Nigeria. Online J Hlth Appl Sci. 2003;2(4):1-5.
5. Bhajee F, Subramony C, Tang S-J, and pepper DJ. Human Immuno deficiency Virus associated gastrointestinal disease: Common Endoscopic Biopsy Diagnosis. Patholog Res Int. 2011;247923:7.
6. Feitosa G, Bandeira AC, Sampaio DP, Badaró R, Brites C. High prevalence of giardiasis and strongly loidiakas among HIV-infected patients in Bahia, Brazil. Brazilian J Infect Dis. 2001;5(6):339–344.
7. Ministry of Public Health and Cameroon National AIDS Control Committee. Preliminary results of the Demographic and Health Survey (DHS-III). Ministry of Public Health, Yaoundé, Cameroon. 2004; SR107(1).

Table 3. Relationship between diarrhoea and species of intestinal protozoa

| Protozoan species | Number infected | Number with diarrhoeic stool |
|-------------------|-----------------|-----------------------------|
| *E. coli*         | 43              | 4                           |
| *E. histolytica*  | 38              | 28                          |
| I. butschlii      | 7               | 1                           |
| *C. parvum*      | 46              | 32                          |
| I. belli          | 6               | 2                           |
| Microspora        | 25              | 0                           |

Prevalence was relatively high with *C. parvum*. Previous studies conducted revealed *Cryptosporidium parvum* and *Giardia lamblia* as the commonest parasites among HIV infected persons and these were mostly associated with diarrhea [20,21]. These results however are not consistent with the findings of other researchers who observed that the most prevalent protozoan was *G. lamblia* [16]. The prevalence of *C. parvum* was relatively high in the rural area than the urban area and the difference was statistically significant at P ≤ 0.05. This is most probably due to the fact that the transmission of *Cryptosporidium* is through the ingestion of contaminated soil or water both of which were very feasible among the rural population. Also it might be due to the fact that many rural dwellers are more in contact with their domestic animals especially goats, sheep, and dogs, thus increasing their chances of being infected with the parasite. This parasite is one of the major opportunistic infections in HIV/AIDS patients [22]. *Cystoisospora* had the least infection rate. This is in agreement with the findings of Sarfati et al. [23].

Diarrhea is a common symptom in HIV infection and a major sign to AIDS progression with the possibility of various opportunistic infections. The high prevalence of intestinal protozoan in HIV infected patients draws attention to the need to include routine stool examination during visits of HIV/AIDS for early treatment of *C. parvum* and other intestinal parasites. Also health education should be encouraged especially in the rural areas in order to make the inhabitants aware (especially the men) to attend health clinics, submit themselves to examinations and also for them to learn how to prevent infections.

5. CONCLUSION

In conclusion, the overall prevalence of intestinal parasitic infections in HIV-infected patients in this study was high despite the availability of ART.
8. Lawn SD, Myer L, Orrell C, Bekker LG, Wood R. Early mortality among adults accessing a community-based antiretroviral service in South Africa: implications for programme design. AIDS 2005;19(18):2141-2148.

9. Cheesbrough M. District laboratory Practice in Tropical Countries. 2nd ed, Dist Lab Pract. United Kingdom. Trop Ctries Part 2 Cambridge University Press. 2006;1:605.

10. WHO. World Health Organization. Guidelines on standard Operating Procedures for Laboratory Diagnosis of HIV-Opportunistic Infections; 2001. Geneva.

11. Weber R, Bryan RT, Owen RL, Wilcox CM, Gorolkin l, Visvesvara GS. Increased light microscopical detection of microsporidia spores in stool and duodenal aspirates. The enteric opportunistic infections working group. N Engl J Med. 1992;326:161-166.

12. Teklemariam Z, Abate D, Mitiku H, Dessie Y. Prevalence of intestinal parasitic infection among HIV positive persons who are naive and on antiretroviral treatment in Hiwot Fana Specialized University Hospital, Eastern Ethiopia; 2013;6. 2013 ID 324329.

13. Mathur MK, Verma AK, Makwana EG, Sinha M. Study of opportunistic intestinal parasitic infections in human immunodeficiency virus/acquired immunodeficiency syndrome patients. J Glob Infect Dis. 2013;5(4):164-167.

14. Nkenfou CL, Nana CL, Payne VK. Intestinal parasitic infections in HIV infected and non-infected patients in a low HIV prevalence region, West-Cameroon. PLoS One. 2013;8:1-16.

15. Jegede EF, Oyeyi ETI, Bichi AYH, Mbah, HA, Torpey K. Prevalence of intestinal parasites among HIV/AIDS patients attending Infectious Disease Hospital Kano, Nigeria. Pan Afr Med J. 2014;17:295.

16. Haileyesus A, Beyene P. Intestinal protozoan infections among HIV positive persons with and without antiretroviral treatment (ART) in selected ART centres in Adama, Afar and Dire-Dawa, Ethiopia. Ethiopian J Hlth Dev. 2009;23(4):133-141.

17. Faye B, Tine RC, Ndiaye JL, Kintega C, Manga NM, Sow PS, Gaye S. Impact of intestinal parasites on intensity of HIV infection in Senegal. J Antiviral Antiretroviral. 2010;2(1):11-12.

18. Gbgre KK. Prevalence of intestinal parasites in HIV/AIDS infected adult patients in South Western Ethiopia. Ethiop J Hlth Develop. 2003;17(1):71-78.

19. Feleke M, Yenew K, Afework K, Getu D, Moges T, Molla G. Infection with HIV and intestinal parasites among street dwellers in Gondar city Northwest Ethiopia. Japan J Infect Dis. 2006;59(6):400-403.

20. Kurniawan, A, Karyadi T, Dwintasari S W, Sari I P, Yunihastuti E, Djauzi S, Smith HV. Intestinal parasitic infections in HIV/AIDS patients presenting with diarrhoea in Takhta, Indonesia. Trans R Soc Trop Med Hgy. 2009;103(9):893-898.

21. Botero JH, Castano A, Mantaya NM, Ocampo NE, Hurtada MI, Lopera MM. A preliminary study of the prevalence of intestinal parasites in immune compromised patients with and without gastrointestinal manifestations. Rev. Inst Med Sao Paulo. 2003;45(4):197-200.

22. Cimerman S, Cimerman B, Lewi DS. Enteric parasites and AIDS. Sao Paulo Med J. 1999;117(6):1516-1580.

23. Sarfati C, Bourgeois AJ, Menotti F, Liegeois R, Moyou-Somo E, Delaporte, Derouin E, Ngole M, Molina JM. Prevalence of intestinal parasites including microsporidia in human immunodeficiency virus-infected adults in Cameroon: a cross-sectional study. Am J Trop Med Hyg. 2006; 74(1):162-164.

© 2015 Ntonifor et al.; This is an Open Access article distributed under the terms of the Creative Commons Attribution License (http://creativecommons.org/licenses/by/4.0), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Peer-review history:
The peer review history for this paper can be accessed here:
http://www.sciencedomain.org/review-history.php?id=993&id=8&aid=8731