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Prevalence and Predisposing Factors to Intestinal Parasitic Infections in HIV/AIDS Patients in Fako Division of Cameroon

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Abstract **Background**: Understanding the epidemiology of intestinal parasitic infection is essential for the effective management of HIV infection in areas where intestinal parasites are also endemic. Data on the prevalence of intestinal parasitic infection in people living with HIV/AIDS (PLHWA) in Cameroon are sparse. This study was designed to determine the prevalence of intestinal parasitic infections, as well as assess the predisposing factors for the infection in PLHWA in Fako Division of Cameroon. **Methods:** This was a cross-sectional study conducted between April and July 2014. Stool specimen were collected from consented participants and examined for ova, cysts, larvae or oocytes using the Kato-Katz, Formalin-Ether Concentration, Modified Ziehl-Neelsen and Modified field staining techniques. Statistical analysis performed included the Chi-square test and logistic regression. P<0.05 was considered to be statistically significant. **Results:** At the end of the study, 300 participants were enrolled, the majority being females 236 (78.6%). The participants were between 21–70 years (mean±SD = 40±10) of age. The overall prevalence of intestinal parasites was 82.6% (95% CI: 78.4 – 87.0). The prevalence of infection was associated with age, being more prevalent in the age group 51–60 years (p=0.032). Intestinal protozoa were more prevalent than intestinal helminthes (74.3% vs 11.3%). The parasites isolated included: *Cryptosporidium parvum* (44.0%), *Blastocystis hominis* (25.0%), *Microsporidium* spp. (21.0%), *Entamoeba histolytica* (7.3%), *Ascaris lumbricoïdes* (4.3%), *Isospora belli* (4.3%), *Trichuris trichiura* (2.3%), hookworm (2.7%), *Hymenolepis nana* (1.3%), *Strongyloïdes stercoralis* (0.7%), *Cyclospora cayetatensis* (3.7%) and *Giardia lamblia* (3.3%). The predisposing factors for infection with intestinal parasites included poor educational background (OR=0.33, p=0.02), unskilled worker (OR=0.27, p=0.04), well as a source of drinking water (OR=2.6, p=0.03), and living with cats as pets (OR=3.06, p=0.01). **Conclusion:** A very high prevalence of intestinal parasitic infection was observed in PLHWA. Major predisposing factors for intestinal parasites infection included primary level of education, ownership of cats as pets, wells as source of drinking water and having a blue collar job. Routine screening for intestinal parasites should be instituted as part of HIV care in Fako division of Cameroon to improve on the management of HIV.

Keywords: *HIV, protozoa, helminthes, prevalence, predisposing factors, Fako Division, cameroon*

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1. Background

Intestinal parasites are highly endemic in Sub-Saharan Africa [1], a region which is currently experiencing the highest burden of HIV/AIDS [1]. With the overlap in the distribution of both diseases, co-infection with one or more intestinal parasites and HIV is therefore common in the region [2]. The incidence of Intestinal parasitic infections is approximately 50% in developed countries whereas it reaches up to 95% in developing countries [2]. Cameroon is among the Sub-Saharan African countries with an overlapping high rate of HIV and intestinal parasitic infections. The prevalence of intestinal parasitic infection observed in Cameroon ranges from 27.9% reported by Lehman et al. [3] in Douala (in the Littoral region) to 57.48% reported by Marius et al. [4] in Yaounde (in the Centre region). Information on the prevalence of parasitic infections and HIV coinfection are not readily available from the other regions of the country.The presence of opportunistic parasites such as *Cryptosporidium parvum*, *Cyclospora cayetanensis*, *Isospora belli* and *Microsporidia* are well documented in patients with HIV/AIDS [5]. Non opportunistic parasites such as *Entamoeba histolytica, Giardia lamblia, Trichuris*

trichura, Ascaris lumbricoides, Strongyloides stercoralis and *Ancylostoma duodenale* are frequently encountered in developing countries but are not currently considered opportunistic in HIV/AIDS patient [6]. Many factors have contributed to the increase and reinvasion of newly emerging intestinal parasites including the increasing migration of people due to political instability, war, economical problems, and travel to developing countries [7]. Also poverty and malnutrition are some of the factors that contribute to concomitant infections of both HIV and intestinal parasites in Sub-Saharan Africa [8].

The link between intestinal parasites and HIV/AIDS is thought to be bidirectional, with the presence of concomitant infections skewing the immune response of the host towards predominantly a type-2 immune response (Th2) as well as an increase in the immune system activation [9,10,11]. The presence of chronic immune activation and a dominant Th2 immune response may increase the host susceptibility thereby promoting HIV infection and disease progression [12,13,14]. On the other hand, the progressing development of AIDS in HIV is a risk factor for the acquisition of other infective agents including parasites which are the proximate cause of death of AIDS patients [15,16,17]. The effect of intestinal parasites on the HIV RNA levels is not clear either with reports suggesting that treatment of intestinal parasitic infections may reduce HIV RNA levels [18].

Whatever the outcome may be of intestinal parasitic and HIV co-infection, it is obvious that co-infection between these pathogens is common particularly in developing countries where the factors favoring transmission of parasites are all in place This negatively impacts on the health status and outlook of people living with HIV/AIDS [19]. Understanding the actual burden of intestinal parasites in HIV is therefore imperative in improving on the management of HIV/AIDS.

The objectives of this study performed in Fako division of the South West region was to i) determine the prevalence of intestinal parasitic infection in people living with HIV/AIDS in the region, and ii) assess the predisposing factors for infection with intestinal parasites in the target group.

2. Materials and Methods

2.1. Study Area

This is part of the study conducted on "Intestinal parasitic infections in HIV/AIDS patients attending treatment centers in Fako-Division Cameroon: prevalence and predisposing factors" at the Faculty of Health Sciences, University of Buea, Cameroon. The other part of the study has been reported elsewhere [20]. Briefly, participants were enrolled in the HIV treatment centers in the Regional Hospital of Buea and Limbe found in Buea and Limbe respectively. Buea and Limbe are the two major urban centers in Fako division in the South West Region. These two HIV treatment centers are the only two centers in Fako serving the entire population of Fako. Limbe is the head quarter of the Cameroon Development Corporation (CDC), one of the largest employers in Cameroon after the government. These two towns attract people from other parts of the country for work, education or leisure. With a prevalence of 8%, the south west region has one of the highest HIV prevalence in the country [21].

2.2. Study Design and Setting

This was a hospital based cross-sectional study in which participants of the Buea and Limbe Regional Hospitals HIV treatment centers were enrolled between April and July 2014.

2.3. Sampling Technique

A time limited sampling technique was used, where patients were consecutively recruited into the study. Participants were enrolled into the study provided they gave their consent and met the inclusion criteria.

2.4. Study Population

HIV patients greater than or equal to 21 years (according to Cameroon´s cut-off age for adulthood) attending the HIV treatment centers of the Regional Hospitals in Buea and Limbe were approached to take part in the study. Participants were selected based on whether they met the inclusion criteria HIV patients irrespective of their ages and gender were eligible to participate. Patients were randomly recruited into the study from these treatment centers until sample size of 313 was attained from April to July 2014. Although there was a respondent rate of 95%, up to 8 participants filled questionnaires with the investigator and provided only blood samples but did not provide stool samples while the remaining 5 provided stool samples, incomplete questionnaire information and no venipuncture. Thus a total sample size of n=300 participants was attained. Excluded from the study were individuals that were on any anti-parasitic medication. Children were also excluded from the study due the low prevalence of HIV in this age group.

2.5. Study Variables

The study participants were grouped as follows: Urban or rural location, those with ≥ 21 years, their level of education, their occupation (blue collar and white collar jobs): Blue-collar jobs (Participants who do manual work with hands) and white-collar jobs (participants who do work which do not involve manual labour) and personal/environmental hygienic practices.

2.6. Data Collection

2.6.1. Administration of Questionnaire

The data collection has been described elsewhere [20]. Briefly, a self-administered structured paper based questionnaire was issued to consented participants to obtain information on the socio-demographic, medical history and potential risk factors related to food-borne, waterborne, person-to-person, and zoonotic transmission of intestinal parasites. The questionnaires were explained in Pidgin English to those to those who were not literate. The participants guided on how to answer the questionnaire

and were expected to summit the completed questionnaire prior to specimen collection.

2.7. Specimen Collection

The participants were provided with a wide open neck, clean and sterile container to provide stool specimen. The participants were instructed on how to produce stool specimen and also to take it to the laboratory within 1hr of time of collection.

2.8. Analysis of Stool Specimen

Stool specimens were analysed using the Kato-katz and formol ether concentration techniques, and smears were examined using the modified field and modifies Ziehl-Neelsen staining techniques.

Formol ether concentration technique: Using an applicator stick, about one gram of stool was emulsified in about 7ml of 10% formol water in a screw-cap tube. The proceeding was done as described by Cheesbrough [22]. In this techniques we were able to detect both protozoa cysts, oocysts and helminthes eggs. Also this technique has as prediction to concentrate protozoa cysts, oocysts and helminthic eggs.

Kato-Katz Technique: Fresh stool samples were analyzed by the kato-katz technique for the qualitative and quantitative detection of helminthes eggs [22].

Smears prepared from the formol ether concentration techniques were stained using the Modified field staining technique for the detection of spores of *Microsporidium* species [23], and the Modified Ziehl-Neelsen staining technique for the detection of oocysts of *Cryptosporidium* species, *Isospora belli*, and *Cyclospora cayetanensis* [24].

2.9. Ethical Considerations

Ethical Clearance was obtained from the Institutional Review Board of the Faculty of Health Sciences of the University of Buea. Administrative authorization was obtained from the South West Regional Delegation of the Ministry of Public Health and the District Health Service. The purpose of the study alongside the role of the participants were well explained in the consent form to the participants in English, French and the local Pidgin English languages and participation could only take place after the participant had read and signed the informed consent form voluntary.

2.10. Statistical Analysis

Data were analysed using SPSS Version 20.0. Armonk, NY: IBM Corp. Statistics performed included the Chi-square test for group comparison and the logistic regression analysis for the measure of association. P-value less than 0.05 considered as statistically significant at 95% confidence interval (CI). Where appropriate the Fisher exact test was used to determine p-values.

3. Results

By the end of the study, 300 participants successfully summited completed questionnaire and stool specimen. Among them were 21.3% males and 78.6% females. The ages of the participants ranged between 21-70 years (mean ± SD = 40±10). Majority of the participants were from rural 54% than urban 46% settings.

The overall prevalence of intestinal parasitic infection in HIV/AIDS patients observed in this study was 82.6%, 95% CI: 78.4 – 87.0. Intestinal protozoa were more prevalent than intestinal helminthes [74.3% vs. 11.3%]. The most common parasitic infection was *Crptosporidium parvum* 44%, while the least prevalent was *Strongyloides stercoralis* 0.7% (Figure 1).

In this study, the prevalence of intestinal parasitic infection was higher in females 84.3% than in males 76.5% (Figure 2). However this difference was not observed to be significant (p = 0.14). The prevalence of intestinal parasites was higher in HIV/AIDS patients resident in rural areas 84.4% than those residing in urban centers 80.5% (Figure 2). But the difference was not statistically significant (p = 0.54). The prevalence of intestinal parasites was highest in the age group 51 – 60years (90.3%) (Figure 2). A significant association was observed in the prevalence of intestinal parasites with age (p = 0.032).

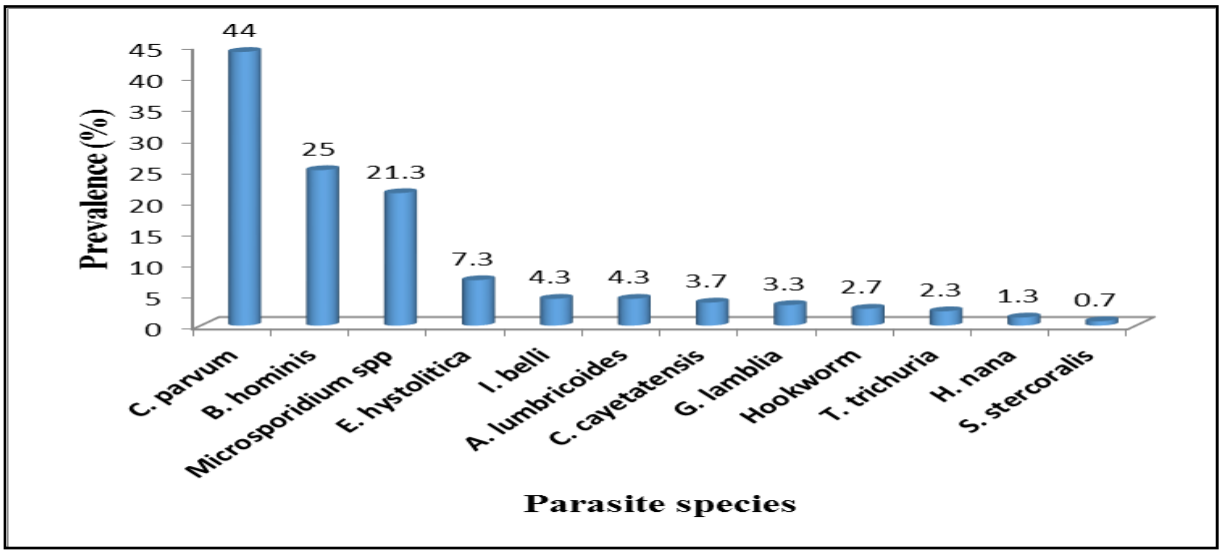

Figure 1. Prevalence of Intestinal Parasites in the HIV/AIDS patients from April to July 2014

Predisposing factors for the infection with intestinal parasites included: the level of education, being common in HIV patients with primary education or less (p = 0.02), unskilled profession (p = 0.04), wells as a source of drinking water (p = 0.03), and the presence of cats as pets (p = 0.01) (Table 1).

Table 1. Predisposing factors for intestinal parasitic infections among HIV patients in Fako Division (n=300)

Demographic variables	Number Examined n(%) N=300	Number infected with any parasites n(%)	COR	95%CI	P-values
Gender					
Male	64(21.3)	49(76.5)	1		-
Female	236(78.7)	199(84.3)	0.607	0.308-1.194	0.14
Location					
Urban	139(46.3)	112(80.5)	1		-
Rural	161(53.7)	135(84.4)	0.798	0.44-1.44	0.45
Age group					
21 – 30	45(15.0)	36(80.0)	1		-
31 – 40	118(39.3)	99(83.9)	0.76	0.31-1.85	0.55
41 – 50	90(30.0)	71(78.8)	1.07	0.44-2.60	0.88
51 – 60	31(10.3)	28(90.3)	0.42	0.10-1.73	0.33
61 – 70	16(5.3)	14(87.5)	0.57	0.11-2.98	0.71
LEVEL OF EDUCATION					
Tertiary	20(6.7)	13(65)	1		-
Secondary	111(37.0)	92(82.8)	0.38	0.13-1.08	0.06
≤ primary	169(56.3)	143(84.6)	0.33	0.12-0.92	**0.02**
OCCUPATION					
Students	12(4.0)	7(58.3)	1		-
White collar	231(77.0)	193(83.5)	0.27	0.08-0.91	**0.04**
Blue collar	40(13.3)	34(85.0)	0.24	0.05-1.04	0.09
Unemployed	17(5.7)	14(82.3)	0.30	0.05-1.63	0.21
INCOME LEVELS (FRS)					
>50000	80(26.7)	62(77.5)	1		-
<50000	220(73.3)	186(84.5)	0.62	0.33-1.19	0.15
PERSONS PER HOUSEHOLD					
< 5	152(50.7)	123(80.9)	1		-
≥ 5	148(49.3)	125(84.5)	1.28	0.70-2.33	0.41
SOURCE OF DRINKING WATER					
Pipe borne	247(82.3)	206(83.4)	1		-
Well	23(7.7)	15(65.2)	2.60	1.06-6.73	**0.03**
Stream	30(10.0)	27(90.0)	0.55	0.16-1.92	0.43
PRESENCE OF TOILET					
Private Water closet	57(19.0)	44(77.2)	1		-
Private pit latrine	128(42.7)	108(84.3)	0.62	0.28-1.36	0.24
Public pit latrine	111(37.0)	94(84.6)	0.61	0.27-1.37	0.23
Open field(no toilets)	4(1.3)	2(50)	3.38	0.43- 26.4	0.22
HAND WASHING HABIT Before Eating					
Always	84(28.0)	68(80.9)	1		-
Sometimes	143(47.7)	123(86.0)	0.69	0.33-1.42	0.31
Never	73(24.3)	57(78.0)	1.19	0.54-2.59	0.65
SOURCE OF FOOD					
Home cooked food	167(55.7)	143(85.6)	1		-
Road side foods	39(13.0)	30(76.9)	1.78	0.75-4.22	0.18
HCF+RSF	94(31.3)	75(79.7)	1.50	0.77-2.93	0.22
PRESENCE OF PETS					
None	220(73.3)	184(83.6)	1		
Dogs	56(18.7)	49(87.5)	0.73	0.30-1.74	0.47
Cats	24(8.0)	15(62.5)	3.06	1.24-7.54	**0.01**
ANIMAL REARING					
None	149(49.7)	117(78.5)	1		-
Goats	46(15.3)	42(91.3)	0.34	0.11-1.04	0.05
Fowls	68(22.7)	58(85.2)	0.63	0.28-1.37	0.24
Pigs	34(11.3)	29(85.2)	0.63	0.22-1.75	0.37
Rabbits	3(1.0)	2(66.6)	1.82	0.16-20.8	0.52

CI = Confidence Interval; COR = Crude Odd Ratio.

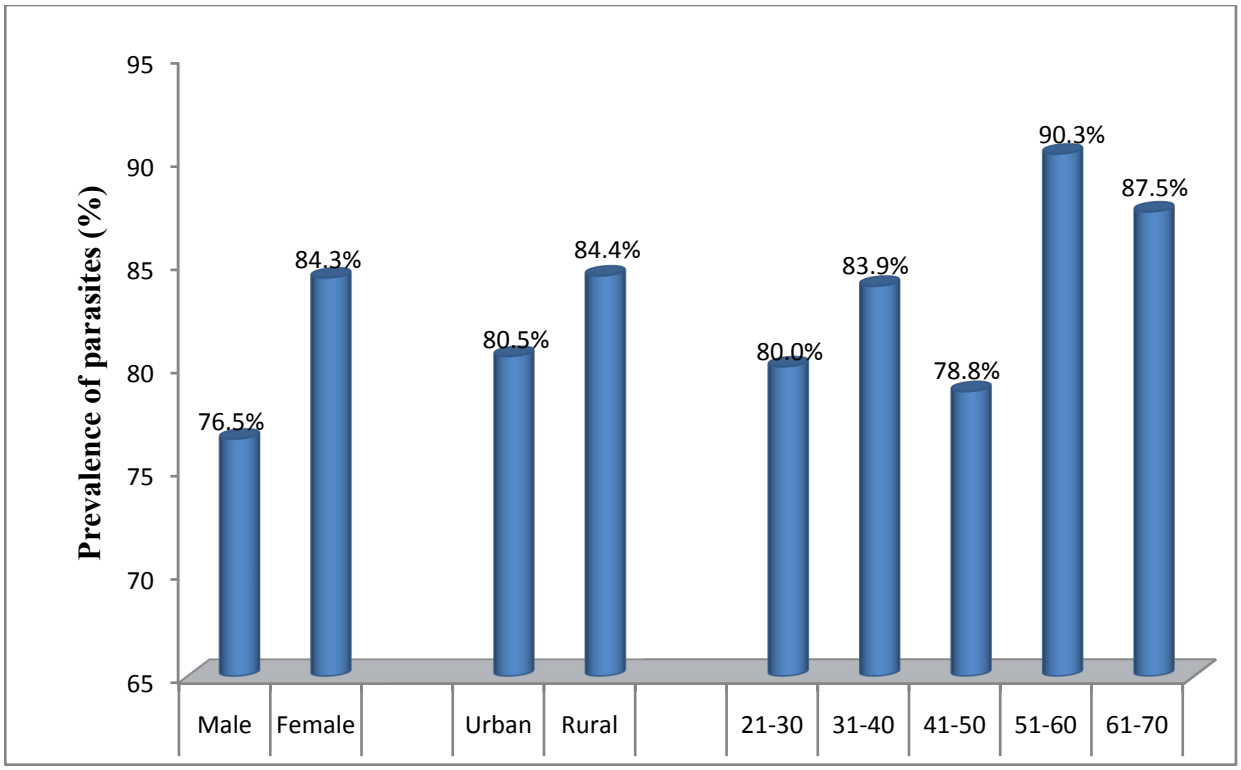

Figure 2. Intestinal parasites in study participants based on gender, age and location in HIV/AIDS patients Fako Division, Cameroon

4. Discussion

There is a high burden of parasitic infections in Fako Division in the South West Region of Cameroon. The interaction between HIV and other infective agents including parasites have long been recognised to adversely influence the health status of people living with HIV/AIDS. Understanding the epidemiology of intestinal parasitic infection is essential for the effective management of HIV infection. Data on the prevalence of intestinal parasitic infection in people living with HIV/AIDS in Cameroon are sparse. Overall, a prevalence of 82.6% was observed in this study, which is higher compared to the 27.9%, 33%, and 57.48% observed in studies performed elsewhere in Cameroon by Lehman *et al*. [3], Sarfati *et al*. [25], and Marius *et al*. [4] respectively. The differences in the prevalence reported in these studies and the present study could be attributed to the difference in the methodology. In this study, intestinal parasites were isolated using the formol ether concentration and the kato-katz techniques in addition to methods involving staining with the modified field stain and the modified Ziehl-Neelsen stain meanwhile the other studies performed elsewhere at best involved the use of only one concentration and one staining technique. The prevalence of intestinal parasites was also high compared to that reported in other countries, 27.9% in Apulia, Italy [26] and the 28.1% and 42.9% observed in Abuja and Abeokuta respectively in Nigeria [27]. However, the prevalence observed in this study was similar to the 81.8% reported in Tanzania [28] and the 79.3% in Osun state of Nigeria [29].

In this study, the prevalence of intestinal parasites was higher in females (84.3%) than in males (76.5%). However, the difference was not observed to be statistically significant ($p = 0.14$). This finding is similar to other studies performed elsewhere [7,26,28]. This could be explained in that females are the ones more engaged with domestic chores and farming which exposes them to intestinal parasites. Contrary to this, is a study performed in Nigeria in which the prevalence was observed to be higher in males than in females [30]. This may be due to differences in the study area, study population and the activities of these males which expose them than the females. A significant association was observed between the prevalence of intestinal parasites and age ($p = 0.032$), with the prevalence being higher in the age group 51–60 year. This is in accordance with a study performed in Nigeria [31]. This high rate in patients above 50 years could be attributed to the immunosuppression due to the presence of HIV/AIDS coupled to the weakened immune system associated with ageing.

In this study, intestinal protozoa (74.3%) were more prevalent that intestinal helminthes (11.3%). The prevalence of intestinal protozoa observed in this study was lower compared to the 80.8% reported in Kenya [32]. Protozoa infection varies per location and hygienic practices differences in prevalence may be also due to differences in study population. The prevalence of intestinal helminthes observed in this study was lower compared to the 37.04% observed in Ethiopia [8], but higher than the prevalence reported by Njunda *et al*. [33] in Yaounde-Cameroon. Soil transmitted helminthes is very common with children. However, children were not included in our study as with other studies which explains the difference in the prevalence of intestinal helminthes. The most prevalent intestinal protozoa was *Cryptosporidium parvum* (44%) meanwhile *Ascaris lumbricoides* (4.3%) was the most prevalent helminthes. The prevalence of *Cryptosporidium parvum* was higher than that reported elsewhere in Cameroon [3,4,25]. *Cryptosporidium parvum* is mostly associated with unclean drinking water sources. Shortage of drinking water supply is common in the study area.

Hence, this promotes use of unclean drinking water sources by the inhabitants thus, further predisposes them to this parasite especially in the immunocompromised individual.

Predisposing factors for infection with intestinal parasites observed in this study included the level of education, the prevalence was observed to be higher in individuals with only primary education. Ostan *et al*. [34] had demonstrated the relationship that exists between increasing level of education and decreasing incidence of intestinal parasites. Other factors included the occupation of the individual whereby the prevalence was observed to be higher in unskilled workers which confirms the findings of Assefa *et al*. [8]. The source of drinking water was also significant risk factors for the infection, with intestinal parasites more common in individuals who used water from wells for drinking. This finding is similar to that reported in Ethiopia [35]. This could be attributed to the poor quality of the wells which are often not dug very deep due to the stony nature of the terrain in the study area and as such are easily contaminated by sewage and runoff. Ownership of cats as pets was also observed to be a significant risk factor for infection with intestinal parasites. The plausibility of *Cryptosporidium* being passed from cats to humans has been suggested in a few earlier studies. For example, in 1983 Koch *et al.* [36] described cryptosporidiosis in an immunocompromised patient whose cat also harbored *Cryptosporidium* oocysts. Similarly, Egger *et al*. [37] reported a case in which an 8-year-old boy contracted cryptosporidiosis during a visit to a farm; *Cryptosporidium* oocysts were found in a cat but not in the investigated calves on the farm, and thus the authors suggested that the cat was the source of infection. However, neither of these cases was confirmed by molecular methods. Another case of unusual zoonotic *Cryptosporidium* transmission has been described from Peru [38]. In a recent study, the HSP70 sequences from the cat and the patient were 100% identical to each other (1,885/1,885 bp), but identical sequences were also found in most of the other investigated *C. felis* isolates from cat and humans [39].

In this study, rearing of domestic animals was not observed to be a predisposing factor for infection with intestinal parasites which is unusual owing to the fact that people in the study area especially in the rural settings are farmers, rearing animals such as goats, rabbits, chicken etc. Thus they come in contact with their lither thereby exposing them to infection with intestinal parasites. This finding is contrary to studies performed elsewhere [40,41,42,43]. Studies using molecular techniques are required in the area to investigate the presence of zoonotic transmission of intestinal parasites.

Although this study provides data on the actual burden of intestinal parasitic infections in people living with HIV/AIDS in Fako Division which could impact the management of HIV in the area, it was subject to several limitations. Firstly, the result only represent people living with HIV/AIDS in Fako Division and may not be generalized to all HIV – infected persons in Cameroon. Secondly the questionnaires used to determine the risk factors for intestinal parasites was self-administered which could have affected the accuracy in determining the predisposing factors for the transmission of intestinal parasites in the study population. However to minimize this, questions and benefits of the study were well explained to the participants prior to response.

5. Conclusion

A high prevalence of intestinal parasites was observed in people living with HIV/AIDS in Fako division of Cameroon. A significant association was observed between the prevalence of intestinal parasites with age, being more prevalent in individuals between the ages of 51 – 60. Overall, *Cryptosporidium parvum* was observed to be the most prevalent intestinal parasite meanwhile *Ascaris lumbricoides* was the most prevalent helminthes infection. The major predisposing factors for infection with intestinal parasites included having a poor educational background, ownership of cats as pets, use of wells as a source of drinking water and being blue collar workers. Intestinal parasitic infections are still huge burden in HIV/AIDS patients in the Fako Division. This implies parasitic co-infections in HIV/AIDS individual should not be neglected whether they are symptomatic or asymptomatic. Continues screening and public health surveillance systems should be put in place to combat this dilemma. In addition to the health talks by health care providers on anti-retroviral therapy, talks on basic hygienic practices should be enhanced. There should be a continuous surveillance of intestinal parasites in HIV/AIDS patients to check changes in the fauna of infection for proper laboratory testing and management of these patients. Due to the importance of knowledge on microorganisms implicated in infectious diarrhea and on the individual and collective impact of infection, there is a need for more studies on this subject especially at the molecular level.

List of Abbreviations

ARV	Antiretroviral
ART	Antiretroviral Therapy
HIV/AIDS	Human Immunodeficiency Virus/Acquired Immunodeficiency syndrome
IPIs	Intestinal parasitic infections
PLHIV	People living with HIV
HAART	Highly Active Antiretroviral Therapy
VCT	Voluntary counseling and testing
WHO	World Health Organization
EPG	Egg per gram

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Conflict of Interest

The authors declare that they have no competing interest.

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