Original article

Diarrhoea-associated parasitic infectious agents in AIDS patients within selected Addis Ababa Hospitals

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Abstract: The aim of the study was to determine the prevalence of gastrointestinal parasites in Acquired Immunodeficiency Syndrome (AIDS) patients with chronic diarrhea. This prevalence was compared with two control groups: Human Immunodeficiency Virus (HIV) seronegative diarrheal patients and HIV seropositive individuals without diarrhoea. Stool specimens from clinically diagnosed hospitalized AIDS patients in some hospitals in Addis Ababa were screened for parasite infection. Of 147 AIDS patients with chronic diarrhoea, 74 (50.3%) were infected with one kind or more of parasites. Out of 56 non-AIDS (seronegative) diarrhoeal patients, 41.1% (23/56) and out of the 43 non-diarrhoeal (seropositive) patients, 41.9% (18/43) were infected by a variety of intestinal protozoa and helminths. The parasites detected in AIDS patients were *Cryptosporidium* spp, *Isospora* spp, *Blastocystis* spp, *Ascaris lumbricoides*, *Giardia lamblia*, *Strongyloides stercoralis*, *Taenia spp*, *Trichuris trichiura*, *Entamoeba histolytica*, and *Hook worm spp*. Among the intestinal parasites, *Cryptosporidium spp* was exclusively associated with AIDS patients. The high proportion of the study subjects who had diarrhoea in the absence of identifiable parasitic infections suggests that other infectious agents (eg. Bacteria and Virus) or mechanisms other than infectious agents, are responsible for the diarrhoea. [*Ethiop. J. Health Dev.* 1999;13(3):169-173]

Introduction

Diarrhoea is the clinical manifestation of HIV-1 infection in both developing and developed countries. In tropical countries chronic diarrhoea associated with weight loss ("slim disease") is often the presenting illness of HIV-1 infected individuals (1). This diarrhoea-wasting syndrome in association with a positive HIV-1 serology test is an AIDS-defining illness in the World Health Organization's classification (2,3).

The acquired immunodeficiency syndrome (AIDS) caused by human immunodeficiency virus (HIV) infection predisposes to several parasitic diseases such as *Pneumocystis carnii* pneumonia and *Toxoplasma* encephalitis. Among intestinal parasites, the intracellular coccidia, *Isospora belli*, and *Cryptosporidium parvum* are clearly opportunistic (4). The microsporidian *Enterocytozoon bieneusi* may also be a significant enteric pathogen (5).

Investigations for intestinal parasitic infections in AIDS patients in Africa has focused on patients with diarrhoea. *Isospora* and *Crypotosporidium* have been found consistently in such patients at prevalence rates varying from 8% to 32% and 12% to 19%, respectively, among patients with chronic diarrhoea (1,6,7,14). In nearly all studies, opportunistic infections were more common than the more commonly encountered parasitic protozoa and helminths which are probably more prevalent in the general community (17). Most of the clinical manifestations of HIV-1 infections result either from the reactivation of preexisting latent pathogens or exposure to locally predominant pathogens. Consequently, clinical presentations of AIDS and the pathogens responsible in different geographical areas reflect the differing prevalence of endogenous infections. On the other hand, some reports

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indicate that there is no convincing evidence on the etiological agents isolated from African AIDS

patients with diarrhoea that their prevalence or incidence is increasing due to the HIV epidemic (15).

Patterns in developing countries, where hygiene is poor and intercurrent infection rates are high, differ in several important ways from patients in the industrialized countries. The commonest presentation of AIDS in Africa is 'slim disease' or enteropathic AIDS, which is characterized by severe loss of weight and chronic watery diarrhoea and prolonged fever (9,16,17). The role of HIV in diarrhoea, malabsorption, and other gastrointestinal disorders has been elucidated (8). However, the presence of the virus in bowel mucosa of individuals with symptoms and no known pathogen suggests that HIV could give rise to this disorder. In particular, HIV infection of neuroendocrine cells (eg. Enterocromiffin cells) could result in disorders of intestinal motility and function (8,9,16).

Intestinal nematodes, amoebae, and other protozoa have not been shown to be opportunistic in AIDS patients. There is no convincing evidence to show that, for parasitic diarrhoeal infections, their prevalence or incidence is increasing due to the HIV epidemic (13,15). For reasons which are unclear, parasites such as *S. stercoralis* and *E. histolytica*, where cell-mediated immune response is thought to be significant, do not appear to be opportunist infections (13). Although strongloidiasis and AIDS often coexist in regions of endemicity such as Central Africa, only few cases of disseminated disease in people infected with HIV have been reported (10-13).

The aim of this study was to determine the prevalence of gastrointestinal parasitic agents in AIDS patients with chronic diarrhoea.

Methods

The study was conducted between January 1995 and May 1996. The sample size for the study was computed by considering a 95% level of confidence, 80% power and an expected ratio of 1:3 (Not ill/ill). Two hundred forty six stool samples were examined in this study, 147 from AIDS patients with chronic diarrhoea, 56 from HIV seronegative diarrhoeal patients (control group I) and 43 from HIV seropositives without diarrhoea (control group II). During the study period, patients (cases, control groups I and II) visiting the study sites were included until the required sample size was reached. A questionnaire relating to the history and clinical status was completed for each subject included in the study by the AIDS coordinator physicians and trained counselors in hospitals where this study was conducted. Statistical analysis was performed using EPI-INFO software to test differences between the cases and the two control groups.

In the hospital laboratories, 10 ml of venous blood was taken from each patient using a vacuum system. Serological testing for HIV-1 antibodies was done with the Vironostika Uniform II (Organon Teknika, Holland). Reactive sera were retested using Wellcozyme ELISA (Murex, UK) (18,19) in the National Referral Laboratory for AIDS (NRLA) which is located at the Ethiopian Health and Nutrition Research Institute (EHNRI). Stool specimens were collected fresh and processed in the clinical parasitology laboratory of the EHNRI within four hours of collection. Parasite detection was made by examination of stool specimens by the following methods: fresh hospital specimens were first examined as wet mounts using normal saline followed by Formal-ether concentration (1). Air-dried thin smears were stained by modified Ziehl-Neelsen technique (20) for *Cryptosporidium* and *Isospora* oocysts. The smears were fixed in methanol for three minutes, stained with carbol fuschin for 60 minutes, decolorized in 2% H_2SO_4 for one minute, washed in running tap water, counter-stained with 5% malachite green for five minutes, washed in running tap water, and air dried. The thin-walled oocysts stain an intense red and are easily recognized against the green background under high power or oil immersion for detailed visualization.

Results

The age profile of the study and control subjects is similar to the HIV/AIDS profile in the general population reported by the Ministry of Health (21) and the normal population pattern reported by the Central Statistical Authority (22), where both young and older individuals are represented in the

sample (Table 1).

Table 1: **Age and gender distribution of the study population (cases) and the control groups (controls I and II), in selected Addis Ababa hospitals, 1995/96.**

Age (year)	Sex	Cases No. (%)	Control I No. (%)	Control II No. (%)
15-24	M	7(4.8)	5(8.9)	5(11.6)
	F	9(6.1)	5(8.9)	3(7.0)
25-34	M	62(42.2)	20(35.7)	16(37.2)
	F	26(17.9)	12(21.4)	8(18.6)
35-44	M	21(14.3)	7(12.5)	4(9.3)
	F	7(4.8)	3(5.4)	2(4.7)
>45	M	12(8.2)	3(5.4)	4(9.3)
	F	2(1.4)	1(1.8)	1(2.3)
Total		47(100)	56(100)	43(100)

Oocysts of *Cryptosporidium species* were isolated from 38/147 (25.9%) AIDS patients with chronic diarrhoea. In addition, among the opportunistic parasites frequently associated with AIDS, 2/147(1.4%) *Isospora belli* and 1/147 (0.7%) *Blastocystis* species were found in 74/147 (50.3%) of the AIDS patients with chronic diarrhoea (Table 2). The only parasite clearly more prevalent in AIDS patients was *Cryptosporidium* (P<0.001), although *Isospora* and *Blastocystis* were also found only in AIDS patients.

Parasites were identified in 74 out of 147 (50.3%) AIDS patients with chronic diarrhoea, and in 18 out of 43 (41.9%) who did not suffer from diarrhoea. Chronic diarrhoea occurred in all 38 patients with cyptosporidiasis which was the only infection clearly associated with this symptom.

The parasites detected in stools of all AIDS hospital patients with diarrhoeal consisted of *A. Lumbricoides* (11.8%), *E. histolytica* (8.2%), G. lamblia (4.1%), *S. stercoralis* (3.4%), Taenia spp (3.4%) and *T. trichiura* (7.5%). These common parasites often occurred as multiple infections (Table 3).

Table 2: **Parasite species identified in AIDS patients with chronic diarrhoea (cases), HIV negative diarrhoeal patients (control I) and HIV seropositive individuals without diarrhoea (control II).**

Parasite species	Cases(N = 147) n (%)	Control I (N = 56) n (%)	Control II(N = 43) n (%)
Cryptosporidium spp	38(25.9)**	0	0
A. lumbricoides	17(11.8)	12(21.4)	7(16.3)
S. stercoralis	5(3.4)	1(1.8)	0
G. lamblia	6(4.1)	2(3.6)	2(4.6)
Isospora belli	2(1.4)	0	1(2.4)
T. trichiura	11(7.5)	7(12.5)	6(14.0)
E. histolytica	12(8.2)	3(5.4)	3(7.0)
Taenia spp	4(3.4)	2(3.6)	3(7.0)
Blastocystis spp	1(0.7)	0	1(2.4)
S. mansoni	0	1(1.8)	1(2.4)
Hook worm spp	0	2(1.8)	2(4.6)
Total infected	74(50.3)	23(41.1)	18(41.9)

** : P<0.001

Table 3: **Prevalence of single and multiple parasitic infections in AIDS patients with diarrhoea and the control groups in selected hospitals in Addis Ababa, 1995/96.**

Subjects	Parasites detected in the study subjects			
	One No. (%)	Two No. (%)	Three No. (%)	Four No. (%)
AIDS patients (n = 74)	54(73.0)	15(20.3)	4(5.4)	1(1.4)
Control group I (n = 23)	16(69.6)	7(30.4)	0	0
Control group II (n = 18)	10(55.6)	8(44.4)	0	0

Discussion

These findings suggest that patients with AIDS do not have more intestinal parasite infections than the control groups. Only *Crytosporidium*, a recognized opportunistic pathogen was more prevalent in the AIDS group in which it was strongly associated with chronic diarrhoea. Associations between AIDS, other parasite infections, and chronic diarrhoea were not evident. Of course, there are other important causes of diarrhoea, such as viral, bacterial and microsporidian infections, for which this study was not designed.

Diagnosed cases of *I. belli* were considerably fewer in this study than reported in Zaire (7%), Uganda (13%), and Zambia (16%), which could possibly be either due to more sensitive detection methods or a reflection of low prevalence in this study population (23). In addition *Blastocystis* species, which has been reported as a causative agent of diarrhoea in a number of cases in patients with AIDS, was detected in one case of AIDS with diarrhoea and in an HIV seropositive individual, but none in HIV seronegative patients with Diarrhoea(24).

The finding suggests that *Blastocystis* species may be associated with HIV infection and AIDS. Therefore, it will be necessary to conduct further studies to determine the importance of this parasite in relation to HIV/AIDS in Ethiopia.

Our findings tend to support the view that the more 'common' parasites *(Ascaris, Strongyloides, G.lamblia, T. trichiura, Taenia and E.histolytica)* are not opportunistic in AIDS patients (13). Identification of these common parasites in up to 12-21% of AIDS patients and the controls is a reflection of poor environmental hygiene.

The high proportion of AIDS patients who had diarrhoea in the absence of identified parasite infections strongly indicates the existence of other diarrheaogenic agents or mechanisms. The detection of these will require more comprehensive and better controlled studies.

In conclusion, diarrhoeal illnesses remain a major clinical problem for patients infected with HIV-1, particularly those with AIDS. Opportunistic enteric pathogens, for which there is no effective treatment, the emergence of new opportunistic infections, and the enlarging pattern of drug resistance continues to be a challenging task. However, better understanding of HIV-1-induced mucosal immunosuppression, sound clinical management, careful diagnostic evaluation, development of newer antimicrobial agents, and judicious patient management should help meeting this challenge and can be necessary in attempting to improve the quality of life.

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