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ASSOCIATION OF MULTIPLE INTESTINAL PARASITOSIS AND SOME SPECIFIC PARASITES WITH HUMAN IMMUNO DEFICIENCY VIRUS SERO-POSITIVE STATUS IN CALABAR

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ABSTRACT

The association between intestinal parasites with Human Immunodeficiency Virus (HIV) seropositive status in Calabar was evaluated using fresh stool samples collected from 175 HIV seropositive subjects. Samples were collected from both male and female subjects with age range of 1-70 years (mean, 35 years) and examined macroscopically for consistency, presence of blood, mucus and adult worms. Microscopic examination was carried out using direct smear method in saline and iodine. Stool concentration was by ether shake technique and modified Ziehl Neelsen method was used for the detection of *Crptosporidium* spp. In all, eight different parasites were detected from both groups of subjects. HIV seropositive subjects had a higher parasite load (4.33 +_ 0.24). Odds ratios associating *Crptosporidium* spp., *Giardia Lambia* and *Strongyloides Stercoralis* with HIV seropositive status were 106.9 (95%CI 4. 11-5.22), 24.0 (95%CI 2.61-3.25) and 22.1 (95%CI 2.25-3.68) respectively. Infections involving four or more parasites were exclusive to the HIV seropositive subjects and was significantly associated ($p<0.001$) with HIV seropositive status. Parasitic infections may be employed for the presumptive diagnosis of HIV infections where clinical syndromes are inapparent especially in the rural settings where HIV test kits may not be available but microscopy is possible.

Key words: Multiple parasitosis, CALABAR, NIGERIA, HIV seropositive.

INTRODUCTION

The clinical expression of HIV infections is not only very diverse but also complex in different populations, and may vary according to the relative frequency of other endemic opportunistic infection (United States Public health Services/Infectious Disease Society of America, 1995, Kelly, 1998, Crowe *et al.*, 1999). Its association with several parasitic agents has been recognized for more than a decade (Luby and Horan, 1994, Fischer *et al.*, 1995, Mocroft *et al.*, 1997). It has been established that as the body immune system wanes as a result of selective destruction of $CD4^+$ lymphocytes by HIV (Carswell, 1993, Huges *et al.*, 1994, Del Rio-Chiroboga *et al.*, 1996), the individual becomes more prone to prevailing endemic infections (Colebunder and Latif, 1991, Carswell, 1993 Huges *et al.*, 1994, Luby and Horan, 1994, Fischer *et al.*, 1995, United States Public health Services/Infectious Disease Society of America, 1995, Del Rio-Chiroboga *et al.*, 1996, Mocroft *et al.*, 1997, Kelly, 1998, Crowe *et al.*, 1999). This problem is further compounded in most third world developing countries like Nigeria where sanitation is often neglected, thus enhancing the endemicity of some of these parasites (Cross.

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1981, Ejezie, 1981, Knotic, 1981 and Ndifon, 1991).

Some intestinal parasites have been listed as indicators of defects in cell-mediated immunity (Pantalco *et al.*, 1993, Pantalco and Fauci, 1996), while a few others are being used for defining a person as having AIDS. But in the immunosupressed patients, they are regarded as opportunistic pathogens (United States Public health Services/Infectious Disease Society of America, 1995, Kelly, 1998, Crowe *et al.*, 1999, Fischer *et al.*, 1995, Mocroft *et al.*, 1997,) This inapparent distinction between an intestinal parasite and commensal may pose some treatment problems, especially where HIV sero-status of the patient is not known or where clinical manifestations of HIV infections are not obvious (Volberding, 1990, Cooper and Lawfon Ar., 1994).

This study is therefore aimed at determining the range of the intestinal parasites of HIV seropositive subjects in Calabar and to assess the relationship between specific intestinal parasites and HIV sero-status of the subjects.

MATERIALS AND METHODS

Single stool samples were collected from a total of 355 subjects seen at the University of Calabar Teaching Hospital (UCTH) AIDS screening centre between March 1996 and May 1999. The subjects were made up of 175 persons who had tested HIV sero-positive based on reactive ELISA test (ELISA WEELCOZYME; WELLCOME BOROUGH, DARTFORD, UK) and 180 HIV sero-negative males and females aged 1-70 years who gave informed consent. In the case of minors informed consent was obtained from their parents.

STOOL EXAMINATION

Stool samples were examined macroscopically for consistency, presence of pus, blood, or adult worms, and microscopically using direct smear technique in saline and iodine to detect trophozoites, cysts, ova and larvae of parasites. Concentration for parasites was done using ether shake method (Robinson, 1968, Nelvin and Brook, 1982). Modified Ziehl Neelsen technique was employed for the detection of *Cryptosporidium* spp. (Baxby and Hart, 1986, Cheesbrough, 1987).

RESULT

Intestinal parasites and their prevalence rates detected in the stool samples of HIV seropositive subjects and their matched controls are presented in table 1.A total of 8 different types of parasites were seen during the study. *Giardia lambia, Cryposporidium* spp. and *Stronyloides stercoralis* were the most prevalent intestinal parasites among HIV seropositive subjects with rates of 76.0%, 70.9% and 69.7% respectively. *Ancylostoma duodenale* (26.1%), *Trichuris trichiura* (18.3%) and *Ascaris lumbricoides* (13.3%) were the most prevalent parasites among the control subjects.

The distribution of multiple parasites infections according to the HIV sero-status of the subjects is presented in table 2. Multiple parasitic infections involving four or more parasites were found in 50(33.3%) of the HIV-positive subjects and none among their matched controls. Similarly, multiple infections of 2 or 3 parasites were found in a high proportion (59.0%) of the HIV-positive subjects as compared to the matched controls with a rate of 26.7%. Statistical analysis using the Chi-square (x^2) revealed a significant association ($p<0.001$) between multiple parasitic infection and HIV infection.

DISCUSSION

This study reveals a high level of parasitosis in both groups of subjects studied. Although we have no baseline data on endemic parasitic infection in this area, high levels of parasitic infections have been reported in some Nigerian communities (Okpala, 1961, Nnokso, 1991, Obianniwe, 1994). Our finding in this study therefore confirms the endemicity of these parasites. The comparative high level of parasitic load of 4.33 ± 0.1 found among the HIV seropositive subjects as against 0.98 ± 0.07 in the control subjects may strengthen the evidence for the influence of cell-mediated immunity on the

TABLE 1: DISTRIBUTION OF INTESTINAL PARASITES ACCORDING TO HIV SERO STATUS OF THE SUBJECTS STUDIED.

Parasitic agents	HIV Seropositive cases		HIV-Negative controls		ODDS Ratio
	No. with	No. without	No. with	No. without	(95% confidence interval)
Ascaris lumbricoides	98	77	24	156	8.3(1.59 – 2.66)
Cryptosporidium spp.	124	51	4	176	106.9(4.11 – 5.22)
Entamoeba histolytica	44	131	20	160	2.68(0.4 – 1.55)
Giardia lamblia	133	42	21	159	24.0(2.61 – 3.75)
Ancylostoma duodenale	105	70	47	133	4.24(1.01 · 1.89)
Strongyloides stercoralis	122	53	17	163	22.1(2.51 – 3.68)
Taenia spp.	18	157	11	169	7.76(0.03 – 1.11)
Trichuris trichiura	113	62	33	137	7.57(1.77 2.27)
Mean (x) no of parasited	4.33 + 0.24		0.98 + 0.07		

TABLE 2: DISTRIBUTION OF MULTIPLE PARASITIC INFECTIONS ACCORDING TO SERO-STATUS OF SUBJECTS.

Profile of Distribution	Number (%) of Subjects involved		
Probability	**HIV positive (+) (n = 175)**	**HIV Negative (-) control (n = 180)**	**chi square (x^2)**
Combination of 5 Parasites [a,b,c,d,e]	9(10.9)	0(0.0)	**P<0.01**
Combination of 4 [a,b,c,d]	31(17.7)	0(0.0)	**P<0.01**
Combination of 3 [a, b,e]	43(24.6)	5(2.8)	P<0.01
Combination of 2 [a ,c,]	46(26.3)	35(19.4)	P<0.01
One parasite only	26(14.9)	37(20.5)	P<0.01
No with ≥ 2 parasites	139(75.4)	37(20.5)	P<0.01

a = *Cryptosporidium* spp. c = *Strongyloides stercoralis* e = *Trichuris trichiura*
b = *Giardia lamblia* d = *Ascaris lumbricoides*

intestinal parasitic load of HIV positive subjects (Colebunder and Latif, 1991, Carswell, 1993, Huges *et al.*, 1994, Luby and Horan, 1994, Fischer *et al.*, 1995, United States Public health Services/Infectious Disease Society of America, 1995, Del Rio-Chiroboga *et al.*, 1996, Mocroft *et* *al.*, 1997, Crowe *et al.*, 1999). This must however be interpreted cautiously as we did not have any information on the subjects CD4$^+$ counts (Carswell, 1993, Luby and Horan, 1994, Crowe *et al.*, 1999), nor the time when they became infected. Therefore we cannot determine whether the parasitic load was entirely due to HIV infection. However, cases of high levels of opportunistic infections have been reported in HIV/AIDS patients (Huges *et al.*, 1994, Kelly, 1998).

Our finding of multiple parasitic infections involving four or more parasites to be exclusive to the HIV positive subjects examined, and our establishment of a statistically significant relationship (p<0.001) between multiple parasitic infection and HIV infection, may be an important instrument for the presumptive diagnosis of HIV in rural communities. In these communities where facilities for HIV diagnosis may be lacking, it may be employed as an informed yardstick for requesting for HIV tests in clinically inapparent cases.

The highly statistically significant association (p<0.001) found in this study between *Cryptosporidium* spp., *Gardia lamblia, Strongyloides stercoralis, Taenia* spp. and *Ascaris lumbricoides* and HIV sero-positive status is in line with the reports of other authors (Huges *et al.*, 1994, Kelly, 1998).

The strong association between *Cryptosporidium* spp. (OR, 106.95:95%CI=4.11-5.22) and HIV confirms previous reports (Baxby and Hart, 1986) and further strengthens the case for employing the presence of this parasite for defining the HIV/AIDS status of an individual. The exact pathogenic mechanisms that may relate *Giardia lambia* and *Strongyloides stercoralis* whose odd ratios were 24.0 (95%CI=2.61-3.75) and 22.1(95%CI=2.51-3.68) respectively with HIV infection, are not immediately clear to us and calls for further research.

The data obtained in this study further provide insight and a simple way of presumptively defining the HIV status of the individuals in the rural communities where serodiagnosis kits may not be available (Berkley, 1989). Interestingly, Stanley (1996) working at the National Institute of Allergy and Infectious Diseases, Bethesda, USA, has observed that activation of host immune system of HIV infected persons by opportunistic infective agents and immunizations tend to boost the replication of HIV in such persons. This observation may form the basis for the strong association of multiple parasitosis with HIV seropositive status in the study population. This may further explain why HIV/AIDS disease typically progresses in areas of the world where a person's immune system may be constantly challenged by parasites and other microorganisms (Stanley, 1996).

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