Am. J. Trop. Med. Hyg., 85(4), 2011, pp. 776–781
doi:10.4269/ajtmh.2011.10-0713

Alterations in T Cell Subsets in Human Immunodeficiency Virus–Infected Adults with Co-infections in Southern Mozambique

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Abstract. T cell activation and depletion of naive T cells are hallmarks of human immunodeficiency virus (HIV) pathogenesis. This study explored the relationships between certain co-infections (including syphilis, hepatitis B and C, human T cell lymphotrophic viruses I and II [HTLV-I/II], Kaposi sarcoma–associated herpesvirus [KSHV], *Plasmodium falciparum* malaria, and tuberculosis), and levels of activated CD8 and CD4 T cell subsets as well as naive and memory CD4 T cells in HIV-infected adults in a rural area of southern Mozambique. We found that syphilis infection and to a lesser extent HTLV-I/II seropositivity were independently associated with higher CD8 T cell activation (CD8+ CD38+ HLA-DR+) whereas only syphilis was associated with higher CD4 T cell activation. Furthermore, KSHV and HTLV-I/II seropositivities were independently associated with a lower percentage of naive CD4 T cells (CD4+ CD45RA+ CD62L+). These results highlight the importance of screening and prompt treatment of syphilis, and raise questions as to whether HIV-positive persons with certain chronic viral co-infections should initiate combined antiretroviral therapy at higher CD4 cell counts.

INTRODUCTION

Persons infected with human immunodeficiency virus (HIV) type 1 and living in sub-Saharan Africa are commonly co-infected with other pathogens, which include viral, bacterial and parasitic microorganisms. The effect of co-infections and their treatment on HIV pathogenesis is the subject of an ongoing debate. Although many studies suggest that certain co-infections increase HIV RNA levels, their impact on HIV/acquired inmmunodeficiency syndrome (AIDS) pathogenesis and disease progression is less clear.[1] The CD4 cell count is often used as a correlate of disease progression but other parameters of T cell populations such as activation or T cell maturity phenotypes may be of equal importance when assessing HIV pathogenesis.

Immune activation is the strongest correlate of HIV/AIDS disease progression in the absence of treatment.[2] It has been hypothesized that HIV-induced immune activation occurs by mechanisms that include translocation of microbial particles from a damaged intestinal lumen to the systemic circulation, activation of macrophages/dendritic cells, decreased activity of regulatory T cells, and increased production of interferon-α. This activation is thought to fuel HIV replication and is absent in non-pathogenic simian immunodeficiency virus infection of sootey mangabeys.[3] Combined antiretroviral therapy (cART) has been shown to effectively reduce CD8 T cell activation in populations in Europe and Africa. However, independently of HIV infection, it has been shown that persons from Africa have a higher proportion of activated circulating T cells and a more mature T cell phenotype than persons from Europe.[4–6] It is unknown what proportion of this higher activation is caused by co-infections (viral infections, tuberculosis, malaria) and what is caused by genetic differences. Because this excess activation may contribute to fuelling HIV pathogenesis, efforts to elucidate whether the control of co-infections can benefit the prognosis of persons infected with HIV is crucial.

In addition to immune activation, HIV infection also severely impairs the pool of naive CD4 T cells, which is thought to lead to a progressive inability to mount responses to novel antigens and to a lower probability of immune reconstitution after cART. Infection of naive CD4 T cells by HIV is rare. However, depletion of naive T cells occurs likely by several simultaneous mechanisms that include decrease in thymic output, sequestration of naive T cells in lymph nodes, and chronic immune activation causing cells to acquire memory phenotypes.[7] The impact of co-infections on the dynamics of naive and memory T cell pools in HIV-infected persons is largely unknown.

This study explored the relationships between the presence of certain co-infections (tuberculosis, syphilis, and *Plasmodium falciparum*) and viral co-infections (human T lymphotropic virus [HTLV]), hepatitis B virus [HBV], hepatitis C virus [HCV], and Kaposi sarcoma-associated herpesvirus [KSHV]), and levels of activated and naive T cell subsets in advanced stage HIV-infected adults in a rural area of southern Mozambique.

METHODS

Study population and patient visits. This study was conducted during April 2006–November 2008 at the Centro de Investigação em Saúde de Manhiça/Manhiça District Hospital in Manhiça District in southern Mozambique. The present study is an ancillary study from a prospective surveillance cohort designed to assess immune reconstitution inflammatory syndrome in this area.[8] Patients attending the HIV/AIDS voluntary counseling and testing services at Manhiça District Hospital and meeting criteria for initiation of cART were invited to participate in the study if they were more than 18 years of age and lived in the study area under demographic surveillance. All patients received clinical care and follow-up for HIV and other co-infections according to national guidelines.

The study was reviewed and approved by the institutional review boards of the Mozambican National Bioethics

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Committee and the Hospital Clinic of Barcelona Ethics Review Committee. Written informed consent was obtained from all study participants.

HIV-1 viral load determinations. Plasma HIV-1 viral load levels were measured by using a reverse transcriptase–polymerase chain reaction technique (Amplicor Monitor-v1.5; Roche, Basel, Switzerland) with cryopreserved samples. The lower limit of detection of the assay was 400 copies/mL.

Immunologic determinations. The CD4 cells were counted by using flow cytometry after staining fresh whole blood samples with labeled antibodies (CD4, CD3, CD8, and CD45 in TruCount tubes; Becton Dickinson Biosciences, San Jose, CA). For analysis, CD4 cell counts were categorized as greater than or equal to or less than 200 cells/μL.

To access the percentage of naive and activated CD4 and CD8 T lymphocytes, cell staining and flow cytometry were performed on fresh peripheral mononuclear blood cells. Activated T cells were defined as those CD8 or CD4 T cells that expressed CD38 and HLA-DR surface markers. All CD38+ HLA-DR+ double positive CD4 or CD8 T cells were included irrespective of intensity of expression (dim or bright). Naive T cells were defined as those CD8 or CD4 T cells that expressed CD45RA and CD62L. Mature memory phenotype CD4 T cells were defined as those that expressed CD45RO. In all analyses, peripheral blood mononuclear cells were first gated on the lymphocyte population by forward and side scatter plot, then on CD4 or CD8 T cells. Only the gated CD4 or CD8 T cells were assessed in the final plots for expression of activation markers (CD38 and HLA-DR), naive markers (CD45RA and CD62L), or a pan-memory marker (CD45RO).

Assessment of co-infections. Analyses for co-infections were performed at the pre-cART visit. For HBV, rapid testing for HBV surface antigen was performed by using the Determine assay (Abbott Laboratories, Abbott Park, IL). For syphilis, rapid plasma reagin was used for screening (Human Diagnostics, Wiesbaden, Germany). All rapid plasma reagin–positive samples were confirmed by a using a treponemal enzyme-linked immunosorbent assay (Trepanostika; (BioMérieux, Boxtel, The Netherlands).

The remaining co-infections were assessed by using the following enzyme-linked immunosorbent assays: HCV (Human Diagnostics), HTLV I/II (HTLV I/II Murex; Abbott Laboratories), lytic antibodies to KSHV (Biotrin, Dublin, Ireland).

Tuberculosis cases included confirmed and suspected cases. Confirmed cases had either a sputum smear–positive result after staining smears with Ziehl-Nielsen stain and a positive result by optical microscopy, or a culture-positive result. Suspected cases had a compatible chest radiograph or clinical response to anti-tuberculosis drugs.

Plasmodium falciparum malaria was diagnosed by using thick blood smears. Briefly, Giemsa-stained thick blood smears were examined by microscopy. Positive slides were those with at least 1 parasite per 100 high-power microscopy fields.

Statistical methods and definitions. Univariable and multivariable linear regression was used to identify factors associated with outcomes expressed as continuous variables. Multivariable linear regression was performed by using a forward-stepwise procedure with $P < 0.10$ from univariable analysis as an entry criteria, and $P > 0.15$ from the Wald test of coefficients as a remove criteria. Results from estimated models were expressed as coefficients and 95% confidence intervals. All univariable coefficients and P values are shown in the tables and those variables retained in the multivariable model after forward stepwise regression are presented. Those variables not retained in the model do not show a P value.

The CD4 cell counts were categorized as immunocompetent (≥ 200 cells/μL) versus immunocompromised (< 200 cells/μL). HIV RNA was categorized as high (≥ 5 $\log_{10}$ copies/mL) versus low (< 5 $\log_{10}$ copies/mL), World Health Organization stage was categorized as AIDS (stages III–IV) versus non-AIDS (stages I–II). Body mass index was categorized as normal (≥ 18.5) versus underweight (< 18.5).

Outcomes used in linear regression analysis were % CD8 T cell activation, % CD4 T cell activation, % naive CD4 T cells, and % memory CD4 T cells. Regression diagnostics showed that the variables for % CD8 and CD4 activation were normally distributed. However, the variables for percent naive and memory CD4 T cells were non-normally distributed. Statistical analysis was performed by using STATA version 9.0 (StataCorp., College Station, TX).

RESULTS

Study patient characteristics and co-infections. Approximately 60% of the 136 patients enrolled were female, and median age at cART initiation was 36 years (Table 1). Most patients (75.2%) started cART with a CD4 cell count < 200 cells/μL, 73.7% were in an advanced disease stage (WHO stage III–IV), and more than 95% had HIV RNA levels greater than 10^5 copies/mL. One hundred-thirty patients with all baseline measurements were assessed in this study.

At pre-cART visit, tuberculosis, the most common co-infection in HIV patients in sub-Saharan Africa, was present in one-third of the patients (Table 2). Approximately 9.1% (95% confidence interval [CI] = 4.8–15.3%) of the patients had diagnosis of syphilis. *Plasmodium falciparum* malaria was present in 5.1% (95% CI = 1.4–8.9%) of the patients. Viral infections assessed were HCV, HBV, KSHV, and HTLV-I/II. The most prevalent viral co-infection was KSHV, which had a seroprevalence of 44.7% (95% CI = 36.0–53.6%). Hepatitis B surface antigen was present in 13.6% (95% CI = 8.3–20.7%) of the patients, and HCV seropositivity was present in 6.1%

TABLE 1
Clinical and immunologic characteristics of the study population, Mozambique*

Characteristic	No. (%), n = 130
Sex	
M	55 (41.3)
F	78 (58.7)
WHO stage	
I–II	35 (26.3)
III–IV	98 (73.7)
CD4 cell count < 200/μL	100 (75.2)
	Median (IQR)
Age, years	36 (28–45)
HIV RNA $\log_{10}$ copies/mL	5.1 (4.8–5.4)
CD4 parameters	
Counts, cells/μL	134 (71–199)
% Activated	33.7 (24.6–46.0)
% Naive	7.5 (2.4–17.5)
% Memory	87.7 (73.2–93.9)
CD8 parameters	
Counts, cells/μL	586 (239–1,125)
% Activated	66.1 (57.9–75.5)

*WHO = World Health Organization; IQR = interquartile range; HIV = human immunodeficiency virus.

Table 2
Prevalence of co-infections in HIV-infected adults at pre-cART visit, by serologic results, unless otherwise indicated, Mozambique*

Co-infection	No. positive/no. tested	%	95% CI
HBV†	18/132	13.6	8.3–20.7
HCV	8/132	6.1	2.7–11.6
HTLV-I/II	14/132	10.6	5.9–17.2
KSHV	59/132	44.7	36.0–53.6
Syphilis	12/132	9.1	4.8–15.3
Tuberculosis‡	40/136	29.4	21.7–37.2
Pf malaria‡	7/136	5.1	1.4–8.9

Serologic results were available for 132 of 136 eligible patients. HIV = human immunodeficiency virus; cART = combined antiretroviral therapy; HBV = hepatitis B virus; HCV = hepatitis C virus, HTLV = human T cell lymphotropic virus, KSHV = Kaposi sarcoma-associated herpesvirus; Pf = *Plasmodium falciparum*.
† Rapid test for hepatitis B surface antigen.
‡ See Methods for diagnosis.

(95% CI = 2.7–11.6%). There were no double viral hepatitis infections. Approximately 10.6% (95% CI = 5.9–17.2%) of the patients had positive serologic results for HTLV-I/II.

Association of co-infections with level of pre-cART T cell activation. To determine whether co-infections was associated with pre-cART CD8 or CD4 T cell activation, univariable and multivariable linear regression was performed. At the pre-cART visit, the median baseline percentage of activated CD8 T cells and of activated CD4 T cells was 66.1% (interquartile range [IQR] = 57.9–75.5%) and 33.7% (IQR = 24.6–46.0%), respectively (Table 1). Univariable linear regression analysis showed that HIV RNA ≥ 5 $\log_{10}$ copies/mL, tuberculosis, positive serologic results for HTLV, syphilis infection, and body mass index ≤ 18.5 showed a significant or borderline association with a higher level of pre-cART CD8 T cell activation (Table 3).

Multivariable stepwise linear regression included variables shown in Table 3 with a univariable P value < 0.10 and CD4 cell counts. Variables found to be independently associated with higher CD8 T cell activation included HIV RNA ≥ 5 $\log_{10}$ copies/mL, syphilis, and positive serologic results for HTLV. A positive syphilis test result was associated with an 11.2% increase in CD8 T cell activation. Seropositivity for HTLV was associated with a 7.1% increase in CD8 T cell activation, and having an HIV RNA level ≥ 5 $\log_{10}$ copies/mL was associated with a 25.1% increase in CD8 T cell activation as compared to HIV RNA level < 5 $\log_{10}$ copies/mL (Table 3). None of the patients had syphilis and HTLV infection.

In addition to being associated with CD8 T cell activation, syphilis infection also showed a significant association with CD4 T cell activation in a multivariable linear regression model (including all variables with univariable $P < 0.10$ shown in Table 4). A positive syphilis test result was associated with a 12.6% (95% CI = 1.4–23.8%, $P = 0.03$) increase in CD4 T cell activation (Table 4). An HIV RNA ≥ 5 $\log_{10}$ copies/mL was also independently associated with higher CD4 T cell activation, but having CD4 cell counts ≥ 200 cells/μL was associated with less CD4 T cell activation (Table 4).

Association of co-infections with levels of pre-cART naive T cells. Potential associations between co-infections and naive T cells were assessed. At pre-cART, the median percentage of naive CD4 T cells expressing CD62L and CD45RA was 7.5% (IQR = 2.4–17.5%) (Table 1). Univariable linear regression showed that KSHV seropositivity and to a lesser extent HTLV-I/II seropositivity and age were associated with a lower percentage of naive CD4 T cells (Table 5). Conversely, HBV infection was associated with a higher percentage of naive CD4 T cells (6.9%) although the association did not reach statistical significance. Multivariable linear regression (including all variables in Table 5 with a univariable $P <$ 0.10 and CD4 cell counts) showed that age, KSHV, and HTLV-I/II were independently associated with a decrease in percentage of naive CD4 T cells (Table 5). Four persons had KSHV and HTLV-I/II co-infections. None of the co-infections were associated with changes in levels of naive CD8 T cells.

Multivariable linear regression was performed to assess associations of KSHV, HTLV-I/II, and HBV infections with percentage of CD45RO-expressing CD4 T cells, which are considered to be a mature phenotype (model including age, HBV, HTLV-I/II, KSHV, and CD4 counts). HTLV-I/II, KSHV, and age were associated with an increase in percentage of CD45RO-expressing CD4 T cells (HTLV-I/II, $P = 0.033$; KSHV, $P = 0.065$; and age, $P = 0.076$, respectively).

DISCUSSION

The findings of this study suggest that syphilis infection and to a lesser extent HTLV seropositivity are associated with increased levels of pre-cART CD8 T cell activation and that

Table 3
Associations between pre-cART co-infections and percentage of activated CD8 T cells (CD8+ CD38+ HLA-DR+), by univariate and multivariate linear regression (n = 130), Mozambique*

Factor	Crude difference†	95% CI	P	Adjusted difference†	95% CI	P
Pre-cART baseline factors						
CD4 cells	–4.39	–10.28 to 1.49	0.142			
HIV RNA	23.23	12.48–33.99	< 0.0001	25.11	14.53–35.68	< 0.0001
WHO status	3.39	–2.43 to 9.20	0.251			
BMI	5.94	0.29–11.58	0.039			
Co-infections (+ vs. –)						
HBV	–2.16	–9.68 to 5.35	0.570			
HCV	3.01	–7.80 to 13.82	0.583			
KSHV	1.25	–3.90 to 6.41	0.631			
HTLV-I/II	7.80	–0.48 to 16.08	0.065	7.08	–0.57 to 14.73	0.070
Syphilis	7.61	–1.28 to 16.49	0.093	11.20	2.93–19.48	0.008
Pf malaria	–1.12	–12.62 to 10.37	0.847			
Tuberculosis	4.71	–0.89 to 10.30	0.098			

*Values are CD4 cell counts ≥ 200 vs. < 200 cells/mL; HIV RNA ≥ 5 $\log_{10}$ vs. < 5 $\log_{10}$ copies/mL; BMI ≤ 18.5 vs. > 18.5; WHO status III/IV vs. I/II. cART = combined antiretroviral therapy; HIV = human immunodeficiency virus; WHO = World Health Organization; BMI = body mass index; HBV = hepatitis B virus; HCV = hepatitis C vírus, KSHV = Kaposi sarcoma-associated herpesvirus; HTLV = Human T cell lymphotropic virus; Pf = *Plasmodium falciparum*.
†The difference in percent activated CD8 T cells refers to the regression coefficient in linear regression analysis. Adjusted difference refers to the multivariable model including those variables with univariate $P < 0.10$ entered into a forward stepwise model. Final adjustment includes the variables appearing in the multivariate model (HIV RNA, HTLV-I/II, syphilis).

TABLE 4

Associations between pre-cART co-infections and percentage of activated CD4 T cells (CD4+ CD38+ HLA-DR+), by univariate and multivariate linear regression (n = 130), Mozambique*

Factor	Crude difference†	95% CI	*P*	Adjusted difference†	95% CI	*P*
Pre-cART baseline factors						
CD4 cells	–13.27	–20.77 to –5.77	0.001	–11.06	–18.53 to –3.58	0.004
HIV RNA	19.71	4.92–34.49	0.009	18.51	3.90–33.13	0.013
WHO status	1.17	–6.55 to 8.87	0.767			
BMI	0.54	–7.12 to 8.19	0.890			
Co-infections (+ vs. –)						
HBV	–1.35	–11.29 to 8.60	0.789			
HCV	–0.76	–15.06 to 13.55	0.917			
KSHV	1.43	–5.38 to 8.25	0.678			
HTLV-I/II	4.73	–6.32 to 15.79	0.399			
Syphilis	11.30	–0.41 to 23.01	0.058	12.60	1.37– 23.84	0.028
Pf malaria	0.20	–14.97 to 15.38	0.979			
Tuberculosis	1.41	–6.02 to 8.90	0.704			

*Values are CD4 cell counts ≥ 200 vs. < 200 cells/mL; HIV RNA ≥ 5 $\log_{10}$ vs. < 5 $\log_{10}$ copies/mL; BMI ≤ 18.5 vs. > 18.5; WHO status III/IV vs. I/II. cART = combined antiretroviral therapy; HIV = human immunodeficiency virus; WHO = World Health Organization; BMI = body mass index; HBV = hepatitis B virus; HCV = hepatitis C vírus, KSHV = Kaposi sarcoma-associated herpesvirus; HTLV = human T cell lymphotropic virus; Pf = *Plasmodium falciparum*.

†The difference in percent activated CD8 T cells refers to the regression coefficient in linear regression analysis. Adjusted difference refers to the multivariable model including those variables with univariate $P < 0.10$ entered into a forward stepwise model. Final adjustment includes the variables appearing in the multivariate model (HIV RNA, HTLV-I/II, syphilis).

syphilis is also associated with increased CD4 T cell activation. Additionally, the results show an independent association of KSHV and HTLV infection with lower levels of pre-cART naive CD4 T ells.

We report the prevalence of some key co-infections among a group of HIV-infected adults initiating cART in Mozambique. Tuberculosis and KSHV are common in sub-Saharan African HIV- patients and their epidemiology has been amply described. Syphilis is also common and representative of sexually transmitted infections frequently associated with HIV infection. Some of the other infections assessed in this study such as hepatitis B and C have been studied less in sub-Saharan Africa but were present in 13.6% and 6.1%, respectively, of the HIV-infected patients initiating cART. The prevalence of HTLV-I/II in HIV-infected adults was 10.6% in this semi-rural area of Mozambique. This value is slightly higher than the 4.1% prevalence of HTLV-I in HIV co-infected patients in Maputo, Mozambique.[9] Although the serologic testing used in the current study did not distinguish between HTLV-I and HTLV-II, worldwide distribution strongly suggests that most infections in this region of southern Africa are HTLV-I,[10] and studies in Mozambique have not found HTLV-II.[11] Apart from tuberculosis, the relation of these co-infections to HIV pathogenesis and progression is largely unknown.

One of the hallmark features of HIV pathogenesis is chronic immune activation. Markers of CD8 T cell activation are the strongest correlate of HIV/AIDS disease progression in the absence of treatment.[2,12,13] Another T cell population that is severely impaired by HIV infection is the pool of naive T cells involved in mounting responses to novel antigens. HIV infection preferentially infects central memory CD4 T cells, and CD4 naive T cells are not considered to be depleted by direct infection.[14] We found that co-infections such as HTLV may affect CD8 T cell activation and naive CD4 T cell depletion, whereas other infections such as syphilis and KSHV may only affect either T cell activation or naive T cell depletion, respectively. Those co-infections that depleted naive T cells (HTLV and KSHV) did so independently of age. This finding is of importance because aging is known to lead to thymic involution and decreases in naive T cell output.

TABLE 5

Associations between pre-cART co-infections and percentage of CD4 naive T cells (CD4+ CD45RA+ CD62L+), by univariate and multivariate linear regression (n = 130), Mozambique*

Factor	Crude difference†	95% CI	*P*	Adjusted difference†	95% CI	*P*
Pre-cART baseline factors*						
CD4 cells	2.33	–2.96 to 7.62	0.386			
HIV RNA	7.24	–2.95 to 17.42	0.160			
WHO status	2.97	–2.25 to 8.20	0.262			
BMI	2.15	–3.02 to 7.33	0.412			
Age	–4.36	–8.95 to 0.24	0.063	–4.52	–9.06 to –0.01	0.050
Co-infections (+ vs. –)						
HBV	6.85	0.24–13.45	0.042	5.78	–0.74 to 12.30	0.082
HCV	–3.86	–13.50 to 5.77	0.429			
KSHV	–4.96	–9.49 to –0.43	0.032	–6.84	–11.33 to –2.35	0.003
HTLV-I/II	–7.08	–14.46 to 0.31	0.060	–7.82	–14.97 to –0.67	0.032
Syphilis	–1.26	–9.27 to 6.75	0.756			
Pf malaria	6.47	–3.80 to 16.74	0.215			
Tuberculosis	1.10	–3.98 to 6.18	0.670			

*Values are CD4 cell counts ≥ 200 vs. < 200 cells/mL; HIV RNA ≥ 5 $\log_{10}$ vs. < 5 $\log_{10}$ copies/mL; BMI ≤ 18.5 vs. > 18.5; WHO status III/IV vs. I/II. cART = combined antiretroviral therapy; HIV = human immunodeficiency virus; WHO = World Health Organization; BMI = body mass index; HBV = hepatitis B virus; HCV = hepatitis C vírus, KSHV = Kaposi sarcoma-associated herpesvirus; HTLV = Human T cell lymphotropic virus; Pf = *Plasmodium falciparum*.

†The difference in percent activated CD8 T cells refers to the regression coefficient in linear regression analysis. Adjusted difference refers to the multivariable model including those variables with univariate $P < 0.10$ entered into a forward stepwise model. Final adjustment includes the variables appearing in the multivariate model (HIV RNA, HTLV-I/II, syphilis).

Our study suggested that syphilis and HTLV increased T cell activation independently of HIV RNA levels, although another study has suggested that syphilis increases HIV RNA levels.[15] Because syphilis infection activates the innate immune system, including macrophages and dendritic cells, this could lead to increased activation of T cells independently of HIV RNA. Our results did not show a difference in HIV RNA levels between patients with or without co-infection with syphilis. Assessment of activation after syphilis treatment would be useful to confirm a causal relationship between syphilis and T cell activation. However, this was not possible in our study.

It has been hypothesized that immune activation may be related to naive T cell depletion. In our study, HTLV was associated with CD4 T cell activation, with a decrease in naive CD4 T cells and with an increase in CD45RO-expressing memory CD4 T cells. Although CD45RO is a non-specific marker of mature phenotype T cells, these results suggest that the dynamics of these T cell populations are all affected by HTLV infection. HTLV-I shares a similar tropism with HIV and has indeed been suggested to accelerate progression of HIV.[16] However, syphilis did not follow this pattern because it only affected the activated T cell pool. KSHV on the other hand decreased naive CD4 T cells without affecting T cell activation. Because relationships between T cell activation, naive T cell depletion, and HIV progression are complex, co-infections affecting only T cell activation may be acting by more indirect mechanisms.

There have been reports of tuberculosis, helminth infections, and HCV associated with increased T cell immune activation and/or faster HIV progression.[17] In the case of HCV, only HCV viremic patients may show increased T cell activation.[18,19] The current study included patients in advanced stages of HIV infection and did not assess HCV viremia. In the case of tuberculosis, our results suggested that tuberculosis could be associated with higher T cell activation. However, after entry into multivariable analysis, tuberculosis lost its significant association with increased T cell activation. It is thought that increased T cell activation caused by tuberculosis may only be detectable in patients with less advanced HIV/AIDS than in our patients. The proportion of those patients co-infected with HTLV was equal in patients with and without tuberculosis.

A limitation of the study was that of measuring potential synergistic effects of multiple infections. The current findings using multivariable regression suggest that the infections observed to affect T cell subsets do so independently of each other. However, we cannot exclude that concurrent co-infections observed, such as tuberculosis and HTLV or syphilis and KSHV, have different affects than would single co-infections. Larger studies would be required to assess potential interactions. This suggestion is particularly relevant in areas of sub-Saharan Africa, which have a high prevalence of HIV and disease burden.

Another limitation is the analysis of co-infections affecting naive CD4 T cells and memory CD4 T cells. These cell populations show a non-normal distribution. Thus, the results for these populations can only be interpreted qualitatively as a positive or negative effect and the actual numerical amplitude cannot be inferred from our results.

The results of this study highlight the importance of screening and prompt treatment of syphilis, and raise questions as to whether HIV-positive persons with certain chronic viral co-infections should initiate cART at higher CD4 cell counts. Because there is a risk that increased immune activation could decrease the response to cART,[20] the effect of co-infections on cART response also warrants study. Inversely, the impact of cART regimens on co-infections also must be considered.

Recent World Health Organization 2010 recommendations stress that the cutoff for cART initiation in resource-poor countries should be increased to 350 CD4 cells/μL as opposed to 200 cells/μL. Initiating cART earlier is associated with improved survival.[21,22] Earlier cART initiation may be more relevant in the sub-Saharan African context, where it could counteract the potential worsening of immune activation and/or depletion of naive T cells by co-infections that are difficult or impossible to treat.[23]

Received December 20, 2010. Accepted for publication May 2, 2011.

Acknowledgments: We thank the patients for participating in the study; the voluntary counseling and testing center and HIV day hospital staff and data management staff at the Centro de Investigação em Saúde de Manhiça for their dedication and assistance; Deolinda Nhassone and Joaquim Gascon for facilitating the clinical studies; Jose Machado Almeida for laboratory supervision; Elsa Banze, Recifal Iaço, Atanasio Xirindza, and Nelito Ernesto José for their participation in patient logistics and sample processing; and Edgar Ayala for critically reading the manuscript.

Financial support: This study was supported by the Fundació "la Caixa" and the Agència Catalana de Cooperació al Desenvolupament. The Centro de Investigação em Saúde de Manhiça receives core funding from the Spanish Agency for International Cooperation and Development. The voluntary counseling and testing center is supported by the Agència Catalana de Cooperació al Desenvolupament. Denise Naniche was supported by a grant from the Spanish Ministry of Education and Science (Ramon y Cajal).

Disclosure: None of the authors has any conflicts of interest.

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