MAJOR ARTICLE

Concordant HIV Infection and Visceral Leishmaniasis in Ethiopia: The Influence of Antiretroviral Treatment and Other Factors on Outcome

Rachel ter Horst,[1] Simon M. Collin,[2] Koert Ritmeijer,[1] Adey Bogale,[1] and Robert N. Davidson[3]

[1]Médecins Sans Frontières, Amsterdam, The Netherlands; and [2]Department of Social Medicine, University of Bristol, Bristol, and [3]Department of Infection and Tropical Medicine, Northwick Park Hospital, Middlesex, United Kingdom

Background. Coinfection with human immunodeficiency virus (HIV) and *Leishmania donovani* visceral leishmaniasis (VL) in Africa is an emerging, poorly understood disease.

Methods. We evaluated 356 consecutive patients coinfected with HIV and VL treated in Humera, northwest Ethiopia, from February 2003 to October 2006, for risk factors for VL relapse and death and the effect of antiretroviral therapy (ART).

Results. During 2928 patient-months of follow-up, 256 VL episodes and 39 deaths occurred. Among 195 patients receiving ART, 31.3% had ⩾1 VL episode, and 14.4% died. Among 161 patients who did not receive ART, 26.1% had ⩾1 VL episodes, and 6.8% died. A total of 54 patients who received ART and 58 patients who did not receive ART had ⩾1 VL relapse. VL relapse among patients receiving ART was associated with a baseline CD4 cell count <100 cells/μL (hazard ratio [HR], 2.50; 95% confidence interval [CI], 1.21–5.15) and ⩾2 previous VL episodes (HR, 3.74; 95% CI, 1.40–10.02). Failure to clear parasites after VL treatment was usually followed by symptomatic VL relapse. Patients who relapsed showed poor CD4 cell count recovery while receiving ART. ART was partially protective against VL relapse (HR, 0.46; 95% CI, 0.26–0.82). However, 28% of first VL relapses while receiving ART occurred despite a CD4 cell count >200 cells/μL; in 5% of VL relapses, the CD4 cell count had been >200 cells/μL for >6 months. Factors associated with all-cause mortality among patients receiving ART were baseline CD4 cell count <100 cells/μL (HR, 3.20; 95% CI, 1.30–7.87) and VL episodes during follow-up (HR for 1 episode, 4.97 [95% CI, 2.09–11.86]; HR for >2 episodes, 3.22 [95% CI, 1.01–10.23]).

Conclusions. Concordant HIV infection and VL is a major, acquired immunodeficiency syndrome–defining illness with high relapse and mortality rates; ART reduces relapses; and secondary antileishmanial prophylaxis may benefit patients at risk of relapse.

Visceral leishmaniasis (VL; also known as "kala-azar") is a systemic parasitic disease caused by the *Leishmania donovani* species complex. An estimated 500,000 new cases of VL occur annually [1]. In east Africa and south Asia, the parasite is *L. donovani,* and the predominant mode of transmission, via sandflies, is anthroponotic. Humans with VL or post–kala-azar dermal leishmaniasis provide the major reservoir for transmission; thus, incomplete or irregular treatment of VL leads to drug pressure and parasite resistance. In the Mediterranean region and Latin America, a less-pathogenic zoonotic parasite (*Leishmania infantum,* known as *Leishmania chagasi* in Latin America) exists, causing infection in dogs and sporadic VL cases in humans [2].

VL is typically fatal if untreated. In immunocompetent individuals, effective drug treatment reduces *Leishmania* amastigotes to a level undetectable in aspirates. An effective life-long cellular immune response normally develops, and residual parasites are suppressed unless immunodeficiency is present [3]. HIV infection can lead to reactivation of latent *Leishmania* infection or to symptomatic VL at initial infection; in Europe, the risk of developing VL is 100–1000 times greater for HIV-infected individuals than for HIV-uninfected individuals [4, 5]. VL accelerates HIV replication and disease progression, mainly by chronic immune stimulation [6]. The prevalence of patients with

Received 30 November 2007; accepted 23 January 2008; electronically published 17 April 2008.

Reprints and correspondence: Simon M. Collin, Dept. of Social Medicine, Canynge Hall, Whiteladies Rd., Bristol, BS8 2PR, United Kingdom (simon.collin@bristol.ac.uk).

Clinical Infectious Diseases 2008; 46:1702–9

1058-4838/2008/4611-0009$15.00
DOI: 10.1086/587899

concordant HIV infection and VL (hereafter, "HIV-VL coinfection") in Europe has fallen sharply since 1996, when antiretroviral treatment (ART) became standard. In India and particularly in Africa, HIV-VL coinfection is emerging. The AIDS pandemic has expanded to rural areas where VL is endemic, with cases of HIV-VL coinfection reported in 35 countries [7, 8], among which Ethiopia carries the greatest burden. Most studies of HIV-VL coinfection originate in southern Europe and are unlikely to be representative of the situation in East Africa, where the parasite *(L. donovani)* is more pathogenic and its comorbidities are different [9–17]. The Brazilian experience of HIV-VL coinfection resembles that of Europe: high parasite load, parasite dissemination to unusual sites, lower initial and final cure rates, greater susceptibility to drug toxicity, increased risk of drug resistance, and higher rates of death and relapse [18]. Information on HIV-VL coinfection in Africa is scarce; known problems are similar to those mentioned above and include a greater rate of post–kala-azar dermal leishmaniasis [19–23]. In Europe, ART has reduced the incidence of VL [15], has prolonged the intervals between relapses and reduced relapse rates [11, 14, 24], and has improved survival [3].

Médecins Sans Frontières (MSF) has been giving treatment to patients with VL in the Humera area of western Tigray, the main region in Ethiopia where VL is endemic, since 1997. Our VL-affected patient population consists mainly of male (>90%) migrant farm laborers who live and work unprotected from the VL vector for several months among the cotton, sesame, and sorghum fields. Having introduced ART in January 2004, we wished to assess its impact and to identify determinants of VL relapse and survival, to optimize patient care.

METHODS

Study design and population. HIV-VL–coinfected patients were recruited from an integrated HIV and VL treatment program at the MSF clinic in Humera, northwest Ethiopia, from February 2003 through October 2006. The study group comprised patients who received a diagnosis of HIV-VL coinfection at admission and patients who received a diagnosis of HIV/AIDS at admission who had a history of VL and/or who developed VL during follow-up. All investigations and treatments for all patients were free of charge.

HIV testing and treatment. All patients with VL were routinely offered voluntary HIV counseling and testing, generally at 1–2 weeks after initiation of VL treatment, when the patient was well enough to participate. HIV testing was done by 2 rapid diagnostic tests in parallel (HIV-Determine [Abbott Diagnostics] and Unigold [Trinity Biotech]) by use of venous blood [25]. All positive test results were confirmed in the laboratory with a second blood sample by use of the same tests. Patients with discordant test results were asked to return after 6 weeks; if the test results were again discordant, the test result was considered negative. Patients with VL who were infected with HIV were offered follow-up care in our clinic for HIV/AIDS. We provided diagnosis and treatment of opportunistic infections, *Pneumocystis jiroveci* pneumonia prophylaxis, tuberculosis treatment, psychosocial support, shelter for homeless patients, and a feeding program, as appropriate. Since January 2004, HIV-infected patients were offered ART if they had a $CD4^+$ cell count <200 cells/μL, were well motivated for lifelong ART, and had not defaulted from the follow-up clinic. ART was also offered to all HIV-infected patients with primary VL and a $CD4^+$ cell count <350 cells/μL and to all coinfected patients with VL relapse irrespective of $CD4^+$ cell count. Generic antiretroviral drugs were used: the first-line regimen was lamivudine-stavudine-nevirapine, with alternative regimens of lamivudine-zidovudine-nevirapine for stavudine toxicity and lamivudine-stavudine-efavirenz with coadministration of rifampicin for tuberculosis. Second-line regimens were not required during the study period.

Follow-up for patients with HIV/AIDS. $CD4^+$ cell counts were determined (FacsCount flow cytometer; Becton Dickinson) at the first visit or, if the patient was receiving VL treatment, after completion of VL treatment. Follow-up $CD4^+$ cell counts were determined at 3–6 month intervals. Because of our remote location, we could not routinely evaluate HIV-1 RNA levels.

VL diagnosis and test of cure. We diagnosed VL as described elsewhere [21]. We used the World Health Organization case definition of VL [26]: a history of fever for >2 weeks (malaria excluded) in combination with wasting and either splenomegaly or lymphadenopathy. If patients met this case definition, VL was confirmed by a high titer ($\geqslant$1:6400) on *Leishmania* direct agglutination test (DAT) (Royal Tropical Institute) [27] or by the finding of *Leishmania* amastigotes in spleen or lymph node aspirates. In patients with an intermediate DAT titer (1:800–1:3200), aspiration was performed. Patients with a negative DAT titer ($\leqslant$1:400) were evaluated for alternative illnesses and were retested (by DAT and splenic aspirate) if symptoms persisted. As was found previously in this population [21], there was no difference in mean DAT titer between HIV-infected and -uninfected patients. Severely ill patients were aspirated without delay, so that a diagnosis could be made as quickly as possible. In patients with previous antileishmanial treatment, VL was confirmed by a positive aspirate result. A test of cure by spleen or lymph node aspirate was done at the end of treatment for all patients with VL relapse and for patients with primary VL if the response to the treatment was uncertain.

VL treatment regimens. Primary VL was treated with sodium stibogluconate (SSG) (Albert David, Calcutta and International Dispensary Association) at 20 mg/kg/day by intramuscular injection for 30 days. During November 2003–September 2004, 33 men with VL participated in a randomized

trial of miltefosine versus SSG [21]; miltefosine was not available outside this clinical trial period. First relapses of VL were treated with SSG for 40–60 days, depending on the results of the test of cure. Second relapses of VL were treated either with SSG for 40–60 days or with amphotericin B deoxycholate at 1 mg/kg intravenously every other day for 15–20 doses. Additional relapses were treated either with SSG or with amphotericin B; occasionally, the 2 drugs were combined. Secondary prophylaxis was not part of clinical management during the study period.

Data entry and statistical analysis. Demographic and clinical data were collected for each patient from the patient's HIV/AIDS and VL medical records and were entered singly into an Excel (Microsoft) spreadsheet. Statistical analyses were performed using Stata (StataCorp). Hazard ratios for risk factors for our outcomes of interest were estimated using Cox proportional hazards regression. Factors for which there was an association were then included in multivariate Cox regression models to estimate the impact of ART on outcomes. We separated patients according to whether they received ART. Follow-up began at initiation of ART or, for patients who did not receive ART, when the patient registered at the treatment center. Data on patients who defaulted or who transferred out of the program were censored.

The primary outcomes were (1) VL relapse and (2) death (all causes) during follow-up. We based survival analysis of VL relapse on the interval from the start of follow-up to the first VL relapse during follow-up, irrespective of previous relapses. Survival analysis of death was based on the interval from the start of follow-up to death. Secondary outcomes were (1) multiple VL relapses and (2) first episode of VL during follow-up. The time to multiple VL relapses was taken as the interval from the start of follow-up to the first of the patient's relapses during follow-up. Time to the first episode of VL was the interval from the start of follow-up to the patient's first VL episode during follow-up. As a final outcome, we wished to establish whether an immune reconstitution inflammatory syndrome (IRIS) might have occurred. We defined IRIS as the occurrence of the first episode of VL after initiation of ART, with a negative result of a serological and/or parasitological test just before ART initiation and a positive result of a serological and/or parasitological test after ART initiation. We also noted the occurrence of tuberculosis in relation to the start of follow-up.

Ethics approval. Data were collected as part of routine patient care; no additional investigations were performed other than those indicated for medical management. Ethics approval was obtained from the MSF Ethical Review Board. Approval was obtained from the Tigray regional health authorities for publication of the data.

RESULTS

During the study period, 2782 patients with VL were given treatment. Uptake of counseling and testing among patients with VL was 70%, of whom 31% were infected with HIV. Of these HIV-infected patients, 195 commenced ART during the study period. Of the remaining HIV-infected patients (who did not receive ART), 161 had sufficient data for inclusion. Table 1 shows the demographic and clinical characteristics of the patients with HIV-VL coinfection.

Factors associated with VL relapse. Baseline $CD4^+$ cell count <100 cells/μL, tuberculosis coinfection, and previous VL episodes were all risk factors for VL relapse (table 2). Among patients who received ART, $CD4^+$ cell count reconstitution was blunted among those with multiple relapses of VL, compared with patients who did not have a VL relapse (figure 1).

In 18 relapses of VL (in 12 patients) the test of cure showed failure to clear parasites; of the 18 relapses, 15 were followed by further symptomatic VL relapse. One patient had "spontaneous" parasitological cure after a scantily positive test of cure (grade 1), accompanied by a good $CD4^+$ cell count gain. We sought to determine whether a threshold of $CD4^+$ cells might protect against relapse of VL. Among 43 HIV-VL–coinfected patients receiving ART who had a first VL relapse during follow-up, 39 had a $CD4^+$ cell count recorded 2–7 months before the relapse; of these 39 patients, 11 (28%) had $CD4^+$ cell counts >200 cells/μL, and 4 had $CD4^+$ cell counts >350 cells/μL. In 2 (5%) of the 39 patients, the $CD4^+$ cell count had been sustained at >200 cells/μL for >6 months preceding the relapse. There were insufficient patients to allow meaningful analysis of intervals between events (e.g., from the first VL episode to the first relapse, between successive relapses, or from relapse to death) or of factors associated with these intervals.

Factors associated with death (all causes). Baseline or nadir $CD4^+$ cell count <100 cells/μL and >2 VL episodes were associated with higher mortality among patients who received ART and patients who did not receive ART; among the patients who received ART, primary VL was also associated with higher mortality (table 3).

Effect of ART on outcomes during follow-up. Table 4 shows the adjusted hazard ratios for the effects of ART, baseline $CD4^+$ cell count <100 cells/μL, and previous episodes of VL on the primary and secondary outcomes. ART had a protective effect on single or multiple relapses of VL, but there was no evidence of an effect of ART on survival.

IRIS or primary acquisition of VL. Thirteen patients developed their first lifetime VL episode after initiation of ART. All had been evaluated for VL before initiation of ART, and 11 had documented negative DAT results (titer, <1:400) and/or negative splenic aspirate test results; an additional 2 had intermediate serological test results (DAT titer, $\geq$1:400 but <1:

Table 1. Characteristics of patients who received and patients who did not receive antiretroviral treatment (ART).

Characteristic	Patients without ART (n = 161)	Patients with ART (n = 195)	P[a]
Age, years			
Mean (range)	31.3 (20–50)	33.5 (18–60)	.01[b]
Male sex	98.2	90.5	.003
Baseline CD4$^+$ cell count,[c] cells/μL			
⩾300	37.3	2.4	<.001
200–299	18.6	12.1	
100–199	14.4	38.6	
0–99	29.7	47.0	
Tuberculosis			
None	64.2	41.4	<.001
Before study	25.3	39.8	
After entry into study	8.0	16.2	
Before and after entry into study	2.5	2.6	
No. of VL episodes before entry into study			
0	47.5	26.3	<.001
1	37.7	44.9	
2	13.0	22.2	
⩾3	1.9	6.7	
VL episode at entry into study	61.1	27.8	<.001
No. of VL episodes after entry into study			
0	73.5	69.1	<.001
1	21.6	24.7	
2	3.7	2.1	
⩾3	1.2	4.1	
Follow-up status			
Died	6.8	14.4	<.001
Defaulted	63.0	15.0	
Transferred	11.7	4.1	
Active	18.5	66.5	
Follow-up time, months			
Total	1003	1929	
Mean	6.5	10.0	<.001[b]
Median (range)	3.1 (0–36.5)	7.1 (0.5–33.5)	<.001[d]

NOTE. Data are % of patients, unless otherwise indicated.

[a] By χ^2 test, unless otherwise indicated.

[b] By Student's t test, with 354 df.

[c] Baseline data for CD4$^+$ cell count were missing for 28 patients who did receive ART and 44 patients who did not receive ART.

[d] By Kruskal-Wallis test, with 1 df.

1600) and negative aspirate results. All had documented high DAT titers and/or positive parasitological test results after initiation of ART. The mean time at diagnosis of VL was 3 months after initiation of ART (range, 12 days to 9 months). Their mean CD4$^+$ cell count at ART initiation was 130 cells/μL (range, 16–210 cells/μL). Their clinical picture was of classic VL, and standard VL treatment was initiated in all cases. Significant CD4$^+$ cell count reconstitution after the primary VL episode was documented in 8 patients: CD4$^+$ cell counts >50 cells/μL at 6 months in 7 patients and a CD4$^+$ cell count of 37 cells/μL at 6 months and 148 cells/μL at 12 months in 1 patient. VL treatment outcomes were good among this group of patients; none died during treatment, 8 of 8 patients had a negative test of cure, and the remaining 5 patients showed a good clinical response (test of cure was not performed). Two patients subsequently died (after 3 months and 8 months) due to non-VL causes. Relapse occurred in only 1 of the 12 patients with sufficient follow-up time (⩾8 months) after the primary VL

episode. Concomitant herpes zoster IRIS occurred in 2 patients, and tuberculosis IRIS occurred in 2 other patients.

DISCUSSION

Our study represents the largest single-center cohort of HIV-VL–coinfected patients reported to date, larger than all previously reported case series of HIV–*L. donovani* coinfection combined. Ethiopian patients with HIV-VL coinfection, similar to European patients coinfected with HIV and *L. infantum*, have a poor prognosis; many patients have chronic relapsing VL and early death. Factors associated with increased risk of VL relapse were baseline CD4$^+$ cell count <100 cells/μL and a history of VL and tuberculosis. Factors associated with increased risk of death were baseline CD4$^+$ cell count <100 cells/μL and VL during follow-up. ART reduced the risk of relapse by ~50%. Our data did not give meaningful information on the effect of ART on the intervals between VL relapses, reported as longer by some investigators in Europe [28].

Despite the large size of our study, we did not demonstrate an impact of ART on survival, probably because our study suffered from bias due to the substantial and disproportionate

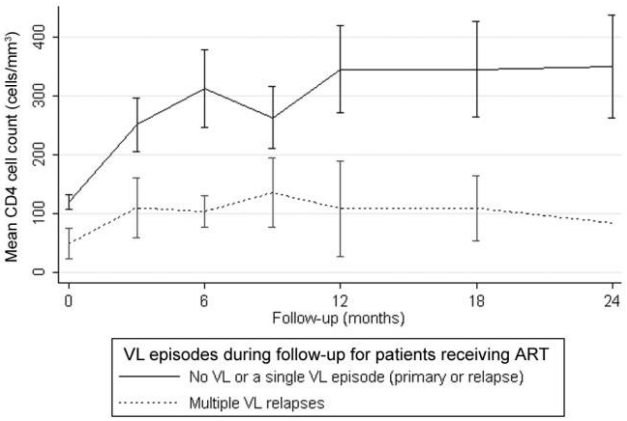

Figure 1. Trends in CD4$^+$ cell counts for patients receiving antiretroviral treatment (ART), including patients with multiple relapses of visceral leishmaniasis (VL) and patients with no VL episode or a single VL episode (primary or relapse). Bars represent 95% CIs.

Table 2. Risk factors for visceral leishmaniasis (VL) relapse during follow-up of patients who received and patients who did not receive antiretroviral treatment (ART).

	Unadjusted hazard ratio (95% CI)	
Risk factor	Patients without ART	Patients with ART
Age	0.99 (0.94–1.04)	1.01 (0.97–1.04)
Sex (male vs. female)	…[a]	1.72 (0.53–5.57)
Tuberculosis		
None	1.00	1.00
Before follow-up	1.34 (0.63–2.86)	1.28 (0.64–2.53)
After follow-up	0.56 (0.13–2.40)	1.18 (0.48–2.90)
Before and after follow-up	6.31 (1.42–28.01)	3.05 (0.88–10.56)
Baseline CD4$^+$ cell count, cells/μL		
⩾100	1.00	1.00
<100	2.05 (0.80–5.26)	2.50 (1.21–5.15)
Nadir CD4$^+$ cell count, cells/μL		
⩾100	1.00	1.00
<100	1.86 (0.72–4.78)	2.30 (1.24–4.26)
No. of VL episodes before follow-up		
0	1.00	1.00
1	4.00 (1.64–9.77)	2.11 (0.79–5.66)
⩾2	2.25 (0.68–7.41)	3.74 (1.40–10.02)
Interval from last VL episode to start of follow-up, months		
<3	1.00	1.00
3–9	1.51 (0.40–5.68)	0.67 (0.31–1.43)
⩾9	0.97 (0.18–5.27)	0.51 (0.20–1.27)
VL episode at start of follow-up	0.66 (0.34–1.30)	0.90 (0.46–1.75)

NOTE. VL, visceral leishmaniasis.

[a] None of 36 events involved female patients.

Table 3. Risk factors for death (all causes) during follow-up of patients who received and patients who did not receive antiretroviral treatment (ART).

Risk factor	Unadjusted hazard ratio (95% CI)	
	Patients without ART	Patients with ART
Age	1.13 (1.03–1.24)	0.99 (0.94–1.04)
Sex (male vs. female)	…[a]	1.05 (0.32–3.51)
Tuberculosis at any time	2.17 (0.54–8.72)	1.12 (0.52–2.41)
Baseline CD4$^+$ cell count, cells/μL		
$\geqslant$100	1.00	1.00
<100	13.82 (1.42–133.97)	3.20 (1.30–7.87)
Nadir CD4$^+$ cell count, cells/μL		
$\geqslant$100	1.00	1.00
<100	12.48 (1.29–121.01)	3.77 (1.61–8.84)
VL episode at start of follow-up	1.30 (0.31–5.45)	1.72 (0.80–3.71)
No. of VL episodes during follow-up		
0	1.00	1.00
1	9.57 (1.07–85.90)	4.97 (2.09–11.86)
$\geqslant$2	16.71 (1.71–163.44)	3.22 (1.01–10.23)
Primary VL during follow-up	…[b]	5.75 (2.25–14.71)
Single relapse during follow-up	3.48 (0.86–14.00)	2.19 (0.86–5.55)
Multiple relapse during follow-up	5.88 (1.37–25.31)	1.71 (0.50–5.77)

NOTE. VL, visceral leishmaniasis.

[a] None of 8 deaths involved female patients.

[b] None of 8 deaths involved patients with primary VL.

number of patients who were lost to follow-up in the group that did not receive ART. We could speculate (not unreasonably) that most of these patients would have died unless they obtained treatment at the place to which they returned (presumably their place of origin), and ART might then show a protective effect. Although ART reduced the risk of VL relapse, 22% of our patients who received ART experienced $\geqslant$1 VL relapse; these patients showed generally poor CD4$^+$ cell count reconstitution despite good adherence to treatment. European studies of HIV-VL–coinfected patients have reported similarly poor CD4$^+$ cell count reconstitution but have attributed this to poor adherence to ART [24]. Thresholds for safe discontinuation of secondary prophylaxis for patients with HIV and *L. infantum* VL have been suggested as CD4$^+$ cell counts of 200 cells/μL [29] and 350 cells/μL [30]. In our study, VL relapses were sometimes seen despite high CD4$^+$ cell counts—in 28% of first relapses during follow-up, the preceding CD4$^+$ cell count was >200 cells/μL, and, in 10% of first relapses, it was >350 cells/μL. Our data suggest that a high CD4$^+$ cell count is required to prevent relapse of *L. donovani* infection. This is con-

Table 4. Effect of antiretroviral treatment (ART) and other factors on outcomes during follow-up.

Outcome	Adjusted hazard ratio[a] (95% CI)		
	ART (vs. no ART)	Baseline CD4$^+$ cell count <100 cells/μL (vs. $\geqslant$100 cells/μL)	$\geqslant$2 Previous VL episodes (vs. 0 or 1 episode)
Primary VL	0.77 (0.27–2.21)	0.91 (0.35–2.37)	NA
Single relapse	0.46 (0.25–0.84)	1.60 (0.85–3.00)	NA
Primary VL or single relapse	0.55 (0.33–0.92)	1.28 (0.75–2.19)	NA
Multiple relapse	0.52 (0.12–2.18)	7.72 (1.54–38.77)	5.17 (1.41–18.99)
Single or multiple relapse	0.46 (0.26–0.82)	2.09 (1.19–3.67)	1.34 (0.70–2.55)
Any VL (primary, single, or multiple relapse)	0.56 (0.34–0.91)	1.66 (1.02–2.70)	NA
Death	1.69 (0.56–5.12)	3.81 (1.55–9.36)	1.87 (0.66–5.29)

NOTE. NA, not applicable; VL, visceral leishmaniasis.

[a] Adjusted for each of the other factors shown plus age and tuberculosis.

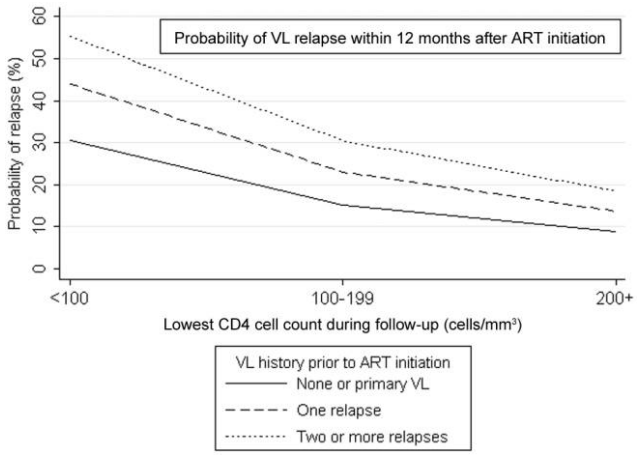

Figure 2. Relapse probability curves for patients receiving antiretroviral treatment (ART), which show the influence of the nadir $CD4^+$ cell count and previous episodes of visceral leishmaniasis (VL) on the probability of VL relapse within 1 year.

sistent with the observation that *L. donovani* is a virulent, anthroponotic species, whereas *L. infantum* has little virulence for immunocompetent humans beyond early childhood. We consider that the lack of clearance of *Leishmania* parasites in our patients holds the patient in a state of immune suppression; hence, they are unable to regain cellular immunity (despite good antiviral efficacy of ART). This leads to recrudescence of parasites and relapses.

We reported 18 instances of failure to clear parasites, despite prolonged treatment with SSG and amphotericin B, suggesting that development of secondary resistance in African *L. donovani* can occur with relative ease. VL treatment failure was almost always followed by further symptomatic VL episodes, demonstrating the importance of achieving an initial parasitological cure. In HIV-VL–coinfected patients, the most likely way of achieving this is by combination VL therapy. In analyses beyond the scope of this study, we found that patients receiving ART who have a history of VL have a mortality rate twice as high as that of patients receiving ART from the same treatment center who do not have VL ($n = 2111$), controlling for age and baseline $CD4^+$ cell count (HR, 2.02; 95% CI, 1.21–3.37).

We consider that there is need for long-term, maintenance antileishmanial treatment for individuals at high risk of VL relapse once a negative test of cure is achieved. Such patients might be identified by the factors we found—for example, a patient with baseline $CD4^+$ cell count <100 cells/μL and $\geqslant$3 previous VL episodes would have a 56% chance of relapsing within 1 year (figure 2). Such patients might then be eligible for secondary prophylaxis (e.g., with pentamidine). It is not yet clear after what VL disease–free interval or at what $CD4^+$ cell count level patients are free of significant risk of VL relapse, but we hope to address these questions in the future. Limited evidence is available for secondary prophylaxis for patients with HIV and *L. infantum* VL. In a randomized trial of 17 patients, 50% of the group that received secondary prophylaxis (3 mg of amphotericin B lipid complex per kg every 21 days) remained relapse free at 1 year, compared with 22% of the group without prophylaxis [31]. A retrospective study of 46 patients reported probabilities of patients remaining relapse-free at 1 year of 9% without secondary prophylaxis, 21% with allopurinol, and 93% with pentavalent antimony [32]. Six patients in Spain received pentamidine at 4 mg/kg every 2 or 4 weeks (3 patients in each group) as secondary prophylaxis. No relapses were observed during a mean follow-up of 8 months (range, 3–12 months) [33]. Pentamidine might be a useful drug for secondary prophylaxis for VL; it has been used safely as secondary prophylaxis against *Pneumocystis carinii* pneumonia [34].

We report an important finding of 13 patients that had a first VL episode after initiation of ART; these patients could be manifesting VL IRIS. Before this study, 6 possible cases of VL IRIS have been reported, all from the Mediterranean region [35].

Our findings lead us to make 5 recommendations for the care of HIV-VL–coinfected patients in Africa. First, given the clear benefits of ART, HIV testing should be widely applied using an opt-out strategy. Second, in Ethiopia but not in other countries, VL has been classified as an AIDS-defining opportunistic infection (in Ethiopian national HIV guidelines). We consider that VL should be an AIDS-defining illness in all countries and a valid entry point to ART irrespective of $CD4^+$ cell count. This may help to reduce VL relapse rates. Third, secondary VL prophylaxis is necessary for individuals at high risk of relapse despite ART. Fourth, parasitological clearance seems to be a crucial end point to VL treatment. Finally, high rates of treatment failure, possibly leading to development of resistant parasites, indicate that combination therapy, instead of monotherapy, should be used to treat HIV-VL coinfection. In selecting a combination regimen, it should be kept in mind that SSG monotherapy has an unacceptable mortality rate in HIV-VL–coinfected patients [21] and, theoretically, may induce an increase in HIV-1 virus replication [36]. We look forward to the licensing in Ethiopia and in other countries of safer drugs for patients with HIV-VL coinfection (e.g., miltefosine, paromomycin, and liposomal amphotericin B), preferably to be used in combination.

Acknowledgments

We acknowledge and thank the Tigray Regional Bureau of Health and Kahsay Abera Hospital Humera.

Financial support. Médecins sans Frontières-Holland.

Potential conflicts of interest. All authors: no conflicts.

References

1. Desjeux P. The increase in risk factors for leishmaniasis worldwide. Trans R Soc Trop Med Hyg **2001**; 95:239–43.
2. Guerin PJ, Olliaro P, Sundar S, et al. Visceral leishmaniasis: current status of control, diagnosis, and treatment, and a proposed research and development agenda. Lancet Infect Dis **2002**; 2:494–501.
3. Pintado V, Martin-Rabadan P, Rivera ML, Moreno S, Bouza E. Visceral leishmaniasis in human immunodeficiency virus (HIV)-infected and non-HIV-infected patients: a comparative study. Medicine (Baltimore) **2001**; 80:54–73.
4. Kubar J, Marty P, Lelievre A, et al. Visceral leishmaniosis in HIV-positive patients: primary infection, reactivation and latent infection: impact of the CD4+ T-lymphocyte counts. AIDS **1998**; 12:2147–53.
5. Paredes R, Munoz J, Diaz I, Domingo P, Gurgui M, Clotet B. Leishmaniasis in HIV infection. J Postgrad Med **2003**; 49:39–49.
6. Wolday D, Akuffo H, Demissie A, Britton S. Role of *Leishmania donovani* and its lipophosphoglycan in $CD4^+$ T-cell activation-induced human immunodeficiency virus replication. Infect Immun **1999**; 67: 5258–64.
7. Cruz I, Nieto J, Moreno J, Canavate C, Desjeux P, Alvar J. Leishmania/HIV co-infections in the second decade. Indian J Med Res **2006**; 123: 357–88.
8. Desjeux P, Alvar J. Leishmania/HIV co-infections: epidemiology in Europe. Ann Trop Med Parasitol **2003**; 97(Suppl 1):3–15.
9. Alvar J, Canavate C, Gutierrez-Solar B, et al. Leishmania and human immunodeficiency virus coinfection: the first 10 years. Clin Microbiol Rev **1997**; 10:298–319.
10. Laguna F, Lopez-Velez R, Pulido F, et al. Treatment of visceral leishmaniasis in HIV-infected patients: a randomized trial comparing meglumine antimoniate with amphotericin B. Spanish HIV-Leishmania Study Group. AIDS **1999**;13:1063–9.
11. Pasquau F, Ena J, Sanchez R, et al. Leishmaniasis as an opportunistic infection in HIV-infected patients: determinants of relapse and mortality in a collaborative study of 228 episodes in a Mediterreanean region. Eur J Clin Microbiol Infect Dis **2005**; 24:411–8.
12. Pintado V, Lopez-Velez R. HIV-associated visceral leishmaniasis. Clin Microbiol Infect **2001**; 7:291–300.
13. Russo R, Laguna F, Lopez-Velez R, et al. Visceral leishmaniasis in those infected with HIV: clinical aspects and other opportunistic infections. Ann Trop Med Parasitol **2003**; 97(Suppl 1):99–105.
14. Lopez-Velez R. The impact of highly active antiretroviral therapy (HAART) on visceral leishmaniasis in Spanish patients who are co-infected with HIV. Ann Trop Med Parasitol **2003**; 97(Suppl 1):143–7.
15. Fernández Cotarelo MJ, Abellan Martínez J, Guerra Vales JM, Martínez Sánchez P, Rodrigo Gómez de la Bárcena M, Salto Fernández E. Effect of highly active antiretroviral therapy on the incidence and clinical manifestations of visceral leishmaniasis in human immunodeficiency virus–infected patients. Clin Infect Dis **2003**; 37:973–7.
16. World Health Organization (WHO). Leishmania/HIV co-infection in southwestern Europe 1990–1998: retrospective analysis of 965 cases. Report WHO/LEISH/2000.42. Geneva: WHO, **2000**.
17. Berman J. Visceral leishmaniasis in the New World and Africa. Indian J Med Res **2006**; 123:289–94.
18. Rabello A, Orsini M, Disch J. Leishmania/HIV co-infection in Brazil: an appraisal. Ann Trop Med Parasitol **2003**; 97(Suppl 1):17–28.
19. Ritmeijer K, Veeken H, Melaku Y, et al. Ethiopian visceral leishmaniasis: generic and proprietary sodium stibogluconate are equivalent; HIV co-infected patients have a poor outcome. Trans R Soc Trop Med Hyg **2001**; 95:668–72.
20. Lyons S, Veeken H, Long J. Visceral leishmaniasis and HIV in Tigray, Ethiopia. Trop Med Int Health **2003**; 8:733–9.
21. Ritmeijer K, Dejenie A, Assefa Y, et al. A comparison of miltefosine and sodium stibogluconate for treatment of visceral leishmaniasis in an Ethiopian population with high prevalence of HIV infection. Clin Infect Dis **2006**; 43:357–64.
22. Wolday D, Berhe N, Akuffo H, Desjeux P, Britton S. Emerging *Leishmania*/HIV co-infection in Africa. Med Microbiol Immunol **2001**; 190: 65–7.
23. Haile T, Anderson SD. Visceral leishmaniasis in northern Ethiopia. East Afr Med J **2006**; 83:389–92.
24. Mira JA, Corzo JE, Rivero A, et al. Frequency of visceral leishmaniasis relapses in human immunodeficiency virus-infected patients receiving highly active antiretroviral therapy. Am J Trop Med Hyg **2004**; 70: 298–301.
25. UNAIDS/WHO Working Group on Global HIV/AIDS/STI Surveillance. Guidelines for using HIV testing technologies in surveillance. Report WHO/CDC/CSR/EDC/2001.16. Geneva: World Health Organization, **2001**.
26. World Health Organization (WHO). Manual on visceral leishmaniasis. Report WHO/Leish/96.40. Geneva: WHO, **1996**.
27. Meredith SE, Kroon NC, Sondorp E, et al. Leish-KIT, a stable direct agglutination test based on freeze-dried antigen for serodiagnosis of visceral leishmaniasis. J Clin Microbiol **1995**; 33:1742–5.
28. Casado JL, Lopez-Velez R, Pintado V, Quereda C, Antela A, Moreno S. Relapsing visceral leishmaniasis in HIV-infected patients undergoing successful protease inhibitor therapy. Eur J Clin Microbiol Infect Dis **2001**; 20:202–5.
29. Soriano V, Dona C, Rodriguez-Rosado R, Barreiro P, Gonzalez-Lahoz J. Discontinuation of secondary prophylaxis for opportunistic infections in HIV-infected patients receiving highly active antiretroviral therapy. AIDS **2000**; 14:383–6.
30. Berenguer J, Cosin J, Miralles P, Lopez JC, Padilla B. Discontinuation of secondary anti-leishmania prophylaxis in HIV-infected patients who have responded to highly active antiretroviral therapy. AIDS **2000**; 14: 2946–8.
31. Lopez-Velez R, Videla S, Marquez M, et al. Amphotericin B lipid complex versus no treatment in the secondary prophylaxis of visceral leishmaniasis in HIV-infected patients. J Antimicrob Chemother **2004**; 53: 540–3.
32. Ribera E, Ocaña I, de Otero J, Cortes E, Gasser I, Pahissa A. Prophylaxis of visceral leishmaniasis in human immunodeficiency virus-infected patients. Am J Med **1996**; 100:496–501.
33. Perez-Molina JA, Lopez-Velez R, Montilla P, Guerrero A. Pentamidine isethionate as secondary prophylaxis against visceral leishmaniasis in HIV-positive patients. AIDS **1996**; 10:237–8.
34. Yeung KT, Chan M, Chan CK. The safety of i.v. pentamidine administered in an ambulatory setting. Chest **1996**; 110:136–40.
35. Lawn SD, Wilkinson RJ. Immune reconstitution disease associated with parasitic infections following antiretroviral treatment. Parasite Immunol **2006**; 28:625–33.
36. Barat C, Zhao C, Ouellette M, Tremblay MJ. HIV-1 replication is stimulated by sodium stibogluconate, the therapeutic mainstay in the treatment of leishmaniasis. J Infect Dis **2007**; 195:236–45.