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Combination Treatment for Visceral Leishmaniasis Patients Co-infected with Human Immunodeficiency Virus in India

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SHORT SUMMARY (40 words):

We describe the safety and efficacy of treating visceral leishmaniasis in patients with HIV-VL co-infection with concurrent intravenous liposomal amphotericin B (AmBisome) and oral miltefosine (Impavido) in India. The regimen was safe, well tolerated, with lower relapse rates than monotherapy.

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ABSTRACT

Background

There are considerable numbers of patients co-infected with Human Immunodeficiency Virus (HIV) and Visceral Leishmaniasis (VL) in the VL-endemic areas of Bihar, India. These patients are at higher risk of relapse and death, but there are still no evidence-based guidelines on how to treat them. In this study, we report on treatment outcomes of co-infected patients up to 18 months following treatment with a combination regimen.

Methods

This retrospective analysis included all patients with confirmed HIV-VL co-infection receiving combination treatment for VL at an MSF treatment centre between July 2012 and September 2014. Patients were treated with 30 mg/kg body weight intravenous liposomal amphotericin B (AmBisome®) divided as six equal dose infusions combined with 14 days of 100 mg/day oral miltefosine (Impavido®). All patients were encouraged to start or continue on antiretroviral therapy (ART).

Results

102 patients (76% males, 57% with known HIV-infection, 54% with a prior episode of VL) were followed-up for a median of 11 months (IQR:4-18). Cumulative incidence of all-cause mortality and VL relapse at 6, 12 and 18 months was 11.7%, 14.5%, 16.6% and 2.5%, 6.0%,13.9% respectively. Cumulative incidence of poor outcome at 6, 12 and 18 months was 13.9%, 18.4% and 27.2% respectively. Not initiating ART and concurrent tuberculosis were independent risk factors for mortality, while no factors were associated with relapse.

Conclusions

In this Bihar based study, combination therapy appeared to be well tolerated, safe and effective and may be considered as an option for treatment of VL in HIV co-infected patients.

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INTRODUCTION

Visceral Leishmaniasis (VL; Kala-azar) is a vector-borne disease caused by *Leishmania donovani* parasites. VL is endemic in the Indian state of Bihar, which accounts for 40% of the worldwide burden of VL[1]. Although the prevalence of Human Immunodeficiency Virus (HIV) in Bihar is considered low (0.2-0.3%), it is one of the few states where prevalence is increasing[2]. A recent study from India has suggested that 2.4% of all patients ≥14 years of age presenting with VL were unknowingly co-infected with HIV[3].

HIV-infected patients are more likely to develop symptomatic VL due to reactivation of dormant *Leishmania* infection acquired prior to being infected with HIV, or due to a much higher rate of clinical manifestation following primary *Leishmania* infection after acquiring HIV. Therefore, VL is generally considered an opportunistic infection in patients with HIV and often presents with atypical clinical features[4]. Co-infected patients are at higher risk of relapse and death and this risk appears inversely correlated with CD4 counts. Furthermore, VL adversely affects the response to antiretroviral treatment [4, 5]. Worse outcomes and the treatment challenges faced by co-infected patients as compared to immunocompetent patients are well documented in the literature [6].

There are currently no evidence-based treatment recommendations for co-infected patients in Asia. Moreover, observational studies by MSF in India have shown that outcomes for HIV co-infected patients receiving 20mg/kg AmBisome® (Gilead Pharmaceuticals, Foster City, CA, USA) were substantially worse than in VL patients not known to be HIV co-infected [7-9], while a recent study in Ethiopia showed that 32% of co-infected patients demonstrated parasitological failure following treatment with 30mg/kg AmBisome despite clinical improvement[10]. Therefore, the MSF VL treatment program in Bihar, in collaboration with the Rajendra Memorial Research Institute (RMRI), chose to treat HIV-VL co-infected patients on a compassionate basis using a combination of 30 mg/kg AmBisome and 14 days of miltefosine (Impavido®, Paladin, Canada). This combination was adopted after consultation of experts, taking into account the synergistic properties of AmBisome and miltefosine[6, 11], and has been used in another centre with promising results [12]. Additionally, the compassionate use of miltefosine in combination with liposomal amphotericin B (at 30 mg/kg total dose) in 111 HIV co-infected VL patients in east Africa seems to suggest substantially higher cure rates and lower failure rates both in primary VL and VL relapse than high-dose AmBisome® monotherapy[12]. In this report, we describe the outcomes up to 18 months following treatment with this combination therapy under routine program conditions in Bihar, India.

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METHODS

We did a retrospective analysis of a clinical cohort of co-infected patients using data collected routinely during MSF's VL care programme activities in Bihar. In August 2013, MSF participated in a pilot study to produce evidence on the field safety and effectiveness of new lower dose treatment modalities recommended by the WHO[9] to treat VL in Bihar (CTRI/2012/08/002891). Patients with HIV/VL co-infection were excluded from the study as these treatments are not recommended for this group[5], however their data were recorded in the trial surveillance register and as suggested in the pilot study protocol were treated on a compassionate basis with a combination regimen of AmBisome® and miltefosine (Figure 1).

Visceral Leishmaniasis and HIV diagnosis

Diagnosis of VL involved a clinical case definition (fever >2 weeks and splenomegaly), which was confirmed using the rK39 rapid diagnostic test (DiaMed-IT-Leish). For immunocompetent patients in India it is 98.8% and 97.6% sensitive and specific respectively[13]; its accuracy in immunocompromised patients had not yet been fully established although is likely to be lower. In cases of suspected relapse, or where there was high suspicion despite negative antibody detection tests, confirmation by splenic or bone marrow aspiration was performed.

All patients diagnosed with VL (both primary and relapses) were offered patient Initiated counselling and testing (PICT) for HIV regardless of known HIV status. HIV testing was performed using Determine-HIV 1/2 RDT, and positive patients were referred to the Ministry of Health HIV testing facility within the same hospital for confirmation using two to three further testing kits as per National Acquired immune deficiency syndrome *(*AIDS) Control Organization (NACO) guidelines[14]. Any discordant tests were confirmed using Western Blot®.

Visceral Leishmaniasis treatment protocol

Patients with HIV-VL co-infection were treated as in-patients using a combination of 30 mg/kg body weight AmBisome divided in 6 equal dose infusions given on alternate days, concurrently with 14 days of oral miltefosine. The dose of miltefosine was calculated according to patient weight (≥25 kg 50mg BD; Weight 12-<25 kg, 50mg OD). Test of cure was not routinely performed, with patients discharged as 'initial cures' once they completed a full course of VL treatment and showed clinical improvement,

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cessation of fever, reduction of spleen size and return of appetite at the time of discharge as per WHO descriptions of treatment response[15].

All newly diagnosed HIV patients were advised and counselled to start ART at the nearest government ART centre as per NACO guidelines[14]. Of note, unlike in the African subcontinent, typical VL in India is not yet considered a stage IV AIDS-defining opportunistic infection, hence ART initiation is not typically offered to all HIV patients with VL regardless of CD4-count[16].

Patient follow-up

At the time of discharge, all patients were instructed to return to the treatment centre if experiencing symptoms of relapse. Routine follow-up visits for all patients were scheduled every month to coincide with collection of ART from the ART centre. Follow-up absentees were actively traced. During each follow-up, patients were clinically screened by a physician for signs of relapse, and parasitological confirmation performed in case of suspicion. CD4-counts and ART adherence information were collected, and communication with ART providers maintained to allow integrated longer-term management of patients.

Data collection and analysis

As with all patients treated in the MSF program, socio-demographic characteristics, clinical, anthropometric, laboratory data and data on adverse events were routinely recorded on patient treatment cards and entered in an electronic database by trained data entry encoders. In the case of co-infected patients, CD4 counts and ART status were also routinely recorded, and the program epidemiologist performed double data entry prior to the retrospective analysis. The primary endpoint for analysis was relapse-free survival during follow-up after the start of treatment. For each patient, person-time at risk was calculated from the date of treatment initiation to the date of the 'death', 'first relapse', 'poor outcome' (defined as either relapse or death), 'lost to follow up' (defined as not attending follow-up visit after discharge) or the date of last visit. All data were censored on November 31st 2014. Cumulative incidence of outcome was then estimated using the Kaplan-Meier method. Comparisons between groups were performed using the log-rank test. For risk factor analysis multivariable Cox proportional hazards modelling was performed. All variables associated with the outcome at the $p<0.05$ significance level in bivariate analysis were considered in a forward step-wise multivariable model, with a significance level of $p<0.05$ used to retain variables in the final model. All data analyses were conducted using SPSS version 21 statistical software (IBM Chicago, IL, USA).

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Ethics consideration

This retrospective analysis of clinical cohort data was approved by the Ethics Advisory Group of the International Union Against Tuberculosis and Lung Disease and met the criteria of MSF's International Ethics Review Committee for a study involving the analysis of routinely collected program data. The compassionate use of the combination regimen for co-infected patients was approved ex-ante (on the basis of protocol CTRI/2012/08/002891) by the MSF International Ethics Review Committee and the Institutional Ethics Review Board of RMRI, Patna. All patients were invited to give informed consent prior to HIV testing and all electronic data were analysed anonymously.

RESULTS

Baseline and clinical characteristics

A total of 102 HIV-VL co-infected patients initiated treatment with AmBisome and Miltefosine combination therapy during the study period. The median length of follow-up was 11 (IQR:4-18) months following VL treatment initiation. The majority (76%) was male. Over half (57%) of patients reported being aware they were HIV positive at the time of VL diagnosis; the remainder were diagnosed with HIV at the same time as VL diagnosis. A total of 39 (38%) cases were diagnosed with VL on the basis of clinical criteria and rk39 rapid diagnostic test alone, while 63 (61.8%) patients had either splenic or bone marrow biopsy for additional parasitological confirmation. Baseline CD4-counts were available for 73 patients with a median CD4-count of 169 (IQR:88-230) cells/µl. Of these, 67% had CD4-count <200 cells/µl. All but eight of the cohort received ART treatment; of those who did not, half died within four months of completing treatment for VL. Of those receiving ART, 52 (51%) were already established on ART at the time of initiating VL treatment; the remainder started after completion of VL treatment. Baseline and clinical characteristics of patients are summarized in Table 1 and laboratory parameters are shown in Table 2.

Treatment outcomes

The combination treatment was well tolerated by the majority of patients with minor adverse events recorded among seven patients; five patients reported nausea and vomiting, one patient developed back pain and one patient had rigors. Two patients died after being referred to nearby specialist centres for complications related to HIV before completion of treatment; one died from sepsis secondary to a large scrotal abscess while the other died from bacterial meningitis. Completion of

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treatment was associated with a significant decrease in spleen and liver size at time of discharge from the hospital; however no significant changes in haemoglobin level or body weight were observed by completion of treatment. Kidney and liver function tests were performed after treatment completion on a limited number of patients suspected to have complications; changes in mean values of these tests are shown in Table 3.

Sixteen (15.7%) deaths were identified during the follow-up period. Two patients died before completion of treatment, while four others died within 2 months following treatment completion. Median time to death was 3.3 months (IQR: 1.3-6.5). Cumulative incidence of mortality at 6, 12 and 18 months was 11.7, 14.5 and 16.6% respectively (Supplementary Figure 1). Among the patients discharged as initial cure, eight cases of VL relapse were identified during follow-up with a median time to relapse of 11 (IQR:4-15) months. The estimated risk of relapse was 2.5, 6.0 and 13.9% at 6, 12 and 18 months respectively. In terms of overall VL treatment response, the estimated cumulative incidence of poor outcomes by 6, 12 and 18 months were 13.9, 18.4 and 27.2% respectively. One patient was diagnosed with macular lesions suggestive of PKDL 13 months after completing VL treatment.

Predictors for death, relapse and poor outcome.

No demographic or clinical characteristics were significantly associated with relapse in bivariate analysis, even though a diagnosis with tuberculosis (supplementary table 1) showed a strong association (hazard ratio (HR): 9.5; 95%CI:0.9 –97.9; p=0.06). Concurrent tuberculosis, haemoglobin ≤6 g/dl, baseline CD4-count <100 cell/µl and lack of ART initiation were identified as risk factors for mortality in bivariate analysis. However, only infection with tuberculosis (adjHR:5.3; 95%CI:1.6–17.8; p<0.01) and ART initiation status maintained statistical significance through the forward step-wise approach and were therefore retained in the final multivariable model for mortality (Table 4). Of the 102 patients presenting, 73 had documented CD4 counts at the time of treatment; for the purposes of multivariable modelling multiple imputation was used to complete the dataset; however, this had minimal impact on the model output[17].

In the final multivariable model, failure to start ART was an independent risk factor for mortality compared to those who started ART prior to VL treatment (adjHR:8.0;95%CI:2.0-32.5;p<0.01). ART initiation following VL treatment was similarly associated with higher mortality than those already established on ART, but did not reach statistical significance (adjHR 2.8;CI95%:0.8-9.5;p=0.101).

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In terms of overall poor outcome, only concurrent tuberculosis (adjHR:7.5;95%CI:2.5–22.1; p<0.01) was retained as an independent risk factor (Supplementary Table 2).

DISCUSSION

This is the first report on HIV-VL co-infected patients receiving treatment with a combination of AmBisome and Miltefosine therapy in the Indian subcontinent. Our data suggests that combination therapy is a well-tolerated and effective treatment regimen for an episode of VL in HIV-VL co-infection within the Indian setting. The choice of 14 days of concurrent oral treatment had the added benefit of ensuring compliance as both treatments can be administered during the inpatient stay. However, a high risk of relapse and early death, similar to earlier studies on short and long-term outcomes of co-infected patients is described in this cohort, particularly among tuberculosis patients and those not initiated on antiretroviral treatment. When compared to patients with VL not known to be HIV-infected treated with a lower dose monotherapy of 20mg/kg AmBisome® in the same setting[7], the outcome among co-infected patients observed in our study was considerably worse - mortality and relapse rates at 12 months for patients were 0.9% and 3.7% compared to 14.5% and 6.0% respectively for the co-infected patients described here.

In HIV-VL co-infected patients already taking or initiated on ART, this study demonstrated slightly higher overall mortality but substantially reduced relapse rates compared to co-infected patients treated with a lower dose 20-25mg/kg AmBisome® monotherapy, with mortality and relapse rates at 12 months of 11.2% and 6.4% compared to 8.7% and 16.2% respectively[8]. Concurrent tuberculosis was found to be an independent risk factor for overall poor outcome in our multivariable model, similar to other studies[8, 9]. No other socio-demographic or clinical factors were found to be associated with poor outcomes.

Baseline CD4-counts at the time of VL diagnosis were low in our cohort, with counts <100 cells/µl at baseline being a significant risk factor for mortality in bivariate analysis, consistent with reports from an Ethiopian co-infected cohort [18]. Patients receiving ART had substantially lower mortality than those who did not, confirming results of earlier studies on co-infected patients in the same programme [8, 9] and re-enforcing the need for the central place of ART in the management of this group of patients. Considering relapse, no associations with demographic characteristics were found, which is in keeping with a systematic review describing predictors of VL relapse in HIV-infected patients [19]. However, unlike this review, our study failed to establish low CD4-count and previous history of VL treatment as risk factors for relapse, possibly for lack of power.

This study has several limitations. Being a retrospective analysis of program data, and despite demographic and clinical data related to VL treatment being complete, some important HIV-related

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data were missing; particularly timely CD4-counts following diagnosis. Secondly, all-cause mortality was used in the analysis, and as such we were not able to ascertain documented causes of deaths discovered during follow-up. However, all but two patients were discharged in good clinical condition following treatment completion, and the two deaths occurring before completion of treatment were not considered related to the therapy. Therefore, we believe it is unlikely that any of the 16 deaths recorded in the cohort was a treatment related serious adverse event. The two patients who died after relapsing did so after re-treatment was completed and were discharged with improved clinical condition. Finally, test of cure was not routinely performed on patients unless there was suggestion of treatment failure clinically, of which there were none; therefore the study may have under-diagnosed initial treatment failures.

Presently, the WHO recommends monotherapy with AmBisome® up to a total of 40mg/kg in divided doses for over a month in HIV-VL co-infected patients worldwide; however this recommendation is made on the basis of patient case series involving *L.infantum* in Europe alone [4]. To our knowledge, no case of *L.donovani* infection in a patient with HIV treated with this regimen has been documented in the Indian sub-continent, while high dose monotherapy with AmBisome already appears to be ineffective in African *L.donovani* infection [10], where studies evaluating the combination of AmBisome and miltefosine in co-infected patients have already begun [20]. We therefore suggest that the use of this WHO recommended regimen and the combination described in this study need to be investigated in further studies in order to help establish optimal dosing and safety profiles to help determine the best management of this challenging group of patients.

Considering the high probability of relapse in co-infected patients, there is a need to provide a safe and effective treatment while protecting the limited drugs available from the development of resistance. This is more pertinent since resistance mechanisms to amphotericin B have recently been described [21], and evidence from Ethiopia that high-dose AmBisome® monotherapy was much less effective in HIV-positive VL relapse patients who had received previous treatment with AmBisome® or amphotericin B compared with those who had not[10].

Our findings have a number of implications for policy and practice in India. Current WHO and NACO guidelines describe ‘atypical disseminated VL’ as a stage IV-defining opportunistic infection, rather than simply ‘visceral leishmaniasis’ and therefore do not recommend initiation of ART in all HIV patients with typical VL irrespective of CD4 count. This contrasts with WHO expert committee on VL recommendations delivered in 2010, where typical VL infection in HIV-infected patients is clearly identified as an AIDS-defining illness. These inconsistencies cause confusion in the field when making decisions to start ART in co-infected patients [16]. The outcomes of the recent expert meeting between NACO and the NVBDCP in India to develop guidelines for the management of HIV-VL co-infection [22]

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is a strong first step in developing clearer recommendations and convergence between WHO ART guidelines and VL guidelines. In turn, these study results strengthen emerging evidence that typical VL should be considered as a clear entry criterion in the stage IV definition of HIV, support the need to offer PICT to all patients diagnosed with VL and crucially that extended follow-up of co-infected patients is required to ensure relapses are detected early and treated appropriately. This needs to be done using a co-ordinated multidisciplinary approach between VL and HIV/AIDS programs.

In conclusion, the administration of a combination therapy of AmBisome® and miltefosine appears safe and effective among HIV-VL co-infected patients under programme conditions in India. Early diagnosis of the co-infection, prompt initiation of ART and anti-leishmania therapy, screening and treatment for TB and extended follow-up may lead to more favourable treatment outcomes.

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DISCLOSURES

None of the contributing authors have any conflict of interest.

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FIGURE LEGEND

Figure 1: Flow chart of analysis of 102 HIV VL co-infected patients, Bihar India

Supplementary Figure 1: Kaplan Meier curves showing cumulative incidence of mortality and relapse over time

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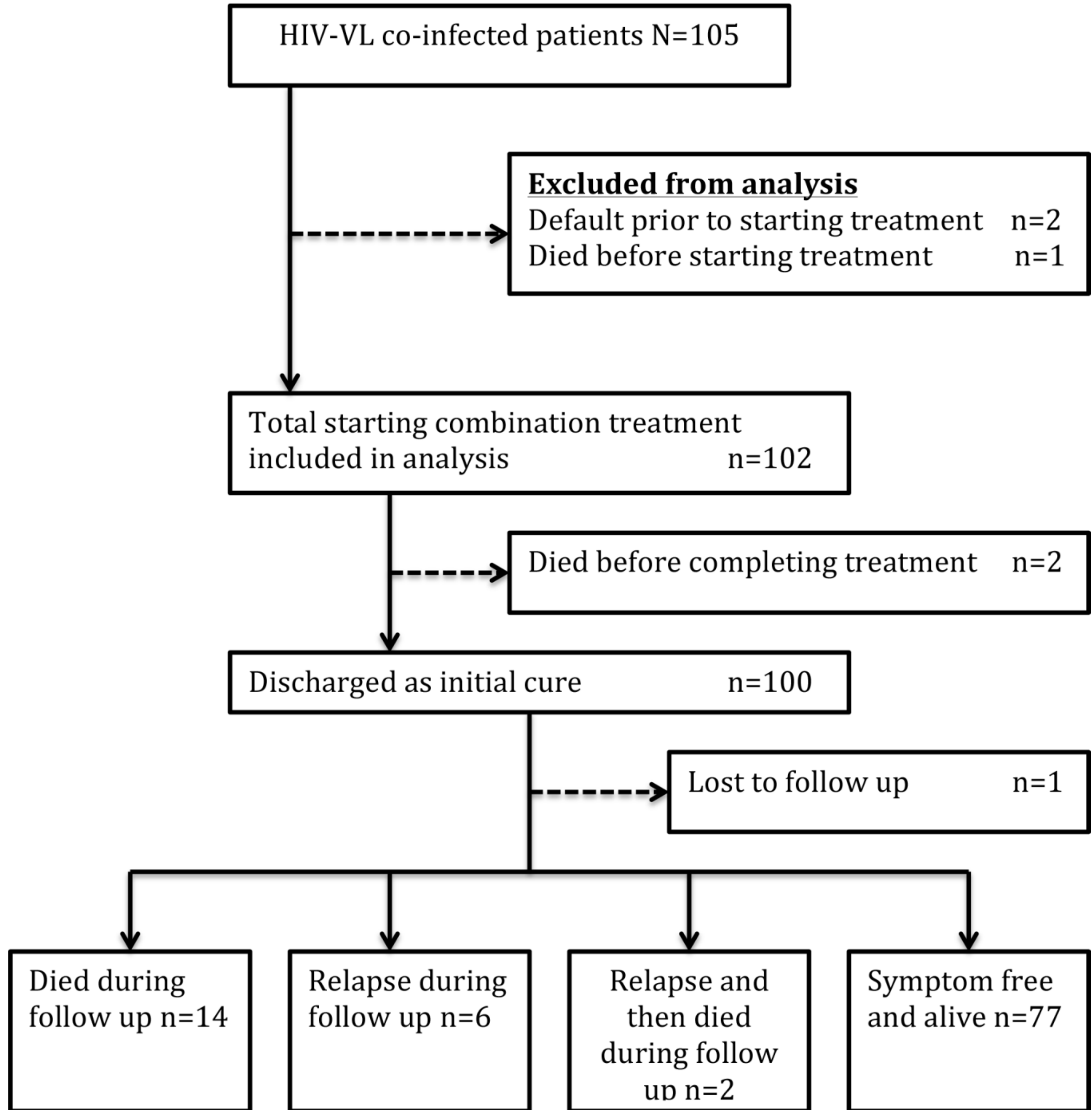

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Table 1: Baseline characteristics of patients with visceral leishmaniasis (VL) and human immunodeficiency virus (HIV) co-infection

Variable	N	%	Median (IQR)
Sex			
Male	77	75.5	
Female	25	24.5	
Age group (years)			36 (30,45)
<15	2	2.0	
15-29	13	12.7	
30-44	58	56.9	
45-59	23	22.5	
≥60	6	5.9	
Time from symptoms onset to diagnosis			8 (4,12)
>4 weeks	64	62.7	
≤4 weeks	38	37.3	
History of previous treatment for VL			1 (0,1)
Second or more relapse	13	12.7	
First relapse	42	41.2	
Primary episode	47	46.1	
VL drug used previously			
None	47	46.1	
AmBisome (20mg/kg body weight)	18	17.6	
Miltefosine	12	11.8	
Amphotericin B	11	10.8	
Sodium Stibogluconate (SSG)	8	7.8	
Fungisome	2	2.0	
AmBisome (5mg/kg body weight) and Miltefosine combination	1	1.0	
Drug Unknown	3	2.9	
Spleen size, (in cm)			8 (6,10)
>6	60	58.8	
3-6	36	35.3	
<3	6	5.9	
Liver size, (in cm)			2 (0,4)
>3	30	29.4	
1-3	33	32.4	
0	39	38.2	
Body mass index (kg/m^2)			17.4 (15.8,18.7)
<16	29	28.4	
≥16	73	71.6	
Concurrent Tuberculosis diagnosis			

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Positive	9	8.8
Negative	93	91.2
HIV status at time of treatment for VL		
Previously diagnosed as HIV positive	58	56.9
Diagnose HIV positive at time of VL treatment	44	43.1

IQR- interquartile range

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Table 2: Baseline laboratory parameters of patients with visceral leishmaniasis (VL) and human immunodeficiency virus (HIV) co-infection

Variable	N	%	Median (IQR)
Hemoglobin (g/dl) (n=102)			8.2 (6.4,9.7)
<6	16	15.7	
6-7	32	31.4	
≥8	54	52.9	
Potassium **(mEq/L)** (n=65)			3.9 (3.5,4.2)
2.5-<3	1	1.5	
3-<3.5	15	23.1	
≥3.5	49	75.4	
Creatinine mg/dl (n=97)			
<1.2	81	83.5	
1.2-2	11	11.3	
>2	5	5.2	
SGPT U/L (n=92)			26.3 (17.1,39.3)
<48	77	41.3	
>48-200	14	56.5	
>200	1	2.2	
SGOT U/L (n-92)			51 (35.3,66.7)
<46	38	41.3	
>46-200	52	56.5	
>200	2	2.2	
Bilirubin (mg/dl) (n=68)			
≥ 1.9	1	1.5	
0.5-1.9	28	41.2	
<0.5	39	57.4	
Platelets count cells/ul (n=89)			146000 (109500,202500)
<150000	46	51.7	
≥150000	43	48.3	
Baseline CD4 count, cells/ul (n=73)*			169 (88.5,230.5)
<100	22	30.1	
100-<200	27	37.0	
200-<350	16	21.9	
≥350	8	11.0	

*window of 3 months prior to VL treatment until 3 months after.
IQR- interquartile range, SGPT-Serum glutamate-pyruvate transaminase, SGOT-Serum glutamic oxaloacetic transaminase

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Table 3. Clinical and laboratory parameters before and after treatment (assessed ≥ day 14) with combination therapy

Variable	Before treatment Mean (SD)	End of treatment Mean (SD)	Within patient difference	p-value
Weight, Kg (n=100)	44.3(8.8)	44.6 (8.7)	0.3(-0.1, 0.6)	0.15
Spleen size, cm (n=101)	8.2 (4.1)	4.1 (3.9)	-4.1 (-4.7, -3.6)	**<0.001**
Liver size, cm (n=101)	2.2 (2.3)	0.8(1.4)	-1.4 (-1.9,-0.9)	**<0.001**
Hemoglobin, g/dL (n=93)	8.2 (2.1)	8.1 (1.8)	-0.2 (-0.4,0.1)	0.27
Potassium (n=17)	3.7 (0.6)	4.3 (0.7)	0.5 (0.2, 0.9)	**0.003**
SGPT, U/L (n=18)	37.8 (33.0)	29.2 (17.2)	-8.6 (-26.1,8.9)	0.31
SGOT, U/L (n=17)	61.9 (56.3)	52.8(43.9)	-9.1 (-46.1,27.9)	0.61
Platelets cells/lL (n=12)	156667 (77713)	222250 (115180)	65583 (30868,100299)	**0.002**

SD- standard deviation; SGPT-Serum glutamate-pyruvate transaminase, SGOT-Serum glutamic oxaloacetic transaminase

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Table 4. Risk factors analysis for mortality in patients with VL-HIV co-infection

Variable	Death N (%)	Survived N (%)	Crude HR (95% CI)	Crude p-value	Adjusted HR (95% CI)	Adjusted p-value
Sex						
Female	6(24.0)	19(76.0)	1.7(0.6,4.8)	0.28		
Male	10 (13.0)	67(87.0)				
Age (years)						
>40	7(17.1)	34(82.9)	1.2(0.4,3.2)	0.74		
≤40	9(14.8)	52(85.2)				
TB diagnosis						
Positive	4(44.4)	5(55.6)	6.6(2.0,22.0)	0.002	**5.3 (1.6, 17.8)**	**0.008**
Negative	12(12.9)	81(86.0)				
History of previous VL treatment						
Yes	7(12.7)	48(87.3)	0.6(0.2,1.6)	0.32		
No	9(19.1)	38(87.1)				
Spleen size (cm)						
>8	6(14.3)	36(85.7)	0.8(0.3,2.2)	0.67		
≤8	10(16.7)	50(83.3)				
BMI (kg/m^2)						
<16	7(24.1)	22(75.9)	2.1(0.8,5.8)	0.13		
≥16	9(12.3)	64(87.7)				
Hemoglobin (g/dL)						
≤ 6	6(30.0)	14 (70)	2.9(1.03,7.9)	0.04		
>6	10(12.2)	72(87.8)				
Baseline CD4 count (cells/ul)*						
<100	8(25)	23(75)	2.0 (1.1,30.8)	0.04		
≥100	8(11.7)	62(88.3)				
ART initiation						
Never started	4(50.0)	4(50.0)	9.1 (2.3,36.5)	0.002	**8.0 (2.0,32.5)**	**0.004**
After VL diagnosis	8(19.0)	34(81.0)	3.2(0.95,10.7)	0.06	**2.8 (0.8,9.5)**	**0.101**
Before VL diagnosis	4(7.7)	48(92.3)				

*73/102 baseline CD4 counts were available; the remainder were completed through multiple imputation

HR – Hazard Ratio; CI – Confidence Interval; VL – Visceral Leishmaniasis; ART- Anti-Retroviral Therapy

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