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# Impact of the immune reconstitution inflammatory syndrome (IRIS) on mortality and morbidity in HIV-infected patients in Mexico<sup>☆</sup>

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## SUMMARY

**Objectives:** To estimate the impact of immune reconstitution inflammatory syndrome (IRIS) on morbidity and mortality in patients starting highly-active antiretroviral therapy (HAART).

**Methods:** A retrospective cohort study of HIV-positive patients starting HAART was conducted at a tertiary care referral center in Mexico City. We estimated the incidence of IRIS, hospitalizations and death rates during the first 2 years of HAART. The relative risk of death (RR) and hospitalization for patients with IRIS were adjusted for relevant covariates using regression methods.

**Results:** During the 2-year follow-up period, 27% of patients developed IRIS (14 IRIS cases per 100 person-years). The relative risk of death among patients who developed IRIS was 3 times higher (95% confidence interval (CI) 1.19–7.65,  $p = 0.03$ ). After adjusting for previous opportunistic infections we still observed a higher death rate among patients with IRIS (RR 2.3, 95% CI 0.9–5.9,  $p = 0.09$ ). An effect modification of IRIS over mortality was observed by previous opportunistic infection.

**Conclusions:** IRIS-associated mortality is strongly confounded by previous opportunistic infection. Patients with AIDS who eventually developed IRIS had the highest risk of death at the 2-year follow-up.

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## 1. Introduction

The use of highly active antiretroviral therapy (HAART) leads to quantitative and qualitative changes in the immune response that have dramatically improved the survival and quality of life of people living with HIV.<sup>1–4</sup> However, HAART initiation is sometimes associated with a paradoxical reaction characterized by a generalized inflammatory response or a localized inflammation of tissues previously infected with opportunistic pathogens.<sup>5,6</sup> The manifestations of this inflammatory response have been grouped together under the term immune reconstitution inflammatory syndrome (IRIS) or immune reconstitution disease (IRD).

Previous observational studies suggest that IRIS has a major effect on morbidity in HIV-infected patients starting antiretroviral treatment, but its impact on mortality is yet to be established.<sup>7–10</sup> Moreover, it has been suggested that IRIS has a role in the early mortality of patients starting antiretroviral (ARV) therapy at

advanced stages of the disease,<sup>11</sup> but to date few studies have addressed this issue.<sup>8,9,12</sup> We conducted a retrospective study in a cohort of HIV-infected patients receiving HAART in an HIV/AIDS clinic in Mexico City. Herein we describe the incidence of IRIS and mortality rate during the first 2 years of ARV therapy in our clinic, and compare mortality rates in patients with IRIS and without IRIS. We also compare the hospitalization rate between groups to estimate the impact of IRIS on morbidity.

## 2. Methods

### 2.1. Study design and population

The source population for this retrospective cohort analysis was all HIV-infected adults receiving medical care at the HIV/AIDS clinic of the Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubirán (INCMNSZ). The INCMNSZ is a tertiary care referral center located in Mexico City that provides medical care for people from different regions of the country. Patients who began ARV therapy between October 2001 and November 2007 were included in this study if they had attended at least one documented medical visit to the HIV clinic after starting HAART. Information through December 31, 2008 was included in the analysis. Patients were considered as lost to follow-up if they had a period of a year or

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longer without visits by that day, or did not complete a 2-year follow-up period. Patients starting treatment during this period at other hospitals who were afterwards referred to continue their care at our clinic were excluded from the analysis. The institutional review board of the INCMNSZ waived the informed consent process for the use of clinical data for retrospective analysis and publication as part of the CCASAnet collaboration (Caribbean, Central and South America Network for HIV Research).<sup>13</sup>

## 2.2. Data collection

The HIV/AIDS clinic has been prospectively recording demographic, clinical, and laboratory information for each patient since 2000, and maintains an electronic database with this information that is updated on a daily basis. The database is sporadically updated with an active search for those patients who were lost to follow-up; the last update occurred in March 2007. All patients starting ARV treatment were identified using the electronic database. Demographic information, medical history prior to HAART initiation, and laboratory baseline information was retrieved from the database. Information on clinical symptoms and conditions after HAART initiation was retrieved from the medical files.

## 2.3. Assessment of IRIS

Using a standardized format, information from patient medical records was collected to evaluate the appearance of symptoms or conditions previously identified as related to IRIS (fever, lymph node enlargement, oral lesions, worsening clinical manifestations of previously diagnosed opportunistic infections (OIs), neurological deficit, pneumonia, tuberculosis, herpes zoster, skin changes, and visual disturbances). All files of patients with any of these symptoms were independently reviewed by two infectious diseases specialists (BCR and JSM) with experience treating HIV-infected patients, to determine whether the clinical information gathered in the files were IRIS events. An IRIS event was defined a priori using the AIDS Clinical Trials Group (ACTG) immune reconstitution inflammatory syndrome generic criteria (revised May 24, 2005). All IRIS events were classified as either 'probable' or 'confirmed'. Patients whose condition did not meet the ACTG criteria were registered as having an 'event not related to IRIS'. Additionally, an IRIS episode was considered either 'paradoxical' or 'unmasking', extrapolating the classification created by the International Network for the Study of HIV-associated IRIS for tuberculosis to all other IRIS-related clinical entities.<sup>14</sup> In brief, an unmasking IRIS event was defined as all new OIs diagnosed after starting HAART with specific atypical features or excessive inflammatory manifestations, while a paradoxical IRIS event was said to be a worsening or recurrent clinical manifestation of a previously diagnosed and successfully treated OI within a 6-month period after starting HAART. We also classified events as severe or not severe, defined a priori as follows. We considered severe IRIS to have occurred when this event led to the patient's hospitalization, to the use of an invasive life support measure, or the requirement of a major invasive diagnostic or therapeutic intervention (e.g., bronchoscopy, abdominal or thoracic exploratory surgery, cerebral biopsy, blood transfusion, or any surgical procedure). Date of start of an IRIS event was considered as the first date when new symptoms of a condition that was finally diagnosed and classified as an IRIS event or worsening of a pre-existing condition were registered in the medical file.

## 2.4. Outcome ascertainment

Hospitalizations and deaths were ascertained from death certificates, medical charts, and the HIV/AIDS clinic database. In

the IRIS group we collected hospitalization data exclusively on those hospital admissions related to the IRIS event, while for the non-IRIS group, all hospitalizations were included in the analysis.

## 2.5. Assessment of other variables

The analysis included other variables to control for potential confounding effects. These variables were modeled as time-fixed, and the value taken was that at the time of HAART initiation.

We considered sex, HIV viral load >75 000 copies/ml (this cut-off value was used because our study included patients from 2001, and at this time the upper limit of viral load detection was 75 000 copies/ml at our center), the presence of OIs before HAART initiation, and type of HAART (protease inhibitors vs. non-nucleoside reverse transcriptase inhibitors (NNRTIs)) as binary variables. Age, weight, and CD4+ T cell counts were classified as continuous variables. Finally, the type of OI previous to HAART initiation, behavioral risk group, and type of HAART regimen were considered categorical variables with discrete values.

## 2.6. Statistical analysis

The primary outcome measure was death during the first 24 months after HAART initiation. The secondary outcome was the first episode of hospitalization after HAART initiation within the first 24 months of HAART. Patients were considered exposed after having had an IRIS event and thereafter, regardless of the duration of the IRIS event. Consequently, subjects contributed to person-time at risk exposed to IRIS only after they had the IRIS event. Patients were censored at death, treatment interruption, last documented visit to the clinic or at 2 years after HAART initiation. Characteristics of patients at the date of HAART initiation are described using proportions and the median and interquartile range (IQR) as measures of central tendency and dispersion. Fisher's exact test and Wilcoxon non-parametric tests were used as appropriate to compare differences in proportions and distribution of characteristics between patients with and without IRIS. Patient follow-up (person-time at risk) began at the date of HAART initiation. The incidence rate of IRIS was calculated by dividing the number of IRIS events by person-time at risk, expressed in number of cases per 100 person-years.

During the exploratory data analysis, we used a Cox proportional hazards model to search for risk factors for IRIS that could also predict death, in order to identify their roles as potential confounders in the final analysis. In order to estimate the relative risk of death, rate ratios (RR) and two-sided 95% confidence intervals (CI) were derived from hazard ratios obtained from the Cox proportional hazard regression model, adjusting for CD4+ T cell counts, weight, and age at baseline. We first fitted a model containing the variables sex, age, weight, baseline CD4+ T cell counts and the presence of OIs diagnosed before starting HAART, HIV viral load and type of HAART regimen, and then removed variables that did not independently predict death (except for CD4+ T cell count which was included in the final model, as it has been consistently observed to be the main predictor of survival among HIV-infected patients) based on an a priori defined  $\alpha$  of 0.2. To evaluate the differentiated effect of IRIS on mortality by different baseline characteristics, we used an interaction term for IRIS with baseline characteristics of interest. The validity of the proportional hazards assumption was assessed graphically with a complementary log–log plot and using the Grambsch–Therneau method.<sup>15</sup> We used Kaplan–Meier estimates to construct survival curves and compared the cumulative hazards function using the log-rank test.

To estimate hospitalization rates, we only considered the first hospitalization after HAART initiation. Patients were censored at

the date of first hospitalization, death, or last documented visit to the clinic or at 2 years after HAART initiation. Hospitalization RR and two-sided 95% CI were estimated using a Poisson regression model to adjust for baseline CD4+ T cell count, weight, and the occurrence of OIs before HAART initiation. The procedures for fitting this regression model were similar to those described to estimate the mortality rate ratio.

Missing data on weight (2.8%), CD4+ T cell count (1.3%), and HIV viral load (8.2%) at baseline was imputed using simple imputations based on multivariate regression models. Statistical analysis was performed using STATA version 10.<sup>16</sup>

### 3. Results

#### 3.1. Cohort patient characteristics

Between October 2001 and November 2007, 413 patients initiated HAART. Of these, 23 (6%) cases were excluded from the analysis because they started treatment at other centers and were referred to our clinic to continue their follow-up. The 390 patients included in the analysis contributed 756 person-years of follow-up, with a median of 730 days of follow-up (interquartile range (IQR) 591–730 days). Demographic and clinical baseline characteristics of the cohort are presented in Table 1. On December 31, 2008 all patients, except those who had died (5%), continued on follow-up or had completed 2 years of follow-up.

Patients were mostly young adults (median age 35 years), most were male (88%), and all had started treatment either with an NNRTI-based regimen (efavirenz, 67%) or a protease inhibitor (33%) regimen. One hundred and twenty-eight patients (33%) had had an OI diagnosed before starting HAART, and 27 (21%) of them had had at least two diagnosed OIs before starting therapy. The most common OIs diagnosed before HAART were *Pneumocystis jirovecii* pneumonia (PCP; 32 events), tuberculosis (21 events), and Kaposi's sarcoma (14 events). Only three patients had cryptococcal meningitis before HAART and none of them developed IRIS. Having a particular OI before HAART did not increase the risk of death.

#### 3.2. Immune reconstitution inflammatory syndrome

During the 2-year follow-up period, 107 (27%) patients developed IRIS, corresponding to an incidence rate of 14 IRIS cases per 100 person-years of follow-up. The median onset of IRIS was 69 days (IQR 20–193 days), and the median duration of the episodes was 17 days (IQR 8–39 days). Of the 107 cases, 16 (15%) were classified as probable and 91 (85%) as confirmed.

The microorganism most frequently associated with IRIS events was varicella zoster, accounting for 32% of all IRIS cases (see Table

2), all of them in patients with no prior herpes zoster episodes and with no severe episodes. The median onset of varicella zoster-related IRIS after HAART initiation was 143 days (IQR 56–332 days). Other frequent presentations of IRIS were *Mycobacterium tuberculosis* (12 events, 11% of all IRIS cases), *Mycobacterium avium* complex (MAC; 10 events, 9%), PCP (six events, 6%), *Cryptococcus neoformans* (five events, 5%), Kaposi's sarcoma (four events), and cytomegalovirus (four events) associated IRIS. Most cases of tuberculosis-associated IRIS<sup>8</sup> presented as tubercular lymphadenitis, in two patients it presented as pulmonary tuberculosis (one unmasking and one paradoxical), and in two others as intestinal tuberculosis (both paradoxical). Tuberculosis-associated IRIS developed in eight of the 21 (38%) patients previously diagnosed with tuberculosis (paradoxical tuberculosis) and accounted for most of the paradoxical IRIS events (see Table 2). Paradoxical tuberculosis-associated IRIS tended to present earlier after HAART initiation (median 15.5 days, IQR 12–26.5 days) than unmasked tuberculosis IRIS (median 58.5 days, IQR 26–76 days). The median onset of presentation in MAC-associated IRIS was 29 days after HAART initiation (IQR 8–58 days), and most cases presented as lymphadenitis (nine cases). Four cases of *C. neoformans* IRIS presented as cryptococcal meningitis and one as lymphadenitis.

Unmasking cases were more frequent ( $n = 87$ , 81%) than paradoxical cases ( $n = 20$ , 19%). Overall, patients with unmasking and paradoxical cases were similar with regards to age, weight, CD4+ T cell count, proportion of males, and patients with a high viral load ( $>75$  000 copies). In total, 17 patients (16%) presented with severe forms of IRIS; those with the paradoxical form were more likely to develop severe events as compared to those with unmasking events (35% vs. 11%,  $p = 0.017$ ) (see Table 2).

Patients with IRIS had on average lower weight and CD4+ T cell count at baseline than patients without IRIS, and were more likely to have had an OI before HAART initiation (Table 1). These variables remained independently associated with IRIS when adjusting for potential confounders (see Table 3).

The most common OIs diagnosed before HAART among the 107 patients who developed IRIS were tuberculosis (12 events; seven disseminated and five pulmonary), PCP (10 events), disseminated histoplasmosis (seven events), and Kaposi's sarcoma (six events). Patients with tuberculosis, but not those with any other OIs before HAART initiation, had an increased risk for developing IRIS (RR 3.5, 95% CI 1.9–6.4) after adjusting for baseline CD4+ T cell count and weight (see Table 3).

#### 3.3. Mortality associated with IRIS

During the 2 years of follow-up, the cumulative all-cause mortality in the cohort was 4.9%, corresponding to a mortality

**Table 1**  
Baseline characteristics of HIV-infected adults receiving medical care at the INCMNSZ

Baseline characteristics	Total (N=390) (100%)	IRIS (n=107) (27%)	No IRIS (n=283) (73%)	p-Value
Male	342 (88%)	96 (90%)	246 (87%)	0.5
Age, median (IQR)	35 (29–42)	34 (28–41)	35 (30–42)	0.46
Weight in kg, median (IQR)	62 (54–70)	58 (48–67)	63 (55–71)	0.0005
CD4+ cell/ml, median (IQR)	87 (32–184)	48 (20–110)	113 (40–205)	<0.0001
HIV-1 viral load, copies/ml				
≥75 000	293 (75%)	86 (80%)	207 (73%)	0.15
<75 000	97 (25%)	21 (20%)	76 (27%)	
HAART regimen				
NNRTI	263 (67%)	73 (68%)	190 (67%)	0.9
Protease inhibitor	127 (33%)	34 (32%)	93 (33%)	
Opportunistic infection before HAART	128 (33%)	55 (51%)	73 (26%)	<0.001
Type of IRIS				
Paradoxical		20 (19%)	NA	NA
Unmasking		87 (81%)		

Results are n (%), unless otherwise specified. INCMNSZ, Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubirán; IRIS, immune reconstitution inflammatory syndrome; IQR, interquartile range; HAART, highly-active antiretroviral therapy; NNRTI, non-nucleoside reverse transcriptase inhibitor; NA, not applicable.

**Table 2**  
Description of 107 patients who presented IRIS, according to clinical presentation

Characteristic	Unmasking n = 87 (81%)	Paradoxical n = 20 (19%)	p-Value
Male	78 (90%)	18 (90%)	1.0
Age, median (IQR)	34 (28–42)	34 (28–37)	0.6
Weight, median (IQR)	60 (48–67)	55 (46–64)	0.25
CD4+ T cells, median (IQR)	52 (22–117)	35 (12–73)	0.12
CD4+ <50 cells/ml	41 (47%)	14 (70%)	0.085
CD4+ <200 cells/ml	77 (89%)	19 (95%)	0.68
HIV RNA viral load >75 000 copies/ml	70 (80%)	17 (85%)	0.46
NNRTI	61 (70%)	12 (60%)	0.27
Previous opportunistic infection	35 (40%)	NA <sup>a</sup> (100%)	NA
Days of onset after HAART initiation, median (IQR)	80 (28–199)	27 (13–114)	0.02
Type of IRIS event			
Varicella zoster (34 events)	34 (39%)	0	<0.01
Mycobacterium tuberculosis (12 events)	4 (5%)	8 (40%)	<0.01
Mycobacterium avium complex (10 events)	8 (9%)	2 (10%)	0.57
Pneumocystis jirovecii pneumonia (6 events)	4 (5%)	2 (10%)	0.13
Cryptococcus neoformans (5 events)	2 (2%)	3 (15%)	0.04
All other (40 events)	–	–	–
Duration of the event in days, median (IQR)	14 (7–39)	25 (18–38)	0.06
Survival time after IRIS event, median (IQR)	42 (20–288)	33 <sup>b</sup>	–
Severe presentation	10 (11%)	7 (35%)	0.017
Deaths	7 (8%)	1 (5%)	0.6

Results are n (%), unless otherwise specified. IRIS, immune reconstitution inflammatory syndrome; IQR, interquartile range; NNRTI, non-nucleoside reverse transcriptase inhibitor; HAART, highly-active antiretroviral therapy; NA, not applicable.

<sup>a</sup> By definition, all paradoxical cases had a previous opportunistic infection.

<sup>b</sup> Only one patient with paradoxical IRIS died.

rate of 2.5 deaths per 100 person-years. The overall survival time after an IRIS event was 39 days (IQR 27–166 days). The mortality rate among patients who did not develop IRIS was 1.8 deaths per 100 person-years in comparison to 4.9 deaths per 100 person-years among patients who had an IRIS event. The crude mortality ratio was 3.02 (95% CI 1.19–7.65,  $p = 0.03$ ) when comparing patients with IRIS to patients without IRIS. After adjusting for age, CD4+ T cell count, and weight, the mortality ratio was 2.86 (95% CI 1.08–7.58,  $p = 0.03$ ). When we introduced the presence of OIs before HAART initiation into the model, the mortality ratio decreased to 2.28 (95% CI 0.87–5.99,  $p = 0.09$ ) (see Table 4).

We observed that the diagnosis of an OI before starting HAART modifies the effect that IRIS has on the 2-year mortality (see Table 5). Figure 1 shows the differences in the 2-year cumulative risk of death according to the presence of an OI previous to HAART and subsequent development of IRIS. The log-rank test ( $p = 0.004$ ) indicates that the risk of death was significantly different across strata, with patients with OIs diagnosed before starting HAART who subsequently developed IRIS being at the highest risk of death in the 2 years of follow-up (RR 8.1, 95% CI 2.4–27.5,  $p = 0.001$ ).

**Table 3**  
Baseline characteristics associated to the risk of IRIS

Baseline characteristic	Relative risk (95% CI)	
	Univariate model	Multivariate model
Male vs. female	1.3 (0.7–2.4)	–
Age <sup>a</sup>	0.9 (0.8–1.1)	–
Weight <sup>b</sup>	0.7 (0.6–0.9)	0.8 (0.7–0.9)
CD4+ T cells (<200 vs. >200 cells/ml)	2.8 (1.5–5.2)	2 (1.1–3.8)
HIV RNA viral load (>75 000 vs. <75 000 copies/ml)	1.0 (0.9–1)	–
Previous opportunistic infection (yes vs. no)	2.8 (1.9–4)	2.2 (1.5–3.3)
Tuberculosis (yes vs. no)	3.8 (2.1–7)	3.5 (1.9–6.4)

IRIS, immune reconstitution inflammatory syndrome; CI, confidence interval.

<sup>a</sup> Increased risk per every 10 years increase in age.

<sup>b</sup> Increased risk per every 10 kg increase in weight.

### 3.4. Hospitalization associated with IRIS

During the 2 years of follow-up, 59 patients (15%) in the cohort were hospitalized at least once, corresponding to an incidence rate of 2.4 hospitalizations per 100 person-years taking into account only the first hospitalization. A greater proportion of patients with IRIS than without IRIS were hospitalized (32% vs. 9%,  $p < 0.001$ ), and patients with IRIS spent on average 6.1 days hospitalized in comparison with 1.4 days among patients without IRIS. The hospitalization rate among patients who did not develop IRIS was 1.2 per 100 person-years in comparison with 6.1 per 100 person-years among patients who developed IRIS. The unadjusted hospitalization rate ratio was 3.6 (95% CI 2.2–6.03,  $p < 0.001$ ) when comparing patients with IRIS to patients without IRIS. After adjusting for age, baseline CD4+ T cell count, presence of OI prior to HAART initiation, and weight, the hospitalization ratio was 3.2 (95% CI 1.9–5.6,  $p < 0.001$ ) (see Table 4).

## 4. Discussion

In this retrospective cohort study of HIV-infected patients starting ARV therapy in Mexico, the crude mortality ratio of patients with IRIS when compared with patients without was 3.02 ( $p = 0.03$ ), but after adjusting for the relevant confounders it decreased to 2.3 ( $p = 0.09$ ). We also observed that the effect of IRIS on mortality differed depending on whether the patient had a previously diagnosed OI or not.

The cumulative incidence of IRIS for this period was 27%, which is consistent with previously published cohort studies in which IRIS has been shown to present during the first weeks after beginning ARV therapy in about 12–45% of patients who start treatment at very low CD4+ cell counts.<sup>5,8,17,18</sup> The variability in the occurrence of IRIS may be explained by the local prevalence of the involved pathogens, the level of immune suppression when starting HAART in different settings, and the pathogenicity of each microbe,<sup>7,17–20</sup> but also by the case-definition used, as will be discussed further.

The presence of an OI before starting HAART, a low weight or body mass index, and a low CD4+ T cell count have previously been identified as risk factors for IRIS,<sup>21–23</sup> and these same factors have

**Table 4**  
Mortality and hospitalization rates for HIV-infected patients starting HAART according to IRIS

	Total (N=390) (100%)	IRIS (n=107) (27.4%)	No IRIS (n=283) (72.6%)	RR <sup>a</sup> (95% CI)	p-Value
Deaths	19 (4.9)	8 (7.5)	11 (3.9)	2.3 <sup>b</sup> (0.9–5.9)	0.09
Hospitalizations	59 (15.1)	34 (31.8)	25 (8.8)	3.2 <sup>c</sup> (1.9–5.6)	<0.001

HAART, highly active antiretroviral therapy; IRIS, immune reconstitution inflammatory syndrome; RR, relative risk; CI, confidence interval.

<sup>a</sup> RR estimated by Cox regression for mortality and Poisson regression for hospitalization.

<sup>b</sup> Adjusted for age, CD4+ T cell count, presence of opportunistic infections prior to HAART initiation, and baseline weight.

<sup>c</sup> Adjusted for baseline CD4+ T cell count, presence of opportunistic infections prior to HAART initiation, and baseline weight.

**Table 5**  
Risk of mortality by IRIS status according to the presence of opportunistic infections before the initiation of HAART<sup>a</sup>

Group	Mortality rate ratio <sup>b</sup> (95% CI)	p-Value
No OIs before HAART		
No IRIS	1	–
IRIS	1.2 (0.1–10.8)	0.86
OIs before HAART		
No IRIS	2.9 (0.9–10.1)	0.09
IRIS	8.1 (2.4–27.5)	0.001

IRIS, immune reconstitution inflammatory syndrome; HAART, highly active antiretroviral therapy; CI, confidence interval; OI, opportunistic infection.

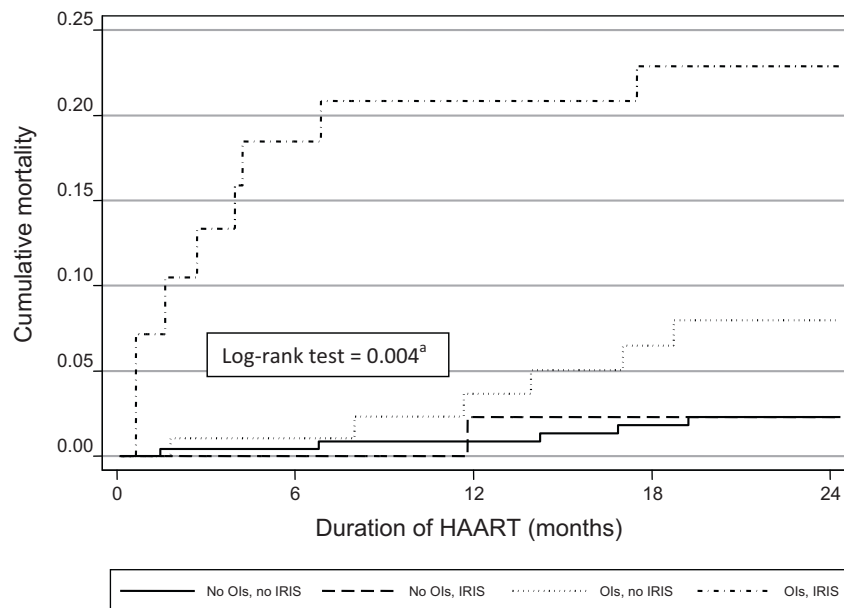
<sup>a</sup> Adjusted for age, weight and CD4+ T cell counts at baseline.

<sup>b</sup> Rate ratios derived from hazard ratios (HR), estimated using a Cox Proportional Hazards model. HR and p-values by stratum were estimated using an interaction term between the binary variables for opportunistic infections and IRIS.

also been found to be predictors of death in HIV-infected patients in general.<sup>24,25</sup> In this study, we controlled the confounding effects of these factors using a multivariate Cox regression model. Our results show that although patients with IRIS indeed have a higher mortality than patients without it, the impact that IRIS may have over mortality in patients receiving HAART is strongly confounded by the fact that patients who develop IRIS are usually those who start HAART at more advanced stages of the disease. The presence of a previous OI appears to be the single most important factor contributing to the increased mortality in patients who developed IRIS in this cohort, as a 31% excess mortality can be attributed to this variable.<sup>26</sup> Previous studies have found conflicting results

regarding the effect of IRIS on mortality. Lawn et al. found a high mortality rate among patients who developed IRIS in a cohort receiving medical care in South Africa.<sup>12</sup> In contrast, in a recently published cohort analysis of 559 ARV therapy-naïve patients starting HAART in a tertiary care center in Uganda, only four out of 99 deaths were directly attributed to IRIS.<sup>9</sup> Similar results were observed in the single cohort study that has prospectively evaluated the contribution of IRIS to mortality.<sup>21</sup> The IRIS cumulative incidence in an tertiary referral center in South Africa was 10% at 6 months of follow-up, with a very low mortality rate attributed to IRIS (two deaths among 44 cases of IRIS) – although it is worth mentioning that most of their cases were deemed to be ‘mild’.

We believe the most plausible explanations for the discrepancies across data are: first, that the studies reporting a higher mortality among patients suffering IRIS have their analysis restricted to specific pathogens and conditions,<sup>8,12</sup> which may in fact show a more severe presentation or may pose greater difficulties for their treatment and medical care (e.g., tuberculosis and cryptococcal meningitis). A recently published systematic review of IRIS seems to confirm this explanation.<sup>27</sup> In addition, the different methods used in the analysis of the information may also account for disparities in the results. For instance, in the two cohort studies carried out in South Africa, particular causes of death were determined and the IRIS-specific mortality was assessed, in contrast to ours, where the overall mortality rate and the mortality risk associated with IRIS were evaluated. Although it is clear that deaths in our study cannot be attributed directly to IRIS events – and we acknowledge this as a limitation of our study – the



**Figure 1.** Kaplan–Meier survival curves comparing the adjusted survival time of patients who developed IRIS with patients without IRIS, according to the presence of opportunistic infections prior to HAART initiation (IRIS, immune reconstitution inflammatory syndrome; OIs, opportunistic infections diagnosed before HAART initiation; HAART, highly-active antiretroviral therapy). Adjusted for age, baseline CD4+ T cell count, and baseline weight. <sup>a</sup>Log-rank test: tests the equality of survivor functions across the four groups.

increased risk of death among those who had IRIS, and particularly for those who had a previous OI and subsequently developed IRIS, deserves further consideration.

We also observed a more than three-fold increase in the risk of hospitalization among patients who developed IRIS after adjusting for the effect of other risk factors independently associated with hospitalization, confirming previous findings that IRIS events have a considerable impact on morbidity in HIV-infected patients recently started on HAART.<sup>7,18</sup>

We recognize that there are several limitations in our study. First, the absence of a complete understanding of the IRIS decreases the reliability of case definitions, so we can expect some degree of misclassification bias.<sup>10</sup> For instance, a recent comparison of expert opinion vs. the two published case definitions for IRIS<sup>28,29</sup> yielded different agreement with expert opinion. Moreover, the sensitivity of each definition varied according to the type of IRIS event,<sup>30</sup> despite the fact that both definitions use similar criteria. The relatively similar incidence rate of IRIS seen in prospective cohorts and in other studies, regardless of the case definition used, is reassuring with regard to the potential sources of error in our own approach. Another constraint of this study is that we registered all-cause mortality, so we cannot claim that the increased mortality in these patients was directly attributable to IRIS. In addition, the general applicability of our findings may be restricted, as our clinic operates in a tertiary-level care center in a geographically accessible, urban setting. However, we think that the mortality directly attributed and/or associated to IRIS would be similar, and more likely higher, in care centers with scarcer resources that may have more limited diagnostic and therapeutic options.

Finally, the strong association between the presence of an OI before HAART, the subsequent development of IRIS, and an increased risk over 2 years of mortality address an important issue observed in patients receiving HAART in developing countries, independently of the direct cause of death. Despite the fact that patients receiving HAART in developing countries have similar proportions of viral load suppression and immune reconstitution (as measured by increase in CD4+ T cell counts) as their counterparts in more advanced nations, it is also true that an excess of deaths occurs within the first few months of therapy among patients receiving HAART in resource-poor countries.<sup>11,31,32</sup> The identification of individuals at higher risk of death after beginning ARV therapy in developing countries, such as those who develop IRIS, may help design different health care strategies to reduce this risk. Our study highlights the necessity of expanding access to HIV voluntary counseling and testing, of raising awareness about AIDS and HIV-testing to increase early diagnosis, and of the need to start treatment earlier,<sup>33</sup> as starting patients on HAART at higher CD4 counts would likely reduce the incidence of IRIS. It also provides rational support for the evaluation of novel strategies to prevent and treat IRIS in those HIV-infected patients at risk for this condition.

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