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Research article

Virologic, clinical and immunologic responses following failure of first-line antiretroviral therapy in Haiti

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Abstract

Background: Since HIV-1 RNA (viral load) testing is not routinely available in Haiti, HIV-infected patients receiving antiretroviral therapy (ART) are monitored using the World Health Organization (WHO) clinical and/or immunologic criteria. Data on survival and treatment outcomes for HIV-1 infected patients who meet criteria for ART failure are limited. We conducted a retrospective study to compare survival rates for patients who experienced failure on first-line ART by clinical and/or immunologic criteria and switched to second-line ART vs. those who failed but did not switch.

Methods: Patients receiving first-line ART at the GHESKIO Center in Port-au-Prince, Haiti, who met WHO clinical and immunologic criteria for failure were identified. Survival and treatment outcomes were compared in patients who switched their ART regimen and those who did not. Cox regression analysis was used to determine predictors of mortality after failure of first-line ART.

Results: Of 3126 patients who initiated ART at the GHESKIO Center between 1 March 2003 and 31 July 2008, 482 (15%) met WHO immunologic and/or clinical criteria for failure. Among those, 195 (41%) switched to second-line ART and 287 (59%) did not. According to Kaplan–Meier survival analysis, the probability of survival to 12 months after failure of first-line ART was 93% for patients who switched to second-line ART after failure and 88% for patients who did not switch. Predictors of mortality after failure of first-line ART were weight in the lowest quartile for sex, CD4 T cell count ≤ 100, adherence $< 90\%$ at the time of failure and not switching to second-line ART.

Conclusions: Patients who failed first-line ART based on clinical and/or immunologic criteria and did not switch to second-line therapy faced a higher mortality than those who switched after failure. To decrease mortality, interventions to identify patients in whom ART may be failing earlier are needed urgently. In addition, there is a major need to optimize second-line antiretroviral regimens for improved potency, lower toxicity and greater convenience for patients.

Keywords: Antiretroviral therapy; second-line therapy; virologic failure; mortality; adherence.

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Introduction

Nearly 7 million HIV-infected patients in resource-poor settings have initiated antiretroviral therapy (ART) since 2003, and as many as 1 million of these patients may have virologic failure with ongoing HIV replication [1–3]. Due to a lack of HIV-1 RNA monitoring in resource-poor settings, these patients will continue on first-line ART until virologic failure progresses to a 50% decrease in CD4 T cell count (immunologic failure) or the recurrence of symptomatic HIV disease (clinical failure). Even then, clinicians may delay switching to second-line therapy, due to the limited availability of second-line medications and the poor specificity of CD4 T cell counts and clinical symptoms for predicting virologic failure [4–7].

In one analysis, computer modelling of ART outcomes in resource-limited settings suggested that HIV RNA testing would not significantly impact survival compared with either CD4 testing or clinical monitoring [8]. However, several studies suggest that where HIV RNA testing is not routinely used, switching to second-line ART is delayed and patients face a high early mortality after switching. A study from Uganda (Home-Based Care Clinical Trial) indicated that clinical and/or immunologic criteria have poor predictive values for ART failure and are associated with AIDS-defining events and increased mortality [9]. Furthermore, the DART trial showed that CD4 testing led to small but significant decreases in the proportion of person-years spent with low CD4-cell counts and lower rates of HIV disease progression and death from the third year on ART as compared with clinical monitoring [10].

In light of these results, we conducted a retrospective study to determine survival rates and other outcomes for patients who switched to second-line ART vs. those who did not after experiencing clinical and/or immunologic failure to first-line ART in Haiti.

Methods

Study design

We conducted a retrospective study of survival rates and other clinical outcomes for patients receiving first-line ART who met clinical and/or immunologic criteria for failure. We compared outcomes for patients who switched to second-line ART or not. As HIV-1 RNA testing is not routinely available for patient monitoring in Haiti, plasma specimens were collected and banked when patients had CD4 T cell counts measured. In this study, we tested banked plasma from these patients for HIV-1 RNA levels and HIV-1 drug resistance by 12 months after initiation of second-line therapy. The institutional review boards at GHESKIO and at Weill Cornell Medical College approved this study.

Setting

Patients received care and were followed up at the Clinic of the Center of the Haitian Group for the Study of Kaposi's Sarcoma and Opportunistic Infections (GHESKIO) in Port-au-Prince, Haiti. Since 1983, the GHESKIO clinic has provided free HIV counselling, testing, prevention and AIDS care for the Port-au-Prince population [11]. With support of the Global Fund Against AIDS, Tuberculosis, and Malaria and the President's Emergency Plan for AIDS Relief (PEPFAR), GHESKIO provides antiretroviral therapy to HIV-infected patients following Haitian National and WHO guidelines [12,13].

First-line therapy for adults and adolescents consisted of zidovudine, lamivudine and efavirenz. Women of reproductive potential received nevirapine in place of efavirenz. Treatment adherence was encouraged through counselling, home visits, pill counts and social support, which included psychological support, food rations, transportation fees and cell phone cards.

After 6 months of first-line ART, patients were evaluated for ART failure with clinic visits every month and CD4 T cell count every 6 months. The WHO defines ART failure as (1) CD4 count below 100 cells/mm^3 after 6 months of therapy; (2) a 50% decrease in the CD4 T cell count; (3) a return of the CD4 T cell count to the pretherapy CD4 baseline or lower; or (4) the development of Stage 3 or 4 HIV events during ART [12–14].

The decision to switch to second-line ART was taken by the clinical team based on the clinical and/or immunologic criteria outlined above. Patients who failed first-line ART switched to second line therapy, which included two new reverse transcriptase inhibitors and a protease inhibitor (lopinavir/ritonavir, indinavir, or nelfinavir).

Population

Inclusion criteria were HIV-1 infected, aged 13 years and older, initiated first-line ART between 1 March 2003 and 31 July 2008 at the GHESKIO Centers, treatment naïve at initiation of first-line therapy, received at least 6 months of first-line ART, subsequently met CD4 and/or clinical criteria for first-line ART failure by WHO criteria. Patients enrolled in clinical trials such as ACTG 5175 and the CIPRA HT001 and who had HIV-1 RNA tests performed for research purposes were excluded from this study.

Clinical measurements

Clinical data through 31 August 2009 were analyzed. Data were retrieved from an electronic medical record and from patient charts. The primary clinical outcome was mortality after failure of first-line ART. Adherence to first-line ART and second-line ART was quantified by examining pharmacy refill records and comparing the actual number of medication refill days during the follow-up period with the expected number of days. An ''adherent'' patient was defined as one whose refill days of antiretroviral medication were at least 90% of the expected number of days. This adherence measure is correlated with virologic outcomes and death [15,16]. We also report treatment-limiting drug toxicities (in the opinion of the physician) that prompted a change in second-line antiretroviral medication.

Laboratory measurements

We recorded patients' CD4 T cell counts, which were performed every 6 months during follow-up, and when clinically indicated. CD4 T cell counts were performed by flow cytometry (Becton Dickinson).

For surveillance purposes, the GHESKIO Center routinely banks a 2 ml plasma sample at -80°C when patients on ART have a CD4 T cell count performed. We tested retrospectively and, in batch, banked plasma samples for HIV-1 RNA levels. We tested samples at the time of second-line ART initiation and 6 to 12 months afterwards. The NucliSens EasyQ HIV-1 PCR Test, v. 1.2 (BioMérieux, Lyon, France), with a lower limit of detection of 50 copies of HIV-1 RNA per millilitre, was used.

We performed HIV-1 drug resistance testing on 6- to 12-month plasma samples that had >1000 HIV-1 RNA copies/ml. The HIV-1 polymerase and protease genes were amplified by PCR and then sequenced on an automated system (Applied Biosystems, Bedford, MA, USA). Mutations at positions in the polymerase and protease genes associated with antiretroviral resistance by the International AIDS Society-USA Drug Resistance Mutations Group were noted [17]. Results of HIV-1 RNA testing and drug resistance testing were provided to primary care providers.

Statistical analysis

The data were analyzed with STATA software version 10 (STATA Corporation, College Station, TX, USA). Proportions were compared by the chi-square test with Yates' correction, for expected CD4 T cell values of <5, by Fisher's exact test. Means and medians were compared by Student's *t*-test and the Wilcoxon rank-sum test, respectively. Kaplan–Meier survival analyses were used to estimate the time from the initiation of second-line ART to death. For patients who did not reach the endpoint, the data were censored at the date of the last visit. The first 6 months of person-time were excluded in the calculation of follow-up time for patients who died after 6 months of first-line therapy.

Cox multivariate analysis was performed to identify predictors of mortality and the association is reported as adjusted hazards ratio (HR). Logistic regression analysis was used to determine predictors of detectable HIV-1 RNA at 6 months and associations reported as adjusted odds ratios (OR). Factors that were considered in the multivariate model

included age, sex, CD4 T cell counts, weight, adherence on first-line ART and adherence on second-line ART. Two-sided hypotheses and tests were adopted for all statistical inferences.

Results

Study population

Between 1 March 2003 and 31 July 2008, 3615 HIV-1 infected patients 13 years and older were initiated on first-line ART at the GHESKIO clinic in Port-au-Prince, Haiti, and 3126 patients were alive and in active follow-up 6 months after initiating first-line therapy and were at risk for ART failure.

Of the 3126, 161 died after at least 6 months of first-line therapy but prior to failure or starting second-line ART. Of the 3126, 482 patients satisfied clinical and/or immunologic criteria for ART failure for a failure rate of 4.8 per 100 person-years. Of these 482 patients, 195 (40%) subsequently switched to second-line ART whereas 287 (60%) did not.

The characteristics of the 482 patients who failed first-line ART, according to whether they switched to second-line ART or not, are summarized in Table 1. The two groups were similar in age, sex, weight, first-line antiretroviral regimen initiated and percentage with tuberculosis at baseline but differed significantly with respect to the median CD4 T cell count (82 vs. 174 cells/mm^3, $p<0.001$), percentage of patients at WHO Stages 3 and 4 HIV disease (47% vs. 32%, $p<0.001$) and time from initiation of first-line ART to failure (19.3 vs. 12.0 months, $p=0.002$). The median time between the first sign or symptom of first-line ART failure and switching to second-line ART was 7 months (interquartile range (IQR) 2 to 15 months).

In patients who switched to second-line ART, the protease inhibitors that were used in the second-line regimens included lopinavir/ritonavir in 146 (75%) patients, indinavir in 34 (17%) patients and nelfinavir in 15 (8%) patients. The nucleoside reverse transcriptase inhibitors used in second-line regimens included abacavir in 135 (69%) patients, tenofovir in 109 (56%) patients, didanosine in 61 (31%) patients, lamivudine in 51 (28%) patients, stavudine in 26 (13%) patients and zidovudine in 8 (4%) patients. The most common second-line antiretroviral regimens used were tenofovir–abacavir–lopinavir/ritonavir (70 patients, 36%), didanosine–abacavir–lopinavir/ritonavir (41 patients, 21%) and tenofovir–lamivudine–lopinavir/ritonavir (23 patients, 12%).

Status at the time of analysis and adherence to ART

At the time of analysis in August 2009, 157 (81%) of the 195 patients who switched to second-line ART were still in active follow-up, 7 (3%) had been lost to follow-up and 31 (16%) had died. Among the 282 patients who failed but did not switch 216 (75%) were in active follow-up, 22 (11%) had been lost to follow-up and 49 (25%) had died. The median follow-up after failure of first-line ART was 22 months (IQR, 12 to 41 months).

Adherence to ART was measured by examining pharmacy refill records and comparing the actual number of medication refill days in the first 6 months with the expected number.

Table 1. Characteristics of the patients at the time of initiation of first-line antiretroviral therapy (ART)

Characteristic	Switched to second line ART ($n=195$)	Did not switch to second line ART ($n=287$)	p-Value
Female sex – no. (%)	104 (53)	150 (52)	0.818
Age			
Median (IQR) years	37 (28 to 44)	37 (30 to 44)	0.904
Weight (kg)			
Men			
Median (IQR)	58 (49 to 68)	56 (47 to 65)	0.164
Women			
Median (IQR)	49 (39 to 60)	49 (43 to 57)	0.907
CD4 T cell count (per mm^3)			
Median (IQR)	82 (31 to 178)	174 (89 to 273)	<0.001
First-line antiretroviral regimen – no. (%)			0.064
Zidovudine, lamivudine, efavirenz	97 (50)	139 (48)	
Zidovudine, lamivudine, nevirapine	61 (31)	75 (26)	
Stavudine, lamivudine, efavirenz	7 (4)	12 (4)	
Stavudine, lamivudine, nevirapine	3 (2)	16 (6)	
Zidovudine, lamivudine, abacavir	13 (7)	11 (4)	
Others	14 (7)	34 (12)	
WHO Stage 3/Stage 4 – no. (%)	91 (47)	92 (32)	<0.001
Tuberculosis – no. (%)	16 (8)	17 (6)	0.330
Time from start of first-line ART to failure (months)			
Median	19.3	12.0	0.002
IQR	10.2 to 29.9	6.0 to 29.9	

IQR, interquartile range; WHO, World Health Organization.

Adherence data were available for 471 (97%) of the patients. Two hundred and fifty-four (54%) had taken 90% or more of the doses of medication prescribed for them until the time of failure. Patients with less than 90% adherence to first-line ART were less likely to be switched to second-line ART than those with 90% or more adherence level (OR, 0.60; 95% CI, 0.41 to 0.87; $p=0.007$). Sixty-three percent of patients who switched to second-line ART had taken 90% or more of the doses prescribed to them during the follow-up period. Before switching to second-line ART, 111 (57%) of the patients had taken 90% or more of the doses of antiretroviral prescribed.

Mortality

There were 80 deaths among the 482 patients (17%) who failed first-line ART during the follow up period: 31 deaths occurred among the 195 patients (16%) who switched to second-line ART and 49 deaths among the 287 patients (17%) who did not switch ($p=0.065$ by the log-rank test). Most frequent causes of death in both groups were wasting syndrome in 48 patients (60%), pulmonary tuberculosis in 15 patients (19%) and sepsis syndrome in 6 patients (8%). Among patients who died after switching to second-line ART, 13 (42%) deaths occurred within 12 months of switching.

According to Kaplan–Meier survival analysis, the probability of survival to 12 months after first-line ART failure was 93% (95% CI, 88% to 96%) for patients who switched to second-line ART after failure and 88% (95% CI, 83% to 91%) for patients who did not switch (Figure 1). The unadjusted HR for the risk of death among those who did not switch to second-line ART as compared with those who switched to second-line ART was 1.5 (95% CI, 1.0 to 2.4).

According to Cox regression analysis, predictors of mortality after failure of first-line ART were weight in the lowest quartile for sex (HR, 3.3; 95% CI, 1.9 to 5.6; $p<0.001$), CD4 T cell count ≤ 100 (HR, 3.2; 95% CI, 1.9 to 5.3; $p<0.001$), adherence $<90\%$ at the time of failure (HR, 1.6; 95% CI, 1.0 to 2.7; $p=0.074$) and not switching to second-line ART (HR, 2.1; 95% CI, 1.2 to 3.4; $p=0.007$).

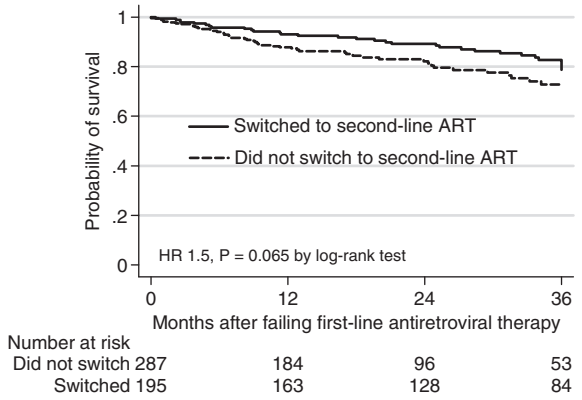

Figure 1. Kaplan–Meier survival estimates for 482 patients who failed first-line ART according to switching status.

Immunologic and virologic response in patients who switched to second-line ART

At the time of first-line ART failure, the median CD4 T cell count in patients who switched to second-line ART was 102 cells/mm^3 (IQR, 54 to 185), whereas that of the patients who did not switch was 144 cells/mm^3 (IQR, 86 to 258). The median CD4 T cell in patients who switched to second-line ART increased to 241 cells/mm^3 (IQR, 180 to 342) within 6 months after switching, whereas that of patients who did not switch increased to 192 cells/mm^3 (IQR, 97 to 328) after 6 months of remaining on first-line ART.

HIV-1 RNA levels were performed in 149 patients with available plasma specimens collected and preserved between 6 and 12 months (median, 9.5 months; IQR, 6.1 to 14.6) after switching to second-line ART. Eighty-six of the patients (58%) achieved virologic suppression (HIV-1 RNA <400 copies/ml) after switching to a second-line regimen. Of these patients, 65 (44%) had HIV-1 RNA <50 copies/ml. Among the 63 patients with HIV-1 RNA >400 copies/ml, the median log HIV-1 RNA was 4.34 (IQR, 3.69 to 5.13). Six patients (10%) had HIV-1 RNA between 400 and 1000 copies/ml, 16 (25%) had between 1000 and 10,000 copies/ml and 41 (65%) had over 10,000 copies/ml. Patients receiving ritonavir-boosted lopinavir were more likely to achieve HIV-1 RNA <50 copies/ml than those receiving unboosted nelfinavir or indinavir (OR, 3.3; 95% CI, 1.3 to 8.3; $p=0.01$).

Adherence less than 90% to second-line ART was the single factor associated with plasma HIV-1 RNA level >1000 copies/ml in multivariate analysis (OR, 7.0; 95% CI, 3.3 to 15.1; $p<0.001$. Analyses at different HIV-1 RNA thresholds (<50 copies/ml, <400 copies/ml) gave similar results.

HIV-1 genotyping results after switching to second-line ART

HIV-1 genotyping was performed for patients with HIV-1 RNA >1000 copies/ml between 6 and 12 months on second-line ART. Results were obtained in 43 patients (Table 2). Thirty-seven of the 43 patients (86%) were found to have at least one drug resistance mutation. Thirty (70%) patients were found to have resistance mutations to the non-nucleoside reverse transcriptase inhibitors. Fourteen (33%) patients were found to have at least two thymidine analogue mutations. The frequencies of specific mutations are presented under Table 2.

Drug-limiting toxicity in patients who switched to second-line ART

Of the 195 patients who switched to second-line ART, 21 (11%) patients required a change in antiretroviral medication. Twelve patients (6%) had a change in the protease inhibitor, whereas 9 (5%) had a change in the nucleoside analogue medication. Among patients requiring a change in protease inhibitors, the reasons for the changes were as follows: five patients required a change in indinavir for nephrolithiasis. Five patients required a change in nelfinavir: three due to diarrhoea and two due to rash. Finally, two patients required a change in lopinavir/ritonavir due to diarrhoea.

Among the nine patients requiring a change in nucleoside analogue, six patients required a change in didanosine (four due to peripheral neuropathy and two due to hepatoxicity), two patients required a change in tenofovir

Table 2. Resistance mutations in 43 patients with HIV-1 RNA level >1000 copies/ml while receiving second-line ART

HIV-1 mutation	Number of patients *n*=43 (%)
Any drug resistance mutation	37 (86)
Mutation conferring resistance to non-nucleoside reverse transcriptase inhibitors (NNRTI)[a]	30 (70)
Reverse transcriptase M184V mutation	23 (53)
M184V and NNRTI mutations	12 (28)
Any thymidine analogue mutation (TAM)[b]	16 (37)
Two or more TAMs	14 (33)
Any PI resistance mutation[c]	17 (40)
At least one major PI resistance mutation	4 (9)

[a]Most frequent NNRTI mutations were: K103N (18), Y181C (7) and G190S (4). Eighteen (60%) patients had more than 2 NNRTI mutations; [b]TAMs were: M41L (8), D67N (5), K70R (9), L210W (2), T215Y/F (12), K219Q/E (8); [c]Major PI mutations were: D30N (1), V32I (1), M46I (2), I47V (1), V82A (3), L90M (1). Minor PI mutations were: L10F/I/V (12), L33F (1), F53L (1), I54V (2), A71VT (9), N88D/S (2), I85V (1).

due to nephrotoxicity and one patient required a change in abacavir due to rash.

Discussion

Our study examined mortality rates and other outcomes in patients who failed first-line ART in Haiti. In the absence of HIV-1 RNA testing, physicians in resource-limited countries wait until patients progress to a decrease in CD4 T cell counts (immunologic failure) or to overt signs and symptoms of HIV disease (clinical failure) before switching to second-line ART. In our study from Haiti, 16% of patients who failed first-line ART based upon WHO clinical and/or immunologic criteria died in the first year after initiating second-line ART. Mortality was associated with weight in the lowest quartile for sex, low CD4 T cell count and not switching to second-line ART. The mortality observed in our study likely resulted from a delay in recognition of virologic failure due to the lack of HIV-1 RNA monitoring. Our study showed a trend toward higher mortality in patients who did not switch compared to those who switched.

The high early mortality rate seen in our cohort initiating second-line therapy is similar to the high early mortality rate documented for HIV-1 infected patients with advanced AIDS initiating first-line ART in resource-poor areas [18,19]. Because of the low sensitivity and specificity of the WHO immunologic and clinical criteria for ART failure, healthcare providers likely waited until patients showed overt HIV symptoms or low CD4 T cell counts before switching to second-line regimens [20]. This is evident in the delay observed in our study (median time from failure to switching 7.0 months) and the low median CD4 T cell counts at the time of failure. Patients who fail first-line therapy therefore must pass again through a period of advanced immune suppression and high-mortality risk in order to be recognized as treatment failures and initiated on second-line therapy. With this increased mortality, there is clearly a need for virologic monitoring for the early detection of ART failure and switching to second-line appropriately.

A number of studies have shown that CD4 counts are poor predictors of ART failure and have to be used cautiously in monitoring patients where viral load testing is not available [3,9]. The findings of the Home-Based AIDS Care (HBAC) study support our observation that early switching based on clinical/immunologic criteria in the absence of viral load may lead to mistaken and premature switching. Delayed switching if failure goes unrecognized may lead to higher mortality [21].

The Development of Antiretroviral Therapy (DART) trial in Africa showed that CD4 testing resulted in decreases in the proportion of person-years spent with low CD4-cell counts, higher rates of switching from the second year and lower rates of HIV disease progression and death beginning in the third year of ART [10]. These findings support our observation that patients who switched to second-line ART had higher survival compared to patients who did not switch. Thus, HIV-1 RNA testing may allow early identification of patients failing first-line ART and switching to second-line ART, which may help to reduce the high mortality for these patients.

Two recent randomized controlled trials on strategies for monitoring patients receiving ART in resource-limited settings showed that clinical or immunologic monitoring was not non-inferior to virologic monitoring [22,23]. Both studies indicated that close and regular monitoring of adherence, toxicity and tolerability are important to prevent ART failure. HIV-1 RNA testing would still be necessary to verify the effectiveness of ART and prevent the accumulation of drug resistance over time.

On second-line ART, the 15% mortality rate at 1 year in Haiti is three times higher than the 5% 1-year mortality rate reported in South Africa, where viral load monitoring is available [24]. The mortality rate detected in our study was also higher than the 4.4 deaths per 100 person years reported in a recent multicentre study conducted in Africa and Asia [25]. This study included only patients who had received at least 6 months of second-line therapy and therefore did not capture the early deaths seen in our study. Further, studies of second-line treatment outcomes may miss the deaths that occur after patients develop symptoms but before patients are switched to second-line therapy. In our cohort, 2.5% of patients on first-line therapy who meet clinical and/or immunologic criteria for failure die annually without switching to second-line drugs. Our data are similar to a report from Malawi, which also found high rates of early mortality in patients initiating second-line ART [26].

Drug adherence less than 90% was associated with death in patients failing first-line ART. In the present study, only 63% of the patients on second-line therapy took 90% or more of their doses prescribed. Improving adherence does not require expensive laboratory equipment or technology. Pharmacy refill records are a simple tool to identify non-adherent patients. We are developing systems so that non-adherent patients can be identified early in the course of first-line therapy and provided specialized counselling including

peer-group support and frequent contact with the clinic via cell phone. Access to second-line drugs with greater potency, less toxicity and more convenient dosing schedules for patients could improve adherence.

Virologic failure to second-line therapy does not appear to be due to ART drug resistance as few patients developed drug resistance to protease inhibitors. Rates of resistance to NRTI and NNRTI mutations observed in our study are similar to those observed in patients failing first-line antiretroviral therapy and only four patients were found to have at least one major protease inhibitor mutation [27,28]. Therefore, lopinavir/ritonavir regimen should be an effective second-line regimen, and suboptimal adherence appears to be the cause of virologic failure to second-line ART in our population.

Our study has several limitations: This was a retrospective study, not all eligible subjects were included and some patients did not have plasma specimens collected and stored for HIV-1 RNA testing. Further, this was a non-randomized study, and clinicians chose treatment strategies based on individual patient factors. Thus, we cannot exclude the possibility of selection bias.

Conclusions

In summary, we found a high mortality rate after failure of first-line ART in Haiti, associated with CD4 cell count <100 and a delay in switching to second-line ART. In resource-limited settings, ART failure in patients who are monitored by the WHO clinical and immunologic criteria is detected later, when patients have advanced immune suppression. As a result, patients who fail first-line therapy are at high risk for death. Cost-effective and simplified technologies need to be developed so that HIV-1 virologic monitoring can be implemented on a large scale in resource poor settings. Newer second-line antiretroviral drugs with greater potency, tolerability and convenience should be made available in these parts of the world for better adherence.

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Competing interests
RMG served as an ad-hoc consultant to Bristol-Myers, Gilead, Janssen, Merck and ViiV.

Authors' contributions
MC designed the study, carried out the genotyping studies, performed the data analysis and drafted the manuscript. PDL participated in the design of the study and performed the statistical analysis. PS and CG participated in the design of the study and collected clinical data. AA performed specimen collection. RMG contributed to the interpretation of the data and critically revised the manuscript for important intellectual content. WDJ, JWP and DWF provided important advice and insight into the analyses and writing of the paper. All authors have read and approved the final manuscript.

Abbreviations
ART, antiretroviral therapy; DART, development of antiretroviral therapy; HR, hazards ratio; IQR, interquartile range; OR, odds ratios.

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References

1. UNAIDS [Internet]. 2011 World AIDS Day report [cited December 2011]. Available from: http://www.unaids.org.
2. Hosseinipour MC, van Oosterhout JJ, Weigel R, Phiri S, Kamwendo D, Parkin N, et al. The public health approach to identify antiretroviral therapy failure: high-level nucleoside reverse transcriptase inhibitor resistance among Malawians failing first-line antiretroviral therapy. AIDS. 2009;23:1127–34.
3. Leger P, Charles M, Severe P, Riviere C, Pape J, Fitzgerald D. 5-year survival of patients with AIDS receiving antiretroviral therapy in Haiti. N Engl J Med. 2009;361:828–9.
4. Kantor R, Diero L, Delong A, Kamle L, Muyonga S, Mambo F, et al. Misclassification of first-line antiretroviral treatment failure based on immunological monitoring of HIV infection in resource-limited settings. Clin Infect Dis. 2009;49:454–62.
5. Mee P, Fielding KL, Charalambous S, Churchyard GJ, Grant AD. Evaluation of the WHO criteria for antiretroviral treatment failure among adults in South Africa. AIDS. 2008;22:1971–7.
6. Reynolds SJ, Nakigozi G, Newell K, Ndyanabo A, Galiwongo R, Boaz I, et al. Failure of immunologic criteria to appropriately identify antiretroviral treatment failure in Uganda. AIDS. 2009;23:697–700.
7. van Oosterhout JJ, Brown L, Weigel R, Kumwenda JJ, Mzinganjira D, Saukila N, et al. Diagnosis of antiretroviral therapy failure in Malawi: poor performance of clinical and immunological WHO criteria. Trop Med Int Health. 2009;14:856–61.
8. Phillips AN, Pillay D, Miners AH, Bennett DE, Gilks CF, Lundgren JD. Outcomes from monitoring of patients on antiretroviral therapy in resource-limited settings with viral load, CD4 cell count, or clinical observation alone: a computer simulation model. Lancet. 2008;371:1443–51.
9. Moore DM, Awor A, Downing R, Kaplan J, Montaner JS, Hancock J, et al. CD4+ T-cell count monitoring does not accurately identify HIV-infected adults with virologic failure receiving antiretroviral therapy. J Acquir Immune Defic Syndr. 2008;49:477–84.
10. DART Trial Team. Routine versus clinically driven laboratory monitoring of HIV antiretroviral therapy in Africa (DART): a randomised non-inferiority trial. Lancet. 2010;375:123–31.
11. Peck R, Fitzgerald DW, Liautaud B, Deschamps MM, Verdier RI, Beaulieu ME, et al. The feasibility, demand, and effect of integrating primary care services with HIV voluntary counseling and testing: evaluation of a 15-year experience in Haiti, 1985–2000. J Acquir Immune Defic Syndr. 2003;33:470–5.
12. Scaling up antiretroviral therapy in resource-limited settings. Geneva: World Health Organization; 2004.
13. Severe P, Leger P, Charles M, Noel F, Bonhomme G, Bois G, et al. Antiretroviral therapy in a thousand patients with AIDS in Haiti. N Engl J Med. 2005;353:2325–34.
14. World Health Organization. Antiretroviral therapy for HIV infection in adults and adolescents: recommendations for a public health approach. Geneva: WHO; 2010 revision.
15. Grossberg R, Zhang Y, Gross R. A time-to-prescription-refill measure of antiretroviral adherence predicted changes in viral load in HIV. J Clin Epidemiol. 2004;57:1107–10.
16. Kitahata MM, Reed SD, Dillingham PW, Van Rompaey SE, Young AA, Harrington RD, et al. Pharmacy-based assessment of adherence to HAART predicts virologic and immunologic treatment response and clinical progression to AIDS and death. Int J STD AIDS. 2004;15:803–10.
17. Johnson VA, Brun-Vezinet F, Clotet B, Gunthard HF, Kuritzkes DR, Pillay D, et al. Update of the drug resistance mutations in HIV-1: December 2009. Top HIV Med. 2009;17:138–45.
18. Braitstein P, Brinkhof MW, Dabis F, Schechter M, Boulle A, Miotti P, et al. Antiretroviral Therapy in Lower Income Countries (ART-LINC) Collaboration; ART Cohort Collaboration (ART-CC) groups. Mortality of HIV-1-infected patients in the first year of antiretroviral therapy: comparison between low-income and high-income countries. Lancet. 2006;367:817–24.
19. Lawn SD, Myer L, Orrell C, Bekker LG, Wood R. Early mortality among adults accessing a community-based antiretroviral service in South Africa: implications for programme design. AIDS. 2005;19:2141–8.

20. Keiser O, Tweya H, Boulle A, Braitstein P, Schecter M, Brinkhof MW, et al. Switching to second-line antiretroviral therapy in resource-limited settings: comparison of programmes with and without viral load monitoring. AIDS. 2009;23:1867–74.
21. Gsponer T, Petersen M, Egger M, Phiri S, Maathuis MH, Boulle A, et al. The causal effect of switching to second-line ART in programmes without access to routine viral load monitoring. AIDS. 2011;26:57–65.
22. Laurent C, Kouanfack C, Laborde-Balen G, Aghokeng AF, Mbougua JB, Boyer S, et al.; Stratall ANRS 12110/ESTHER Study Group. Monitoring of HIV viral loads, CD4 cell counts, and clinical assessments versus clinical monitoring alone for antiretroviral therapy in rural district hospitals in Cameroon (Stratall ANRS 12110/ESTHER): a randomised non-inferiority trial. Lancet Infect Dis. 2011;11:825–33.
23. Jourdain G, Ngo-Giang-Huong N, Le Coeur S, Traisaithit P, Barbier S, Techapornroong M, et al.; PHPT-3 Study Group [Internet]. PHPT-3: a randomized clinical trial comparing CD4 vs viral load ART monitoring/switching strategies in Thailand. 18th Conference on Retroviruses and Opportunistic Infections, Paper # 44 [cited May 2012]. Available from: http://www.retroconference.org/2011/Abstracts/41399.htm.
24. Fox MP, Ive P, Long L, Maskew M, Sanne I. High rates of survival, immune reconstitution and virologic suppression on second-line antiretroviral therapy in South Africa. J Acquir Immune Defic Syndr. 2009;53:500–6.
25. Pujades-Rodriguez M, Balkan S, Arnould L, Brinkhof MA, Calmy A. Treatment failure and mortality factors in patients receiving second-line HIV therapy in resource-limited countries. JAMA. 2010;304:303–12.
26. Hosseinipour MC, Kumwenda JJ, Weigel R, Brown LB, Mzinganjira D, Mhango B, et al. Second-line treatment in the Malawi antiretroviral programme: high early mortality, but good outcomes in survivors, despite extensive drug resistance at baseline. HIV Med. 2010;11:510–8.
27. Barth RE, Wensing AM, Tempelman HA, Moraba R, Schuurman R, Hoepelman AI. Rapid accumulation of nonnucleoside reverse transcriptase inhibitor-associated resistance: evidence of transmitted resistance in rural South Africa. AIDS. 2008;22:2210–2.
28. Charles M, Noel F, Leger P, Severe P, Riviere C, Beauharnais CA, et al. Survival, plasma HIV-1 RNA concentrations and drug resistance in HIV-1-infected Haitian adolescents and young adults on antiretrovirals. Bull World Health Organ. 2008;86:970–7.