Four-Year Treatment Outcomes of Adult Patients Enrolled in Mozambique's Rapidly Expanding Antiretroviral Therapy Program

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Abstract

Background: In Mozambique during 2004–2007 numbers of adult patients (≥15 years old) enrolled on antiretroviral therapy (ART) increased about 16-fold, from <5,000 to 79,500. All ART patients were eligible for co-trimoxazole. ART program outcomes, and determinants of outcomes, have not yet been reported.

Methodology/Principal Findings: In a retrospective cohort study, we investigated rates of mortality, attrition (death, loss to follow-up, or treatment cessation), immunologic treatment failure, and regimen-switch, as well as determinants of selected outcomes, among a nationally representative sample of 2,596 adults initiating ART during 2004–2007. At ART initiation, median age of patients was 34 and 62% were female. Malnutrition and advanced disease were common; 18% of patients weighed <45 kilograms, and 15% were WHO stage IV. Median baseline $CD4^+$ T-cell count was 153/μL and was lower for males than females (139/μL vs. 159/μL, p<0.01). Stavudine, lamivudine, and nevirapine or efavirenz were prescribed to 88% of patients; only 31% were prescribed co-trimoxazole. Mortality and attrition rates were 3.4 deaths and 19.8 attritions per 100 patient-years overall, and 12.9 deaths and 57.2 attritions per 100 patient-years in the first 90 days. Predictors of attrition included male sex [adjusted hazard ratio (AHR) 1.5; 95% confidence interval (CI), 1.3–1.8], weight <45 kg (AHR 2.1; 95% CI, 1.6–2.9, reference group >60 kg), WHO stage IV (AHR 1.7; 95% CI, 1.3–2.4, reference group WHO stage I/II), lack of co-trimoxazole prescription (AHR 1.4; 95% CI, 1.0–1.8), and later calendar year of ART initiation (AHR 1.5; 95% CI, 1.2–1.8). Rates of immunologic treatment failure and regimen-switch were 14.0 and 0.6 events per 100-patient years, respectively.

Conclusions: ART initiation at earlier disease stages and scale-up of co-trimoxazole among ART patients could improve outcomes. Research to determine reasons for low regimen-switch rates and increasing rates of attrition during program expansion is needed.

Introduction

Globally, during 2004–2009, the number of HIV-infected patients enrolled on antiretroviral therapy (ART) increased more than 10-fold, from less than 400,000 to more than five million, with most new ART patients enrolled in sub-Saharan Africa [1]. Reporting treatment outcomes of patients enrolled in ART programs is important to demonstrate program effectiveness and justify continued funding, while assessment of factors associated with outcomes can help to identify opportunities for program improvement [2,3]. Although many sub-national ART programs in sub-Saharan Africa have reported their treatment experience [4–12], only a few studies can be considered nationally representative due to the large cohort size [6,12], or sampling design [13].

In Mozambique, where about 1.6 million people are HIV-infected and about 473,000 need ART [14], numbers of adult patients (≥15 years old) enrolled on ART increased about 16-fold from less than 5,000 to 79,500 during 2004–2007 [15]. We conducted a retrospective, cohort study among a nationally representative sample of adult ART patients starting ART during 2004–2007, to describe mortality rates, attrition rates (numbers of patients who died, were lost to follow-up [LTFU], or stopped ART per 100 person-years), immunologic treatment failure rates, and determinants of attrition and treatment failure. Secondary treatment outcomes of interest were $CD4^+$ T-cell (CD4) count response and weight gain. Regimen-switch rates were measured to allow comparison with rates reported from other programs.

Methods

Ethics Approval

This study was approved by the Institutional Review Board (IRB) of the United States Centers for Disease Control and Prevention (CDC) and the Mozambican Ethics Review Committee (*Ministerio da Saude Comite Nacional de Bioetica para a Saude*). Both review boards approved the consent procedures. Patient informed consent was not required as only routine, anonymous, operational monitoring data were collected and analyzed.

Eligibility for ART during 2004–2007

During 2004–2007, patients diagnosed with World Health Organization (WHO) stage IV disease, stage III disease with CD4 counts <350/μL, or stage I or II disease with CD4 counts <200/μL, were eligible for ART [16]. Prescription of trimethoprim-sulfamethoxazole combination, or co-trimoxazole (CTX), was indicated for all ART patients. First-line ART regimens included two nucleoside reverse-transcriptase inhibitors (NRTI) and a non-nucleoside reverse transcriptase inhibitor (NNRTI). Second-line regimens contained a protease inhibitor and two new NRTIs. Before initiating ART, HIV counseling and testing (HIV-CT) of patient partners was recommended to facilitate HIV care for infected partners or prevention interventions for sero-discordant couples.

Patient Monitoring

At ART initiation, standardized medical records, recommended by the Ministry of Health (MOH), were completed. These records captured, among other variables: date of visit, sex, age at enrollment, date of birth, marital status, employment status, HIV status of partner, WHO stage, CD4 count, weight, height, hemoglobin level, ART regimen and CTX prescription status. At all subsequent visits, the date of the visit, patient weight, clinical stage and ART regimen prescribed were recorded, while at 6-monthly intervals after ART initiation, hemoglobin measurements, and CD4 counts were recommended to monitor disease progression or improvement. Patients collected medications monthly from clinic pharmacies where the date of scheduled antiretroviral (ARV) pick-up appointments and actual ARV pick-up occurrences were documented.

As in other resource-constrained settings where viral load monitoring is not routine [6,13,17], during 2004–2008, treatment failure was detected using clinical and immunologic criteria. A new or recurrent stage III or IV condition was considered clinical evidence of possible treatment failure; however, confirmation of failure, with a CD4 measurement, was required before switching regimens. All facilities providing ART had access to CD4 assays. Immunologic failure criteria included a CD4 count decline from baseline, a CD4 count <100/μL, or a 50% decline from peak CD4 count, after ≥6 months of therapy [16]. At the limited number of sites with access to viral load testing facilities, viral load measurements were also encouraged.

Treatment Outcome Measures

Our reported mortality rate represents the number of documented deaths during ART per 100 person-years of observed therapy. The attrition rate represents the number of patients lost through attrition (death, LTFU, or stopping ART) per 100 person-years of observed therapy. A patient absent from a health facility in the 90 days preceding data abstraction was considered LTFU, unless the medical record stated that the patient had died, stopped ART, or transferred. The date of LTFU was recorded as the date of the most recent visit, or one day after ART initiation if patients only attended the initiation visit. One-year attrition proportions are reported to allow comparison with published literature [11,18]. Patients transferring to other facilities during the first treatment year were excluded from the one-year attrition proportion. Transfers were censored from time-to-event analyses at the date of transfer.

To allow comparison with other resource-constrained programs [6,17], we report incidence of the first CD4 count meeting immunologic treatment failure criteria. Incidence of clinical failure is not reported because MOH guidelines required that the often inaccurate diagnosis of clinical failure [19] be confirmed with objective CD4 count measurements before a regimen change was considered. To allow comparison with other programs in resource-limited settings, we also report rates of regimen-switch per 100 patient-years [20].

CD4 counts were determined using FlowCount PLG CD4 assay and analyzed with Beckman Coulter Epics XL-MCL Flow Cytometers (both Beckman Coulter, Inc., Johannesburg, South Africa). Blood Hemoglobins were determined with an automated hematology analyzer, Sysmex XT2000i (Sysmex Europe, Inc., Hamburg, Germany) or HemoCue photometer assay (HemoCue AB, Helsingborg, Sweden).

Similar to other cohort studies in resource-constrained settings [6], adherence to ART was estimated by measuring timeliness of patient visits to scheduled medicine pick-up appointments at clinic-based pharmacies [21–23] during the first six months of ART.

Study Design and Population

A retrospective cohort study design was used. Patient-level data were abstracted from standardized, MOH-recommended medical records onto study questionnaires by trained abstractors in November 2008. Only medical records of adult patients, ≥15 years old at ART initiation, who started ART during 2004–2007, were eligible.

Sample Size

Sample size calculations were performed using Epi Info™ software (CDC, Epi Info 2008, Version 3.5.1, Atlanta, GA). To achieve a 95% confidence interval (CI) of ±3.0% around the estimate for 6-month attrition, assuming a design effect of 1.5, and a conservative (i.e. higher than expected) 6-month attrition proportion of 25% [11], a sample size of ≥1,200 patient records was needed. This study was linked with a cost-effectiveness study; we aimed to sample 2,600 medical records to meet the needs of both studies.

Sampling

During study planning in November 2007, MOH-reported data from the end of December 2006 were used to define the clinic sample frame. By December 2006, 43,295 adults had initiated ART at 152 clinics, all of which were managed by the MOH with funding support from several donor agencies including the U.S. President's Emergency Plan for AIDS Relief (PEPFAR). Clinics that had initiated <50 adults on ART by this time were excluded from the sample frame, resulting in 58 clinics, ranging in size from 1–49 adult ART enrollees and supporting only 1,061 adult ART patients, being excluded. Ninety-four clinics, which had each enrolled between 50 and 3,530 adult ART patients, were included in the sample frame; of these clinics, 12 with >1,000 enrollees were selected with certainty, while 18 with 50–1,000 enrollees were selected using probability-proportional-to-size sampling. All 11 provinces in Mozambique were represented by the sample with at least one ART clinic selected from each province. From the 30

selected clinics, ranging in site size from 70–3,530 adult ART enrollees, we aimed to randomly sample 2,600 medical records.

Analytic Methods

Data were analyzed using SAS 9.2 (SAS Institute Inc., Cary, NC), STATA 10 (StataCorp, 2009, Stata Statistical Software, Release 10, College Station, TX), and SUDAAN (Research Triangle Institute, 2005, SUDAAN, Release 9.0.1. Research Triangle Park, NC). Data were weighted and survey design controlled for, during analysis.

Multiple imputation with chained equations was used to impute missing baseline demographic and clinical data [24]. The ice [25–27] procedure in Stata was used to create 20 imputed datasets for each of two outcomes included in multivariable analysis: Attrition and treatment failure. The imputation model included the event indicator, all study variables, and the Nelson-Aalen estimate of cumulative hazard [28]. Missing data were assumed *missing at random* (MAR) and all patients had complete time-to-event data. When comparing baseline characteristics among groups, the χ^2 and *t*-test were used to compare categorical and continuous variables, respectively. Kaplan-Meier curves were used to examine retention proportions stratified by baseline variables. Cox proportional hazards regression models were used to estimate unadjusted and adjusted hazard ratios (AHRs), 95% CIs, and *p*-values. The proportional hazards assumption was assessed separately for each imputed dataset using visual methods and the Grambsch and Therneu test [29]. Estimates were combined across the imputed datasets according to Rubin's rules [24]. This was accomplished using SUDAAN and the mim procedure in Stata [30].

As a secondary analysis, mean CD4 count and weight change over time was estimated to allow comparison with published literature. SAS PROC MIXED was used to fit unweighted polynomial growth curve models with maximum likelihood (ML) estimation to the data [31]. Random effects were specified on the intercept and slope terms. Predicted means and 95% CIs from the polynomial growth curve models were plotted to graphically illustrate change over time.

Results

Characteristics at ART Initiation

Data from medical records of 2,596 eligible, adult ART patients were abstracted and analyzed. Table 1 illustrates analysis results for both original and imputed datasets; in the following text, weighted imputed data are reported unless otherwise stated.

At ART initiation, median age of patients was 34 years, and 62% were female of whom 16% (95% CI, 6–25%) were pregnant. About 53% (95% CI, 50–57%) of patients were married or in a civil union. Partner sero-status was documented in 13% (95% CI, 11–15%) of all records. In un-weighted analysis, 37 (11%) of 330 tested partners were sero-negative.

Active tuberculosis (TB) was frequent at ART initiation with 11% of patients receiving TB treatment. Many patients were underweight; 18% weighed <45 kilograms (kg) at ART start. Advanced disease was common with 15% of patients diagnosed as having WHO stage IV disease.

Median CD4 count at ART initiation was 153/μL; 16% (95% CI, 14–19%) had counts below 50/μL and 50% (95% CI, 47–54%) had counts ranging from 50–200/μL. Median CD4 counts at ART initiation were lower for males than females (139/μL vs. 159/μL, p<0.01).

Median hemoglobin at ART initiation was 10.3 g/dL. Severe anemia (hemoglobin <8 g/dL) was present in 13% (95% CI, 12–

15%) of patients. Median hemoglobin at ART initiation was lower for women than men (9.9 vs. 11.0 g/dL, p<0.01).

Stavudine (d4T), lamivudine (3TC), and nevirapine (NVP) or efavirenz (EFV) comprised 88% of first-line regimens. Only 31% (95% CI, 26–36%) of patients were prescribed CTX at ART initiation; 3% (95% CI, 2–3%) were not prescribed CTX due to allergy.

Mortality and Attrition

Of 2,596 patients sampled, 164 died, 564 were LTFU, and 10 stopped ART indefinitely during 4,001 patient-years of follow-up. Median follow-up duration was 1.3 years (interquartile range, 0.7–2.2 years). Of patients who died, 56% (95% CI, 49–63%) died within 90 days of starting ART. Of patients who were LTFU, 41% (95% CI, 35–48%) were lost within 90 days of starting ART.

Mortality rates were 3.4 deaths per 100 patient-years overall (95% CI, 2.8–4.2), 12.9 deaths per 100 patient-years in the first 90 days (95% CI, 9.9–17.1), and 1.8 deaths per 100 patient-years after 90 days (95% CI, 1.3–2.4).

One-year attrition was 21% (95% CI, 17–25%) with 15% LTFU (95% CI, 11–18%), 5% dead (95% CI, 4–6%), and 1% stopping ART (95% CI, 0–3%). Attrition rates were 19.8 attritions per 100 patient-years overall (95% CI, 17.9–21.9), 57.2 attritions per 100 patient-years in the first 90 days (95% CI, 49.5–66.4), and 13.2 attritions per 100 patient-years after 90 days (95% CI, 11.7–15.0).

Male sex was associated with attrition (AHR 1.5; 95% CI, 1.3–1.8) (Table 2, figure 1A). Attrition risk decreased as age at ART initiation increased (AHR associated with a 10-year age increase was 0.8; 95% CI, 0.8–0.9). Patients starting ART one calendar year later had higher attrition rates (AHR 1.5; 95% CI, 1.2–1.8). Compared to patients with baseline WHO stage I/II, patients with stage IV were at higher attrition risk (AHR 1.7; 95% CI, 1.3–2.4) (Table 2, figure 1B). Patients with weight <45 kg (AHR 2.1; 95% CI, 1.6–2.9) (Table 2, figure 1C), and severe anemia (AHR 1.6; 95% CI, 1.2–2.1) were at higher attrition risk.

Patients not prescribed CTX were at higher risk for attrition compared with patients who were prescribed this drug (AHR 1.4; 95% CI, 1.0–1.8) (Table 2, Figure 1D).

Immunologic Treatment Failure and Regimen Switch

Of 2,596 patients at risk, 486 experienced immunologic treatment failure during 3,481 patient-years of follow-up. The failure rate was 14.0 failures per 100-patient years (95% CI, 13.0-16.0). The regimen-switch rate was 0.6 per 100-patient years (95% CI, 0.4-1.1).

Predictors of immunologic treatment failure included male sex (AHR 1.4; 95% CI, 1.2–1.8) and being single or widowed (AHR 1.3; 95% CI, 1.2–1.5). Risk of immunologic failure decreased as age at ART initiation increased (AHR associated with a 10-year age increase was 0.9; 95% CI, 0.8–1.0).

CD4 Count and Weight Response

Modeled mean gains in CD4 count were 173, 186, 237, 273, and 293/μL at 6, 12, 24, 36 and 48 months of follow-up, respectively (Figure 2A). Compared with males, females tended to start ART with higher CD4 counts and maintain higher counts during the first three treatment years (Figure 2B). Modeled mean weight gains were 2.8, 3.8, 3.8, 3.9 and 3.6 kilograms at 6, 12, 24, 36 and 48 months of therapy (Figure 3A) and were similar for males and females (Figure 3B).

Adherence

About 71% (95% CI, 66–77%) of patients were ≥95% adherent to ART medicine pick-up appointments during the first six months

Table 1. Demographic and clinical characteristics of adult patients at ART initiation – Mozambique, 2004–2007.

	Original Data				Following Multiple Imputation (N = 2,596)	
	Un-weighted Frequency of Observations	Un-weighted Total	Weighted Median with IQR*, or, Percentage with 95% CI		Weighted Median with IQR*, or, Percentage with 95% CI	
Median Age†, No., N, median (IQR)						
Both Sexes	2,596	2,596	**34**	(28–42)	**34**	(28–42)
Female	1,576	1, 576	**32**	(27–39)	**32**	(27–39)
Male	1,020	1,020	**38**	(31–45)	**38**	(31–45)
Female†, No., N, %, (95% CI)	1,576	2,596	**62%**	(59–65%)	**62%**	(59–65%)
Employment status, No., N, %, (95% CI)						
Employed	992	2,268	**46%**	(40–51%)	**45%**	(40–51%)
Student	107	2,268	**4%**	(3–5%)	**4%**	(3–5%)
Unemployed	1,169	2,268	**50%**	(45–56%)	**51%**	(46–56%)
observations missing data**	328	2,596	**13%**			
Active TB, No., N, %, (95% CI)	267	2,564	**11%**	(9–13%)	**11%**	(9–13%)
observations missing data**	32	2,596	**1%**			
WHO Stage, No., N, %, (95% CI)						
I/II	619	1,617	**37%**	(32–42%)	**40%**	(34–45%)
III	739	1,617	**47%**	(43–52%)	**45%**	(40–50%)
IV	259	1,617	**16%**	(13–18%)	**15%**	(13–18%)
observations missing data**	979	2,596	**38%**			
Weight Category, No., N, % (95% CI)						
<45 kg	367	2,061	**17%**	(14–20%)	**18%**	(15–21%)
45–60 kg	1,224	2,061	**59%**	(56–61%)	**57%**	(54–59%)
> 60 kg	470	2,061	**24%**	(21–27%)	**25%**	(22–28%)
observations missing data**	535	2,596	**21%**			
BMI<18.5, No., N, %, (95% CI)	354	1,200	**28%**	(23–33%)	**28%**	(23–33%)
observations missing data**	1,396	2,596	**54%**			
CD4+ T-cell count, No., N, median (IQR)						
Both Sexes	2,254	2,596	**155**	(76–231)	**153**	(74–231)
Female	1,373	1,576	**161**	(88–244)	**159**	(87–243)
Male	881	1,020	**141**	(59–213)	**139**	(59–213)
observations missing data**	342	2,596	**13%**			
Hemoglobin, No., N, median (IQR)						
Both Sexes	1,899	2,596	**10.1**	(8.8–11.6)	**10.3**	(8.8–11.7)
Female	1,182	1,576	**9.8**	(8.6–11.0)	**9.9**	(8.5–11.1)
Male	717	1,020	**11.1**	(9.0–12.5)	**11.0**	(9.2–12.5)
First Line ART Regimens†, No., N, %, (95% CI)						
D4T + 3TC + NVP/EFV	2,315	2,596	**88%**	(82–94%)	**88%**	(82–94%)
AZT + 3TC + NVP/EFV	240	2,596	**11%**	(5–16%)	**11%**	(5–16%)
D4T/AZT + 3TC + ABC	17	2,596	**<1%**	(0–1%)	**<1%**	(0–1%)
Other	24	2,596	**<1%**	(0–1%)	**<1%**	(0–1%)

Abbreviations: CI, confidence interval; IQR, interquartile range; TB, tuberculosis; WHO, World Health Organization; Kgs, kilograms; BMI, body mass index; ART, antiretroviral therapy; D4T, stavudine; 3TC, lamivudine; NVP, nevirapine; EFV, efavirenz; AZT, zidovudine; ABC, abacavir; CTX, co-trimoxazole.
*Median and IQR calculated across 20 imputed datasets.
†Variables with complete data.
**Unweighted sample estimate.
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Table 2. Patient characteristics at antiretroviral therapy initiation associated with attrition and treatment failure.

	Attrition*				Immunologic Treatment Failure*			
	Original	Following Multiple Imputation (N = 2,596)			Original	Following Multiple Imputation (N = 2,596)		
		Rate/100PY	HR (95% CI)	AHR[†] (95% CI)		Rate/100PY	HR (95% CI)	AHR[†] (95% CI)
Sex								
Female	1,576	17.9	1.0	1.0	1,576	13.1	1.0	1.0
Male	1,020	23.0	**1.3 (1.1–1.4)**	**1.5 (1.3–1.8)**	1,020	15.5	**1.2 (1.0–1.4)**	**1.4 (1.2–1.8)**
Age[**]	2,596	–	**0.8 (0.8–0.9)**	**0.8 (0.8–0.9)**	2,596	–	**0.9 (0.8–1.0)**	**0.9 (0.8–1.0)**
Year of ART Start[‡]	2,596	–	**1.6 (1.2–2.0)**	**1.5 (1.2–1.8)**	2,596	–	1.1 (1.0–1.2)	1.1 (0.9–1.2)
Married								
Married/Living together	1,211	21.2	1.0	1.0	1,211	12.4	1.0	1.0
Single/Widowed	1,152	18.3	0.9 (0.8–1.0)	0.9 (0.8–1.1)	1,152	15.7	**1.3 (1.1–1.4)**	**1.3 (1.2–1.5)**
Employed								
Yes	992	18.8	1.0	1.0	992	13.4	1.0	1.0
Student	107	18.8	1.1 (0.7–1.5)	1.0 (0.7–1.4)	107	15.8	1.2 (0.9–1.5)	1.0 (0.7–1.5)
No	1,169	20.8	1.1 (0.8–1.4)	1.1 (0.9–1.3)	1,169	14.4	1.1 (0.9–1.3)	1.1 (0.8–1.4)
Active TB								
No	2,297	18.9	1.0	1.0	2,297	14.0	1.0	1.0
Yes	267	27.9	**1.4 (1.0–2.0)**	1.0 (0.8–1.4)	267	13.9	1.0 (0.8–1.3)	1.0 (0.7–1.3)
WHO Stage								
Stage I/II	619	13.3	1.0	1.0	619	14.6	1.0	1.0
Stage III	739	21.7	**1.5 (1.2–2.0)**	1.1 (0.8–1.6)	739	13.1	0.9 (0.7–1.2)	0.9 (0.6–1.2)
Stage IV	259	35.0	**2.5 (1.9–3.2)**	**1.7 (1.3–2.4)**	259	14.7	1.0 (0.7–1.4)	0.9 (0.6–1.3)
Weight								
>60	470	12.7	1.0	1.0	470	11.7	1.0	
45–60	1,224	18.1	1.4 (1.1– 1.8)	1.2 (1.0–1.6)	1,224	14.3	1.2 (1.0–1.5)	1.3 (1.0–1.7)
<45	367	41.9	2.9 (2.2–3.8)	**2.1 (1.6–2.9)**	367	17.5	1.5 (1.0–2.3)	1.6 (1.0–2.6)
CD4+ T-cell count (cells/μL)								
>200	754	18.8	1.0	–	754	16.7	1.0	–
>50–≤200	1,144	18.5	1.0 (0.8–1.3)	–	1,144	10.2	**0.6 (0.5–0.8)**	–
<50	356	25.8	**1.4 (1.1–1.8)**	–	356	22.1	1.3 (1.0–1.8)	–
Hemoglobin								
≥8.0 g/dL	1,664	17.2	1.0	1.0	1,664	14.0	1.0	1.0
<8.0 g/dL	235	40.5	**2.2 (1.7–2.8)**	**1.6 (1.2–2.1)**	235	14.1	1.0 (0.7–1.5)	0.9 (0.6–1.5)
Prescribed CTX								
Yes	821	15.3	1.0	1.0	821	13.7	1.0	1.0
No	1,775	21.9	**1.4 (1.1–1.8)**	**1.4 (1.0–1.8)**	1,775	14.1	1.0 (0.9–1.2)	1.0 (0.9–1.2)
Adherence								
≥95%	1,263	17.0	1.0	1.0	1,263	13.4	1.0	1.0
<95%	597	28.3	**1.5 (1.0–2.3)**	1.3 (0.8–2.0)	597	15.8	1.2 (0.9–1.6)	1.2 (0.9–1.6)
Site Size								
>1,000 patients	2,109	15.9	1.0	1.0	2109	13.8	1.0	1.0
≤1,000 patients	487	37.3	**2.1 (1.1–3.8)**	1.5 (0.8–2.6)	487	14.6	1.0 (0.7–1.5)	1.0 (0.6–1.5)

Abbreviations: Rate/100PY, rate per 100 person-years; HR, hazards ratio; AHR, adjusted hazards ratio; CI, confidence interval; TB, tuberculosis; WHO, World Health Organization; BMI, body mass index; CTX, co-trimoxazole.
*Stratified by CD4+ T-cell count (cells/μL).
[†]All variables listed in this table were included in the multivariate Cox proportional hazards regression model.
[**]Hazard ratios associated with a 10-year increase in age.
[‡]Date of ART initiation was entered into the model; hazard ratios represent a yearly increase rather than a daily increase.
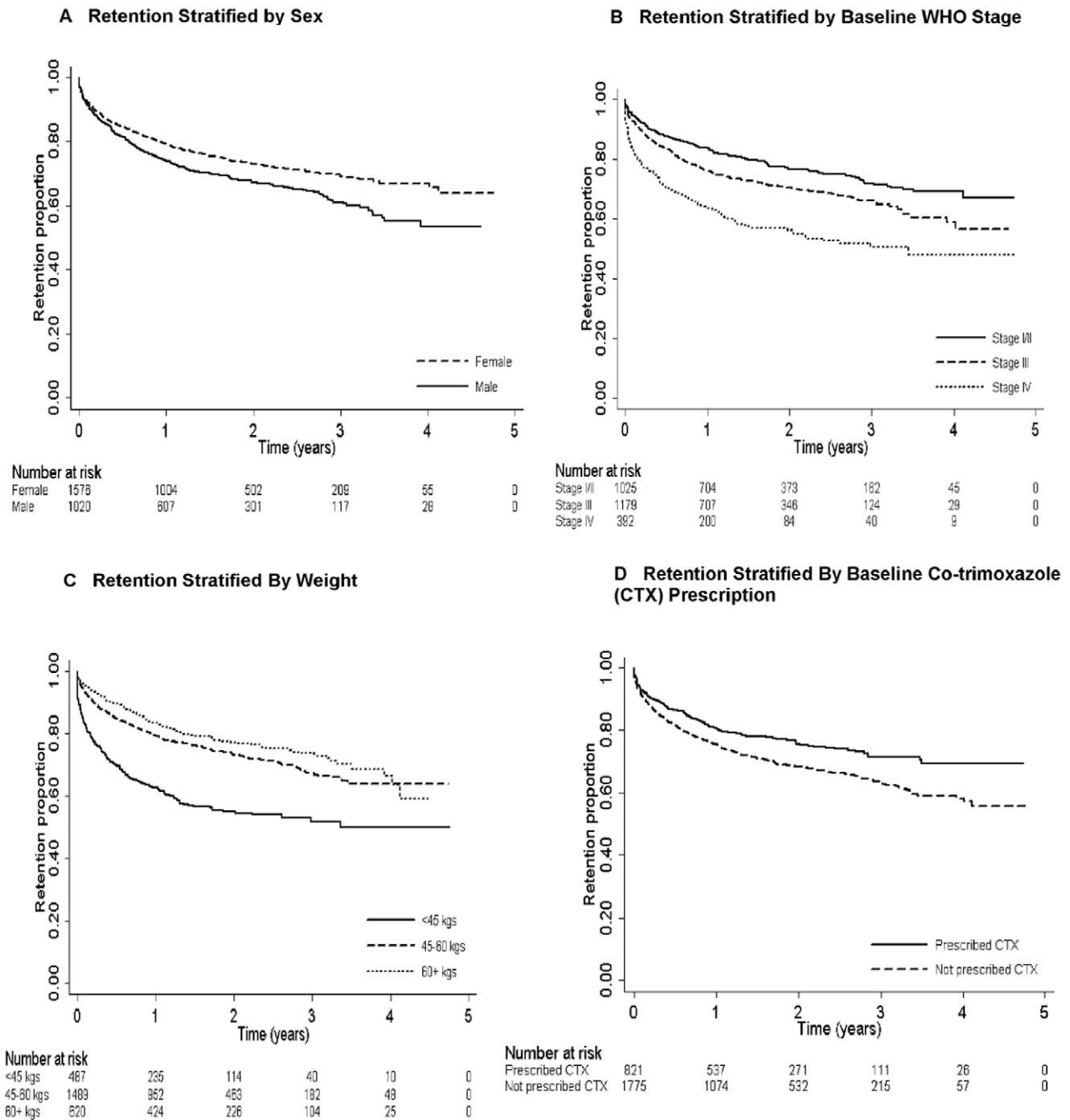doi:10.1371/journal.pone.0018453.t002

A Retention Stratified by Sex

B Retention Stratified by Baseline WHO Stage

C Retention Stratified By Weight

D Retention Stratified By Baseline Co-trimoxazole (CTX) Prescription

Figure 1. Kaplan-Meier Analysis (imputed dataset 1).
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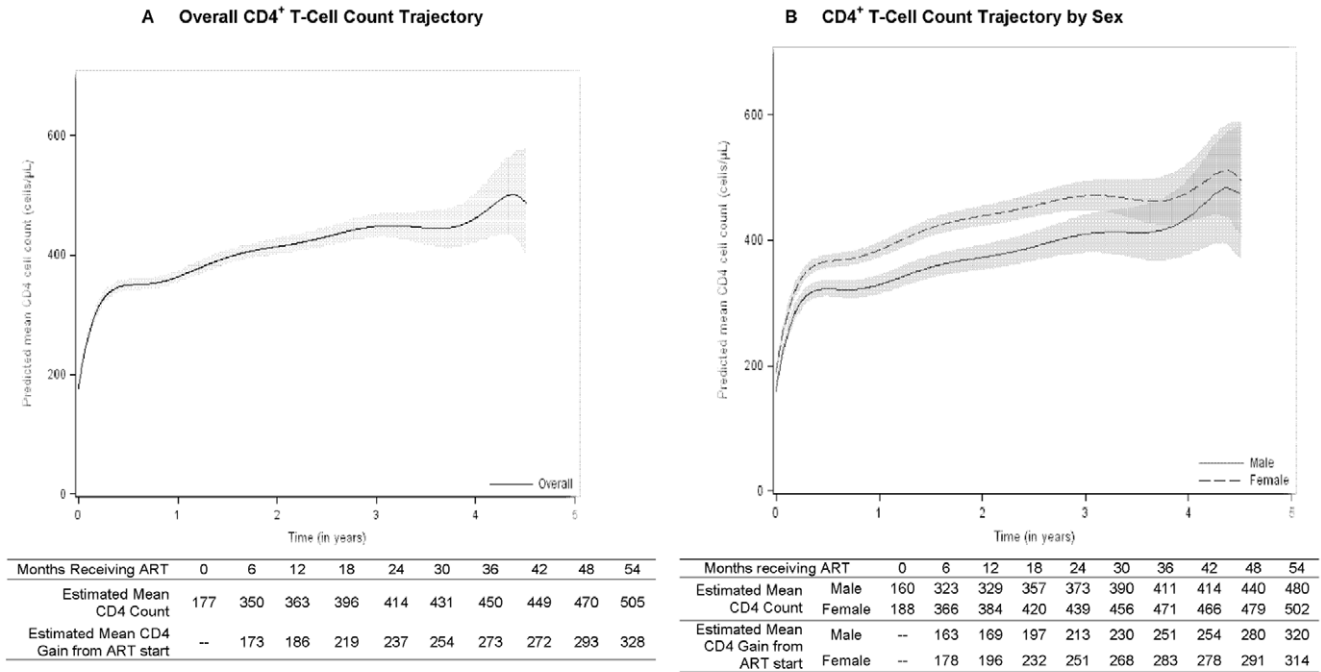
of therapy. Proportions of female and male ART patients who were ≥95% adherent to ART were not significantly different (72% vs. 70%, p = 0.12).

Discussion

This study adds to the limited number of nationally representative ART outcome assessments from sub-Saharan Africa [6,12,13], and has several important findings.
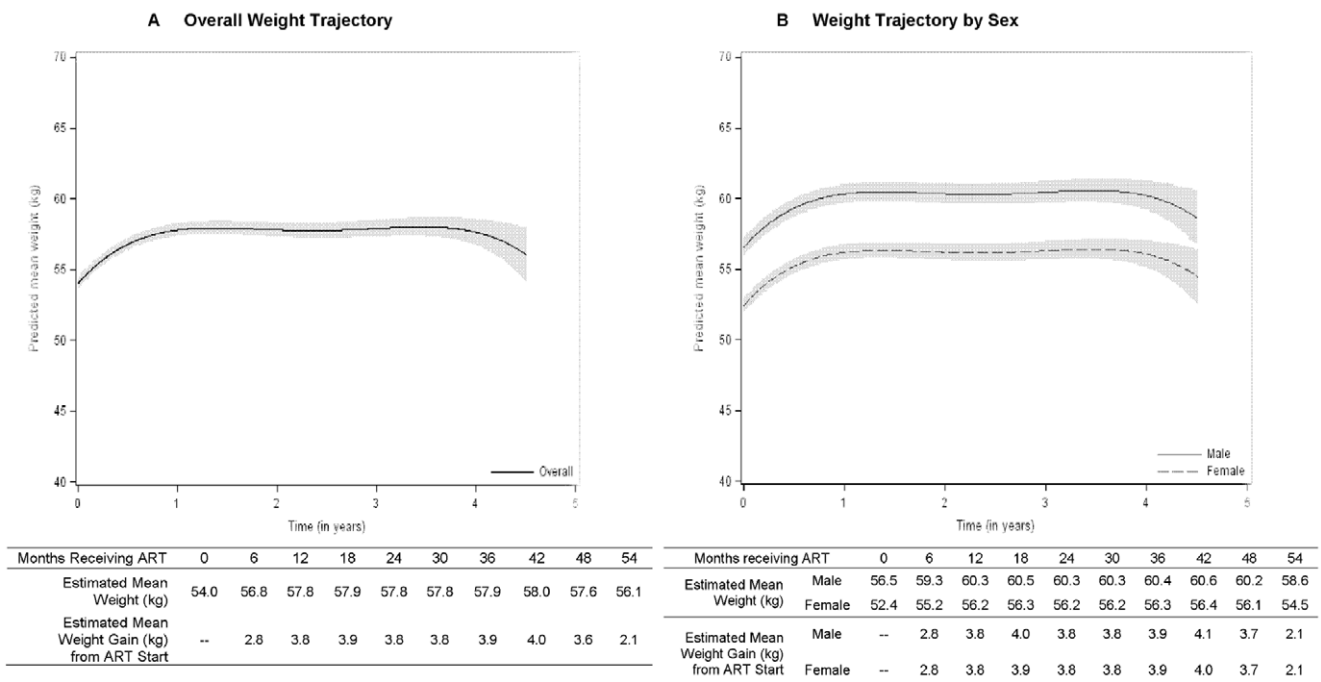
First, rates of mortality, attrition, and immunologic treatment failure for the study period are comparable with reports from other

ART programs in resource-rich [9] and -constrained settings [6,9,11,13,17,18], which is encouraging for program funders and managers. Second, regimen-switch rates were very low, a finding which deserves further research as it possibly indicates delay in switching patients from failing first-line regimens and risk of poor longer term outcomes [20]. Third, as has been reported from other ART programs, male sex, advanced HIV disease, and malnutrition predicted poor outcomes, suggesting that male-specific interventions, starting ART at earlier disease stages, and evidence-based interventions for malnourished patients, could

A Overall CD4+ T-Cell Count Trajectory

B CD4+ T-Cell Count Trajectory by Sex

Months Receiving ART	0	6	12	18	24	30	36	42	48	54
Estimated Mean CD4 Count	177	350	363	396	414	431	450	449	470	505
Estimated Mean CD4 Gain from ART start	--	173	186	219	237	254	273	272	293	328

Months receiving ART		0	6	12	18	24	30	36	42	48	54
Estimated Mean CD4 Count	Male	160	323	329	357	373	390	411	414	440	480
	Female	188	366	384	420	439	456	471	466	479	502
Estimated Mean CD4 Gain from ART start	Male	--	163	169	197	213	230	251	254	280	320
	Female	--	178	196	232	251	268	283	278	291	314

CD4+ T-cell count trajectories were estimated using an eighth degree polynomial growth curve model with maximum likelihood estimation. Random-effects were specified on the intercept, linear slope, and quadratic slope terms. Shaded areas represent 95% confidence intervals around modeled estimates.

Figure 2. Modeled changes in CD4 count for surviving patients initiating antiretroviral therapy during 2004–2007.
doi:10.1371/journal.pone.0018453.g002

A Overall Weight Trajectory

B Weight Trajectory by Sex

Months Receiving ART	0	6	12	18	24	30	36	42	48	54
Estimated Mean Weight (kg)	54.0	56.8	57.8	57.9	57.8	57.8	57.9	58.0	57.6	56.1
Estimated Mean Weight Gain (kg) from ART Start	--	2.8	3.8	3.9	3.8	3.8	3.9	4.0	3.6	2.1

Months receiving ART		0	6	12	18	24	30	36	42	48	54
Estimated Mean Weight (kg)	Male	56.5	59.3	60.3	60.5	60.3	60.3	60.4	60.6	60.2	58.6
	Female	52.4	55.2	56.2	56.3	56.2	56.2	56.3	56.4	56.1	54.5
Estimated Mean Weight Gain (kg) from ART Start	Male	--	2.8	3.8	4.0	3.8	3.8	3.9	4.1	3.7	2.1
	Female	--	2.8	3.8	3.9	3.8	3.8	3.9	4.0	3.7	2.1

Weight trajectories were estimated using a fourth degree polynomial growth curve model with maximum likelihood estimation. Random-effects were specified on the intercept and all slope parameters. Shaded areas represent 95% confidence intervals around modeled estimates.

Figure 3. Modeled changes in weight over time for surviving patients initiating ART during 2004–2007.
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improve future program outcomes. Fourth, CTX prescription at ART initiation, which was documented for about one third of patients, was associated with lower attrition risk, highlighting the need for CTX scale-up among ART patients [32–35]. Fifth, attrition rates appeared to increase as the ART program expanded, a concerning trend that requires further research to better understand underlying causes. Finally, documenting partner sero-status was uncommon, possibly indicating missed prevention opportunities among sero-discordant couples [36].

Treatment Outcomes

Meta-analyses of attrition in adult ART programs in sub-Saharan Africa have reported mean one-year attrition proportions of 25% and 20% for the review periods of 2000-2007 [11] and 2007–2009 [18], respectively. In these studies, LTFU accounted for between 56% [11] and 59% [18] of attrition. Similarly, in our study, one-year attrition was 21% with LTFU accounting for most (71%) of patient loss from the program. This relatively high contribution of LTFU to attrition likely reflects the lack of active patient tracing systems in most Mozambican ART facilities [9,37] due to their cost [37].

Since recent reports show that patient death may account for 29–59% of patients who are LTFU [38–42], we should consider our reported mortality and attrition rates as best- and worst-case estimates of true mortality. Our best-case estimate of mortality (3.4 deaths per 100 person-years) is similar to reports from adult ART programs with passive follow-up systems (2.7 deaths per 100 patient-years [9]) but predictably lower than that reported for programs with active follow-up systems (5.5 deaths per 100 patient-years [9]). Our worst case estimate for true mortality (19.8 deaths per 100 person-years) is comparable with the rate of 16.1 deaths per 100 patient-years reported for a large Zambian cohort [6].

Similar to other programs [4–6,8–10,13,17,43–50], large proportions of documented mortality (56%) and LTFU (41%) occurred within 90 days of starting ART, highlighting the importance of this time period. Evidence based interventions which reduce early mortality are urgently needed [51]. To limit early LTFU from the program, the MOH is considering a pilot program to rapidly trace all patients LTFU in the 90 days following ART initiation.

Post 90-day mortality and attrition rates of 1.8 and 13.2 events per 100 patient-years, respectively, are comparable with post 90-day mortality rates reported from both resource-constrained [6,9,17] and –rich [9] settings (5.0–6.0 deaths per 100 patient-years). Similarly, CD4 count and weight gains [6,9,13,17], and rates of immunologic treatment failure [6,17,52], are similar to those reported from other ART programs.

Rates of Regimen-Switch

Compared with the average regimen-switch rate for resource-constrained programs (2.4 switches per 100 person-years, 95% CI, 2.2–2.6 [20]), our switch rate (0.6 regimen changes per 100 person-years) was low. Limited access to routine viral load testing has likely contributed to low regimen-switch rates [20]. However, even compared with regimen-switch rates among programs lacking routine viral load testing (2.0 per 100 person-years, 95% CI 1.8–2.3 [20]), our switch rates are low. Limited access to second-line therapy, or training in its use, may have contributed to low switch rates, especially at more peripheral health facilities where ART is commonly managed by non-physician clinicians [20,53]. Because low regimen-switch rates may indicate delay in switching patients from failing first-line regimens, which could lead

to poor long-term outcomes [20,54] further research to better understand the reasons for low regimen-switch rates is planned.

Advanced HIV Disease

Similar to other reports, many patients initiated ART with WHO stage IV disease, which was a strong predictor of attrition [5,6,9,44,47,49,55–57]. This suggests that earlier diagnosis and ART is needed to improve program outcomes [51]. Mass HIV testing campaigns, earlier entry into HIV care, and ensuring pre-ART care retention, will likely be needed to reduce absolute numbers of late starters [51]. In addition, the MOH raised the CD4 count threshold for ART initiation among adults with WHO stage I/II disease from 200 to 250/μL in 2008, and from 250 to 350/μL in 2010 [58], which should reduce the proportion of patients initiating ART with end-stage disease.

Male Sex

Similar to other reports [5,6,8,10,55], male sex was associated with poorer outcomes. In our study, males tended to initiate ART at lower CD4 counts than females, suggesting late presentation contributed to poorer outcomes. However, even in multivariate analysis, male sex was associated with poorer outcomes. Reasons for this are unknown, but could relate to differences in health-seeking or adherence behavior [59].

Malnutrition

As in other studies, our marker for malnutrition at ART initiation (weight <45 kg [60]) was independently associated with poor outcomes. In Mozambique, the relative importance of underlying causes of malnutrition among HIV-infected persons is not well understood and deserves further research [61]. Food supplementation and empiric TB treatment for malnourished patients are interventions which require evaluation [62].

Failure to Prescribe CTX

Although CTX prescription is recommended for all ART patients in Mozambique throughout therapy, few (31%) received this drug at ART initiation during 2004–2007. Reasons for this are unknown but may include CTX supply shortfalls and clinician concerns about CTX drug resistance development [32,33,63].

Although several studies have demonstrated an additional benefit of CTX for ART patients [32–35], reasons for this are not fully understood [32]. In Mozambique, where malaria is an important cause of mortality among ART patients [64], the prophylactic action of CTX against this disease [32], may partly explain its association with improved treatment outcomes.

Additional in-service training for ART-service providers is planned to rapidly scale-up CTX prescription. Further, CTX procurement and distribution procedures are being reviewed to ensure that all ART delivery sites have adequate CTX supplies.

Temporal Trends in Attrition

As the ART program expanded during 2004 through 2007, attrition risk for newly enrolled adult ART patients increased, a temporal trend which has been observed in other programs in resource-constrained settings [65,66]. Yearly increases in observed LTFU due to undocumented transfer of patients to more peripheral facilities during the decentralization process, may explain the increases in observed attrition rates [67,68]; however, further research to establish the reasons for increasing rates of attrition is urgently needed [65].

Partner HIV Counseling and Testing

The low proportion of records with documented partner sero-status is concerning because partner HIV counseling and testing (HIV-CT) can facilitate entry into HIV care for infected individuals or appropriate prevention strategies for uninfected partners [36,69–76]. Poor completion of the partner sero-status field in standard MOH medical records may be due to several barriers including hesitancy of patients to disclose HIV status, partner refusal to be tested, or loss to follow-up of the patient before the field can be filled. Further research to identify and overcome these barriers is needed.

Limitations

Limitations primarily relate to the fact that these analyses were based on routinely collected data, which were incomplete for certain baseline and follow-up clinical characteristics.

Summary

Treatment outcomes were comparable with reports from other ART programs; however, the low ART regimen-switch rates and the increasing attrition rates during program expansion are concerning findings which deserve further research. Immediate public health responses that may improve treatment outcomes include initiation of ART at earlier disease stages, especially among males, scale-up of CTX, and evidence-based interventions for malnourished patients; in addition, scale-up of partner HIV-CT could significantly contribute to national HIV prevention efforts.

Author Contributions

Conceived and designed the experiments: AFA FM RWS MS CA LJN TE. Performed the experiments: AFA FM RWS MS CA LJN TE. Analyzed the data: RWS AFA. Contributed reagents/materials/analysis tools: AFA FM RWS MS CA LJN. Wrote the paper: AFA FM RWS MS CA LJN TE.

References

1. World Health Organization. Towards universal access: Scaling up priority HIV/AIDS interventions in the health sector - Progress report (2010) Geneva, Switzerland: WHO; 2010. Avaliable: http://wwwwhoint/hiv/pub/2010progressreport/full_report_enpdf. Accessed 29 January 2011.
2. Boerma JT, Stanecki KA, Newell ML, Luo C, Beusenberg M, et al. (2006) Monitoring the scale-up of antiretroviral therapy programmes: methods to estimate coverage. Bull World Health Organ 84: 145–150.
3. Bennett S, Boerma JT, Brugha R (2006) Scaling up HIV/AIDS evaluation. Lancet 367: 79–82.
4. Laurent C, Ngom Gueye NF, Ndour CT, Gueye PM, Diouf M, et al. (2005) Long-term benefits of highly active antiretroviral therapy in Senegalese HIV-1-infected adults. J Acquir Immune Defic Syndr 38: 14–17.
5. Ferradini L, Jeannin A, Pinoges L, Izopet J, Odhiambo D, et al. (2006) Scaling up of highly active antiretroviral therapy in a rural district of Malawi: an effectiveness assessment. Lancet 367: 1335–1342.
6. Stringer J, Zulu I, Levy J, Stringer EM, Mwango A, et al. (2006) Rapid scale-up of antiretroviral therapy at primary care sites in Zambia: feasibility and early outcomes. JAMA 296: 782–793.
7. Severe P, Leger P, Charles M, Noel F, Bonhomme G, et al. (2005) Antiretroviral therapy in a thousand patients with AIDS in Haiti. N Engl J Med 353: 2325–2334.
8. Coetzee D, Hildebrand K, Boulle A, Maartens G, Louis F, et al. (2004) Outcomes after two years of providing antiretroviral treatment in Khayelitsha, South Africa. AIDS 18: 887–895.
9. Braitstein P, Brinkhof MW, Dabis F, Schechter M, Boulle A, et al. (2006) Mortality of HIV-1-infected patients in the first year of antiretroviral therapy: comparison between low-income and high-income countries. Lancet 367: 817–824.
10. Toure S, Kouadio B, Seyler C, Traore M, Dakoury-Dogbo N, et al. (2008) Rapid scaling-up of antiretroviral therapy in 10,000 adults in Cote d'Ivoire: 2-year outcomes and determinants. AIDS 22: 873–882.
11. Rosen S, Fox MP, Gill CJ (2007) Patient retention in antiretroviral therapy programs in sub-Saharan Africa: a systematic review. PLoS Med 4: e298.
12. Wools-Kaloustian K, Kimaiyo S, Diero L, Siika A, Sidle J, et al. (2006) Viability and effectiveness of large-scale HIV treatment initiatives in sub-Saharan Africa: experience from western Kenya. AIDS 20: 41–48.
13. Lowrance DW, Ndamage F, Kayirangwa E, Ndagije F, Lo W, et al. (2009) Adult clinical and immunologic outcomes of the national antiretroviral treatment program in Rwanda during 2004-2005. J Acquir Immune Defic Syndr 52: 49–55.
14. Multi-Sectoral Technical Group Support for the Fight Against HIV/AIDS in Mozambique. Impacto demografico do HIV/SIDA em Mozambique. Ministry of Health HIV/AIDS Website. Available: http://www.ine.gov.mz/hiv/ImpactoHIV.pdf. Accessed 29 January 29 2011.
15. World Health Organization. Towards universal access - scaling up priority HIV/AIDS interventions in the health sector, (2008) Geneva Switzerland: WHO; 2008. Accessed: http://www.who.int/hiv/pub/2008progressreport/en/. Accessed 29 January 2011.
16. World Health Organization. Antiretroviral therapy for HIV infection in adults and adolescents in resource-limited settings: towards universal access. Recommendations for a public health approach 2006 revision. Available: http://www.who.int/hiv/pub/guidelines/artadultguidelines.pdf. Accessed 29 January 2011.
17. Zhang F, Dou Z, Ma Y, Zhao Y, Liu Z, et al. (2009) Five-year outcomes of the China National Free Antiretroviral Treatment Program. Ann Intern Med 151: 241–251.
18. Fox MP, Rosen S (2010) Patient retention in antiretroviral therapy programs up to three years on treatment in sub-Saharan Africa, 2007-2009: systematic review. Trop Med Int Health 15(Suppl 1): 1–15.
19. Brentlinger PE, Torres JV, Martinez PM, Ghee A, Lujan J, et al. (2010) Clinical staging of HIV-related illness in Mozambique: performance of nonphysician clinicians based on direct observation of clinical care and implications for health worker training. J Acquir Immune Defic Syndr 55: 351–355.
20. Keiser O, Tweya H, Boulle A, Braitstein P, Schecter M, et al. (2009) Switching to second-line antiretroviral therapy in resource-limited settings: comparison of programmes with and without viral load monitoring. AIDS 23: 1867–1874.
21. Fairley CK, Permana A, Read TR (2005) Long-term utility of measuring adherence by self-report compared with pharmacy record in a routine clinic setting. HIV Med 6: 366–369.
22. Lucas GM, Chaisson RE, Moore RD (1999) Highly active antiretroviral therapy in a large urban clinic: risk factors for virologic failure and adverse drug reactions. Ann Intern Med 131: 81–87.
23. Grossberg R, Zhang Y, Gross R (2004) A time-to-prescription-refill measure of antiretroviral adherence predicted changes in viral load in HIV. J Clin Epidemiol 57: 1107–1110.
24. Rubin DB (1987) Multiple Imputation for Nonresponse in Surveys. New York: J. Wiley & Sons. 291 p.
25. Royston P (2004) Multiple imputation of missing values. The Stata Journal 4(3): 227–241.
26. Royston P (2005) Multiple imputation of missing values: update. The Stata Journal 5(2): 188–201.
27. Royston P (2005) Multiple imputation of missing values: update of ice. The Stata Journal 5(4): 527–536.
28. White IR, Royston P (2009) Imputing missing covariate values for the Cox model. Stat Med 28: 1982–1998.
29. Grambsch PM, Therneau TM (1994) Proportional hazards tests and diagnostics based on weighted residuals. Biometrika 81: 515–526.
30. Royston P, Carlin JB, White IR (2009) Multiple imputation of missing values: new features for mim. The Stata Journal 9(2): 252–264.
31. Raudenbush SW, Bryk AS (2002) Hierarchical Linear Models: Applications and Data Analysis Methods. Newbury Park, California: Sage. 491 p.
32. Walker AS, Ford D, Gilks CF, Munderi P, Ssali F, et al. (2010) Daily co-trimoxazole prophylaxis in severely immunosuppressed HIV-infected adults in Africa started on combination antiretroviral therapy: an observational analysis of the DART cohort. Lancet 375: 1278–1286.
33. Lowrance D, Makombe S, Harries A, Yu J, Aberle-Grasse J, et al. (2007) Lower early mortality rates among patients receiving antiretroviral treatment at clinics offering cotrimoxazole prophylaxis in Malawi. J Acquir Immune Defic Syndr 46: 56–61.
34. Alemu AW, Sebastian MS (2010) Determinants of survival in adult HIV patients on antiretroviral therapy in Oromiyaa, Ethiopia. Glob Health Action 3: doi: 10.3402/gha.v3i0.5398.
35. Hoffmann CJ, Fielding KL, Charalambous S, Innes C, Chaisson RE, et al. (2010) Reducing mortality with cotrimoxazole preventive therapy at initiation of antiretroviral therapy in South Africa. AIDS 24: 1709–1716.
36. Crepaz N, Hart TA, Marks G (2004) Highly active antiretroviral therapy and sexual risk behavior: a meta-analytic review. JAMA 292: 224–236.
37. Rosen S, Ketlhapile M (2010) Cost of using a patient tracer to reduce loss to follow-up and ascertain patient status in a large antiretroviral therapy program in Johannesburg, South Africa. Trop Med Int Health 15(Suppl 1): 98–104.

38. Yu JK, Chen SC, Wang KY, Chang CS, Makombe SD, et al. (2007) True outcomes for patients on antiretroviral therapy who are "lost to follow-up" in Malawi. Bull World Health Organ 85: 550–554.

39. Geng EH, Bangsberg DR, Musinguzi N, Emenyonu N, Bwana MB, et al. (2009) Understanding reasons for and outcomes of patients lost to follow-up in antiretroviral therapy programs in Africa through a sampling-based approach. J Acquir Immune Defic Syndr 53: 405–411.

40. Geng EH, Emenyonu N, Bwana MB, Glidden DV, Martin JN (2008) Sampling-based approach to determining outcomes of patients lost to follow-up in antiretroviral therapy scale-up programs in Africa. JAMA 300: 506–507.

41. Bisson GP, Gaolathe T, Gross R, Rollins C, Bellamy S, et al. (2008) Overestimates of survival after HAART: implications for global scale-up efforts. PLoS One 3: e1725.

42. Dalal RP, Macphail C, Mqhayi M, Wing J, Feldman C, et al. (2008) Characteristics and outcomes of adult patients lost to follow-up at an antiretroviral treatment clinic in johannesburg, South Africa. J Acquir Immune Defic Syndr 47: 101–107.

43. Weidle PJ, Malamba S, Mwebaze R, Sozi C, Rukundo G, et al. (2002) Assessment of a pilot antiretroviral drug therapy programme in Uganda: patients' response, survival, and drug resistance. Lancet 360: 34–40.

44. Djomand G, Roels T, Ellerbrock T, Hanson D, Diomande F, et al. (2003) Virologic and immunologic outcomes and programmatic challenges of an antiretroviral treatment pilot project in Abidjan, Cote d'Ivoire. AIDS 17(Suppl 3): S5–15.

45. Seyler C, Anglaret X, Dakoury-Dogbo N, Messou E, Toure S, et al. (2003) Medium-term survival, morbidity and immunovirological evolution in HIV-infected adults receiving antiretroviral therapy, Abidjan, Cote d'Ivoire. Antivir Ther 8: 385–393.

46. Wester CW, Kim S, Bussmann H, Avalos A, Ndwapi N, et al. (2005) Initial response to highly active antiretroviral therapy in HIV-1C-infected adults in a public sector treatment program in Botswana. J Acquir Immune Defic Syndr 40: 336–343.

47. Lawn SD, Myer L, Harling G, Orrell C, Bekker LG, et al. (2006) Determinants of mortality and nondeath losses from an antiretroviral treatment service in South Africa: implications for program evaluation. Clin Infect Dis 43: 770–776.

48. Etard JF, Ndiaye I, Thierry-Mieg M, Gueye NF, Gueye PM, et al. (2006) Mortality and causes of death in adults receiving highly active antiretroviral therapy in Senegal: a 7-year cohort study. AIDS 20: 1181–1189.

49. Zachariah R, Fitzgerald M, Massaquoi M, Pasulani O, Arnould L, et al. (2006) Risk factors for high early mortality in patients on antiretroviral treatment in a rural district of Malawi. AIDS 20: 2355–2360.

50. Makombe SD, Harries AD, Yu JK, Hochgesang M, Mhango E, et al. (2007) Outcomes of tuberculosis patients who start antiretroviral therapy under routine programme conditions in Malawi. Int J Tuberc Lung Dis 11: 412–416.

51. Lawn SD, Harries AD, Anglaret X, Myer L, Wood R (2008) Early mortality among adults accessing antiretroviral treatment programmes in sub-Saharan Africa. AIDS 22: 1897–1908.

52. Zhou J, Paton NI, Ditangco R, Chen YM, Kamarulzaman A, et al. (2007) Experience with the use of a first-line regimen of stavudine, lamivudine and nevirapine in patients in the TREAT Asia HIV Observational Database. HIV Med 8: 8–16.

53. Sherr K, Pfeiffer J, Mussa A, Vio F, Gimbel S, et al. (2009) The role of nonphysician clinicians in the rapid expansion of HIV care in Mozambique. J Acquir Immune Defic Syndr 52(Suppl 1): S20–23.

54. Keiser O, Tweya H, Braitstein P, Dabis F, MacPhail P, et al. (2010) Mortality after failure of antiretroviral therapy in sub-Saharan Africa. Trop Med Int Health 15: 251–258.

55. Lawn SD, Myer L, Orrell C, Bekker LG, Wood R (2005) Early mortality among adults accessing a community-based antiretroviral service in South Africa: implications for programme design. AIDS 19: 2141–2148.

56. Moh R, Danel C, Messou E, Ouassa T, Gabillard D, et al. (2007) Incidence and determinants of mortality and morbidity following early antiretroviral therapy initiation in HIV-infected adults in West Africa. AIDS 21: 2483–2491.

57. Jerene D, Endale A, Hailu Y, Lindtjorn B (2006) Predictors of early death in a cohort of Ethiopian patients treated with HAART. BMC Infect Dis 6: 136.

58. World Health Organization. Antiretroviral therapy for HIV infection in adults and adolescents: Recommendations for a public health approach - 2010 revision. Geneva, Switzerland: WHO; (2010) Available: http://whqlibdoc.who.int/publications/2010/9789241599764_eng.pdf. Accessed 29 January 2011.

59. Nachega JB, Hislop M, Dowdy DW, Lo M, Omer SB, et al. (2006) Adherence to highly active antiretroviral therapy assessed by pharmacy claims predicts survival in HIV-infected South African adults. J Acquir Immune Defic Syndr 43: 78–84.

60. May M, Boulle A, Phiri S, Messou E, Myer L, et al. (2010) Prognosis of patients with HIV-1 infection starting antiretroviral therapy in sub-Saharan Africa: a collaborative analysis of scale-up programmes. Lancet 376: 449–457.

61. Donovan C, Massingue J (2007) Illness, death, and macronutrients: adequacy of rural Mozambican household production of macronutrients in the face of HIV/AIDS. Food Nutr Bull 28: S331–338.

62. Coyne-Meyers K, Trombley LE (2004) A review of nutrition in human immunodeficiency virus infection in the era of highly active antiretroviral therapy. Nutr Clin Pract 19: 340–355.

63. Forgacs P, Wengenack NL, Hall L, Zimmerman SK, Silverman ML, et al. (2009) Tuberculosis and trimethoprim-sulfamethoxazole. Antimicrob Agents Chemother 53: 4789–4793.

64. Marazzi MC, Liotta G, Germano P, Guidotti G, Altan AD, et al. (2008) Excessive early mortality in the first year of treatment in HIV type 1-infected patients initiating antiretroviral therapy in resource-limited settings. AIDS Res Hum Retroviruses 24: 555–560.

65. Cornell M, Grimsrud A, Fairall L, Fox MP, van Cutsem G, et al. (2010) Temporal changes in programme outcomes among adult patients initiating antiretroviral therapy across South Africa, 2002-2007. AIDS 24: 2263–2270.

66. Boulle A, Van Cutsem G, Hilderbrand K, Cragg C, Abrahams M, et al. (2010) Seven-year experience of a primary care antiretroviral treatment programme in Khayelitsha, South Africa. AIDS 24: 563–572.

67. Forster M, Bailey C, Brinkhof MW, Graber C, Boulle A, et al. (2008) Electronic medical record systems, data quality and loss to follow-up: survey of antiretroviral therapy programmes in resource-limited settings. Bull World Health Organ 86: 939–947.

68. Brinkhof MW, Pujades-Rodriguez M, Egger M (2009) Mortality of patients lost to follow-up in antiretroviral treatment programmes in resource-limited settings: systematic review and meta-analysis. PLoS One 4: e5790.

69. Allen S, Tice J, Van de Perre P, Serufilira A, Hudes E, et al. (1992) Effect of serotesting with counselling on condom use and seroconversion among HIV discordant couples in Africa. BMJ 304: 1605–1609.

70. Allen S, Meinzen-Derr J, Kautzman M, Zulu I, Trask S, et al. (2003) Sexual behavior of HIV discordant couples after HIV counseling and testing. AIDS 17: 733–740.

71. Kamenga M, Ryder RW, Jingu M, Mbuyi N, Mbu L, et al. (1991) Evidence of marked sexual behavior change associated with low HIV-1 seroconversion in 149 married couples with discordant HIV-1 serostatus: experience at an HIV counselling center in Zaire. AIDS 5: 61–67.

72. United States Centers for Disease Control and Prevention (2003) Advancing HIV prevention: new strategies for a changing epidemic–United States, 2003. JAMA 289: 2493–2495.

73. Gordon CM, Stall R, Cheever LW (2004) Prevention interventions with persons living with HIV/AIDS: challenges, progress, and research priorities. J Acquir Immune Defic Syndr 37(Suppl 2): S53–57.

74. Gordon CM, Forsyth AD, Stall R, Cheever LW (2005) Prevention interventions with persons living with HIV/AIDS: state of the science and future directions. AIDS Educ Prev 17: 6–20.

75. Janssen RS, Valdiserri RO (2004) HIV prevention in the United States: increasing emphasis on working with those living with HIV. J Acquir Immune Defic Syndr 37(Suppl 2): S119–121.

76. Bunnell R, Ekwaru JP, Solberg P, Wamai N, Bikaako-Kajura W, et al. (2006) Changes in sexual behavior and risk of HIV transmission after antiretroviral therapy and prevention interventions in rural Uganda. AIDS 20: 85–92.