Medicine®

OBSERVATIONAL STUDY

OPEN

Risk Factors for Incident Diabetes in a Cohort Taking First-Line Nonnucleoside Reverse Transcriptase Inhibitor-Based Antiretroviral Therapy

Sumanth Karamchand, MBChB, BPharm, Rory Leisegang, MBChB, Michael Schomaker, PhD, Gary Maartens, MBChB, FCP, Lourens Walters, MSc, BA, Michael Hislop, BSc, Joel A. Dave, MBChB, PhD, FCP(SA), Naomi S. Levitt, MBChB, PhD, FCP(SA), and Karen Cohen, MBChB, MMed(Clin Pharm), MSc(Epid)

Abstract: Efavirenz is the preferred nonnucleoside reverse transcriptase inhibitor (NNRTI) in first-line antiretroviral therapy (ART) regimens in low- and middle-income countries, where the prevalence of diabetes is increasing. Randomized control trials have shown mild increases in plasma glucose in participants in the efavirenz arms, but no association has been reported with overt diabetes. We explored the association between efavirenz exposure and incident diabetes in a large Southern African cohort commencing NNRTI-based first-line ART.

Our cohort included HIV-infected adults starting NNRTI-based ART in a private sector HIV disease management program from January 2002 to December 2011. Incident diabetes was identified by the initiation of diabetes treatment. Patients with prevalent diabetes were excluded.

We included 56,298 patients with 113,297 patient-years of follow-up (PYFU) on first-line ART. The crude incidence of diabetes was 13.24 per 1000 PYFU. Treatment with efavirenz rather than nevirapine was associated with increased risk of developing diabetes (hazard ratio 1.27 (95% confidence interval (CI): 1.10–1.46)) in a multivariate analysis adjusting for age, sex, body mass index, baseline CD4 count, viral load, NRTI backbone, and exposure to other diabetogenic medicines. Zidovudine and stavudine exposure were also associated with an increased risk of developing diabetes.

We found that treatment with efavirenz, as well as stavudine and zidovudine, increased the risk of incident diabetes. Interventions to detect and prevent diabetes should be implemented in ART programs, and use of antiretrovirals with lower risk of metabolic complications should be encouraged.

Editor: Giuseppe Lapadula.
Received: July 29, 2015; revised: December 29, 2015; accepted: January 25, 2016.
From the Division of Clinical Pharmacology (SK, RL, GM, KC), Division of Endocrinology, Department of Medicine (JAD, NSL), Center for Infectious Disease Epidemiology and Research, School of Public Health and Family Medicine, University of Cape Town (MS), Aid for AIDS Management (Pty) Limited (MH), Health Intelligence Unit, Medscheme (Pty) Limited (LW), Chronic Disease Initiative for Africa, Cape Town (JAD, NSL), South Africa.
Correspondence: Karen Cohen, Division of Clinical Pharmacology, K 45 Old Main Building Groote Schuur Hospital, University of Cape Town, Observatory, Cape Town, 7925, South Africa (e-mail: karen.cohen@uct.ac.za).
This study was supported by the South African National Research Foundation.
The authors have no conflicts of interest to disclose.
Supplemental Digital Content is available for this article.

ISSN: 0025-7974
DOI: 10.1097/MD.0000000000002844

(*Medicine* 95(9):e2844)

Abbreviations: AfA = Aid for AIDS, AIC = Akaike Information Criterion, ART = antiretroviral therapy, AZT = zidovudine, BMI = body mass index, d4T = stavudine, EFV = efavirenz, HIV = human immunodeficiency virus, HR = hazard ratio, IQR = interquartile range, LMICs = low- and middle-income countries, NNRTI = nonnucleoside reverse transcriptase inhibitor, NRTIs = nucleoside reverse transcriptase inhibitors, NVP = nevirapine, PI = protease inhibitor, PYFU = patient-years of follow-up, VL = viral load, WHO = World Health Organization.

INTRODUCTION

Access to antiretroviral therapy (ART) has considerably reduced morbidity and mortality associated with human immunodeficiency virus (HIV) infection. However, long-term ART is associated with adverse metabolic effects including dysglycemia and new onset diabetes mellitus.[1,2] With the prevalence of noncommunicable diseases, including diabetes, increasing in low- and middle-income countries (LMICs),[3] patients on ART in LMICs face a dual burden of disease.[4]

A number of antiretroviral drugs are known to cause diabetes, including the nucleoside reverse transcriptase inhibitors (NRTIs) stavudine (d4T) and zidovudine (AZT),[2] and the older protease inhibitors (PIs) indinavir[5] and ritonavir.[6,7] Efavirenz, which is now the preferred nonnucleoside reverse transcriptase inhibitor (NNRTI) for first-line ART in LMICs,[8] is associated with slight increases in blood glucose in randomized controlled trials,[9–13] and, in one study conducted by our group.[14] However, there is no good evidence that efavirenz is associated with an increased risk of developing diabetes.

The aim of our study was to investigate the association between efavirenz use and the incidence of diabetes mellitus in a South African cohort of patients on first-line ART.

METHODS

Study Population and Data Source

The study population comprises South African HIV-infected adults enrolled in a private sector HIV disease management program, Aid for AIDS (AfA). The AfA program collects demographic, laboratory, and clinical data on individuals who registered for HIV benefits. Claim data were captured by AfA from the medical insurance fund claim database. These include laboratory, hospitalization, pharmacy, and medical practitioner claims which were submitted to the scheme for processing either: at the time of the service by the provider (eg, pharmacy,

hospitalization) for direct reimbursement or after the service date by the member where the member had already paid the claim. Reimbursement was subject to established AfA protocols, including protocols for ART initiation, change of ART regimen, and the treatment of certain opportunistic infections. No copayment was required for ART, viral load (VL) and CD4 monitoring, and doctor visits.

Despite being a private sector program, AfA standardized guidelines for HIV management, are similar to the World Health Organization (WHO) guidelines for LMICs.[8] Patients were eligible for ART initiation if their CD4 cell count was below 350 cells/μl or they had WHO stage 3 or 4 illness irrespective of the CD4 count. The recommended initial regimen was a combination of 2 NRTIs and an NNRTI. VL and CD4 counts were monitored every 6 months.

Data linkage to the South Africa death registry allowed ascertainment of deaths and date of death, as previously described.[15,16]

Variables and Definitions

We extracted sex, date of birth, weight, height, Republic of South Africa Identity Number, and date of joining the AfA program from the form completed by the doctor on registering the patient with AfA.

We extracted longitudinal results for CD4 count and VL, and all medication claims for antiretrovirals and concomitant medicines. We created a list of diabetogenic drugs using a pharmacology reference textbook[17] and a review[18] (see Appendix 1, http://links.lww.com/MD/A735). We categorized patients as exposed to diabetogenic drugs if they submitted claims for a diabetogenic drug on 2 or more occasions.

We defined the ART start date as the date on which antiretroviral drugs were first dispensed in the AfA program. The ART starting regimen was the regimen dispensed on this date. The baseline CD4 count, VL, and weight were the values measured closest to the date of ART initiation, within the 12-month window before the ART start date. The primary exposure variable of interest was the NNRTI component of the first-line antiretroviral regimen.

Inclusion and Exclusion Criteria

For this study we included AfA-registered patients who initiated a first-line NNRTI-containing ART regimen from January 2002 to December 2011 and were 19 years or older when starting ART. We excluded patients already on antidiabetic medication before starting ART, and patients with missing South Africa identification numbers (as the identification number was used to determine if death occurred by linkage with the South African death registry).

Endpoint

Incident diabetes was defined, using claims data, as the date on which any of the antidiabetic agents available in South Africa (insulins, metformin, sulfonylureas, alpha glucosidase inhibitors, thiazolidinediones, dipeptidyl peptidase-IV inhibitors, glucagon-like-peptide-1 receptor agonists and meglitinides) were initiated. Patients who started and stopped antidiabetic medication in the months around a pregnancy (from 6 months before delivery until 1 month after delivery) were assumed to have diabetes in pregnancy and were therefore not included as incident diabetes.

Imputation of Missing Data

Some data were missing for baseline CD4 count, baseline VL, baseline weight and height (see Table 1). We imputed 5 datasets with the multiple imputation by chained equations (MICE) module in STATA version 13. The imputation model included the following variables: sex, baseline weight, height, age, CD4 count, and VL, death, incident diabetes, and exposure time. CD4 count and VL were actively imputed, and body mass index (BMI) was passively imputed (using actively imputed baseline weight and height). We checked the results of the imputation model by comparing the imputed data with the actual data.[19,20]

Analysis

We collated and prepared the data for statistical analysis using a relational database (Microsoft SQL Server 2008). We used STATA Version 13 (StataCorp LP, College Station, TX) and R version 3.2.1 (R Development Core Team) for statistical analyses.[21] We compared the incidence of diabetes in patients receiving efavirenz-containing regimens versus nevirapine-containing regimens with a Kaplan–Meier plot and a log-rank test.

We explored the association of efavirenz exposure with the hazard of developing diabetes using a multivariate Cox-proportional hazards model. We adjusted for the following variables: age, sex, baseline BMI, baseline CD4 count, baseline VL, exposure to diabetogenic drugs. For the primary analysis ART was included in the model as time-updated NRTI backbone (AZT-containing, d4T-containing, or other NRTI combination) and time-updated NNRTI (efavirenz or nevirapine); and patients were censored when they died, left the medical insurance scheme, switched to PI-based ART, or reached the end of the study period. We performed a secondary analysis exploring incident diabetes within the first ART regimen only. For this analysis we included 2 additional reasons for censoring: NRTI or NNRTI substitution. We explored the effect of including calendar year in the multivariate model. We performed the following sensitivity analyses: we controlled for the competing risk of death, we constructed a model excluding patients virologically suppressed at baseline.

The proportional hazards assumption was verified by testing interaction effects of analysis time with baseline variables ($\alpha = 0.05$), and graphically in each imputed dataset via log–log plots, amongst others. We performed model selection using the Akaike Information Criterion (AIC) after multiple imputation.[19,22–24]

Ethics

The study protocol was reviewed and approved by the University of Cape Town Faculty of Health Sciences Human Research Ethics committee.

RESULTS

Between January 2002 and June 2011, 62,467 patients commenced ART in the AfA program, of whom 56,298 patients met our inclusion and exclusion criteria (Figure 1) and were included in the analysis. The demographic and clinical characteristics of patients are given in Table 1. Results from the multiple imputations for missing covariates are shown in Table 2. Median follow-up was 1.56 years (interquartile range (IQR): 0.71–2.79 years), 21.7% of patients were followed up for 3 or more years.

We identified new onset diabetes in 1500 (2.66%) patients over 113,297 patient-years of follow-up (PYFU), giving a crude incidence of 13.24 cases per 1000 PYFU (Figure 2). There were 17 pregnancy-associated diabetic events, which were not included as cases of incident diabetes. Exposure to diabetogenic

TABLE 1. Cohort Description

	Whole Cohort	Efavirenz-Containing ART	Nevirapine-Containing ART
Number of patients	56,298	46,666	9632
Age (yr)			
Median (IQR)	38.14 (33.15–44.26)	39.1 (34.1–45.17)	34.05 (29.99–38.65)
Sex			
Male	20,224 (35.92)	18,822 (40.33%)	1402 (14.55%)
Female	36,074 (64.08)	27,844 (59.67%)	8230 (85.44%)
Race			
Asian	148 (0.26%)	126 (0.27%)	22 (0.23%)
Black	53,270 (94.62%)	44,179 (94.67%)	9091 (94.38%)
Mixed	768 (1.36%)	628 (1.35%)	140 (1.45%)
White	989 (1.76%)	825 (1.77%)	164 (1.7%)
Not reported	1123 (1.99%)	908 (1.95%)	215 (2.23%)
Nucleoside reverse transcriptase inhibitor (initial regimen containing)			
Zidovudine	26,917 (47.81%)	25,329 (54.28%)	1588 (16.49%)
Stavudine	22,465 (39.9%)	16,002 (34.29%)	6463 (67.1%)
Other	6916 (12.28%)	5335 (11.43%)	1581 (16.41%)
Exposure to other diabetogenic drugs	22,780 (40.46%)	19,137 (41.01%)	3643 (37.82%)
Baseline height (m)			
Median (IQR)	165 (160–170)	165 (160–170)	163 (158–168)
Missing	19,033 (33.81%)	15,390 (32.98%)	3643 (37.82%)
Baseline weight (kg)			
Median (IQR)	68 (60–79)	68 (60–79)	70 (62–81)
Missing	15,042 (26.72%)	12,349 (26.46%)	2693 (27.96%)
Baseline body mass index (kg/m^2)			
Median (IQR)	25.06 (21.97–29.00)	24.82 (21.80–28.69)	26.52 (23.14–30.49)
Missing	23,178 (41.17%)	18,776 (40.23%)	4402 (45.7%)
Baseline CD4 count (cells/μl)			
Median (IQR)	181 (88–280)	176 (81–277)	201 (121–306)
Missing	948 (1.68%)	769 (1.65%)	179 (1.86%)
Baseline log viral load (copies/ml)			
Median (IQR)	4.78 (3.66–5.37)	4.87 (3.86–5.43)	4.35 (2.59–5.03)
Missing	3464 (6.15%)	2845 (6.1%)	619 (6.43%)
Follow-up time (yr)			
Median (IQR)	1.56 (0.71–2.79)	1.50 (0.67–2.69)	1.90 (0.93–3.34)
Patient years	113,297	89,915	23,382
3 or more years of NNRTI exposure	21.70%	29.86%	20.66%
Reason for censoring			
Diabetes	1500 (2.66%)	1257 (2.69%)	243 (2.52%)
Switch to PI	3706 (6.58%)	2778 (5.95%)	928 (9.63%)
Study end	40,785 (72.44%)	34,186 (73.26%)	6599 (68.51%)
Death	1774 (3.15%)	1488 (3.19%)	286 (2.97%)
Left scheme	8533 (15.16%)	6957 (14.91%)	1576 (16.36%)
Crude diabetes incidence			
Events/1000 patient years	13.24	13.98	10.39

ART = antiretroviral therapy, IQR = interquartile range, NNRTI = nonnucleoside reverse transcriptase inhibitor, PI = protease inhibitor.

medicines occurred in 19,137 (41.01%) of patients taking efavirenz and 3643 (37.82%) of patients taking nevirapine. The Kaplan Meier analysis of incident diabetes in efavirenz versus nevirapine-containing regimens is shown in Figure 2.

The results of the Cox proportional hazard regression analyses, including univariate and multivariate analyses and model selection, are shown in Table 3. Efavirenz-containing ART was associated with a higher risk of developing new-onset diabetes than nevirapine-containing ART, adjusted hazard ratio (HR) 1.27 (95% confidence interval (CI): 1.10–1.46). Zidovudine and stavudine-containing NRTI backbones, older age at baseline, elevated baseline BMI, and exposure to diabetogenic medication were also associated with an increased risk of developing diabetes. We found no association between baseline CD4 and an increased risk of diabetes.

The results of the Cox proportional hazard regression analyses where we censored patients at the time of first drug switch are shown in Table 4; reasons for censoring for this analysis are shown in Supplementary Table 1, http://links.lww. com/MD/A736. The estimated HRs of the variables remained similar after model averaging, confirming the stability of our findings.[25] These findings did not differ from the

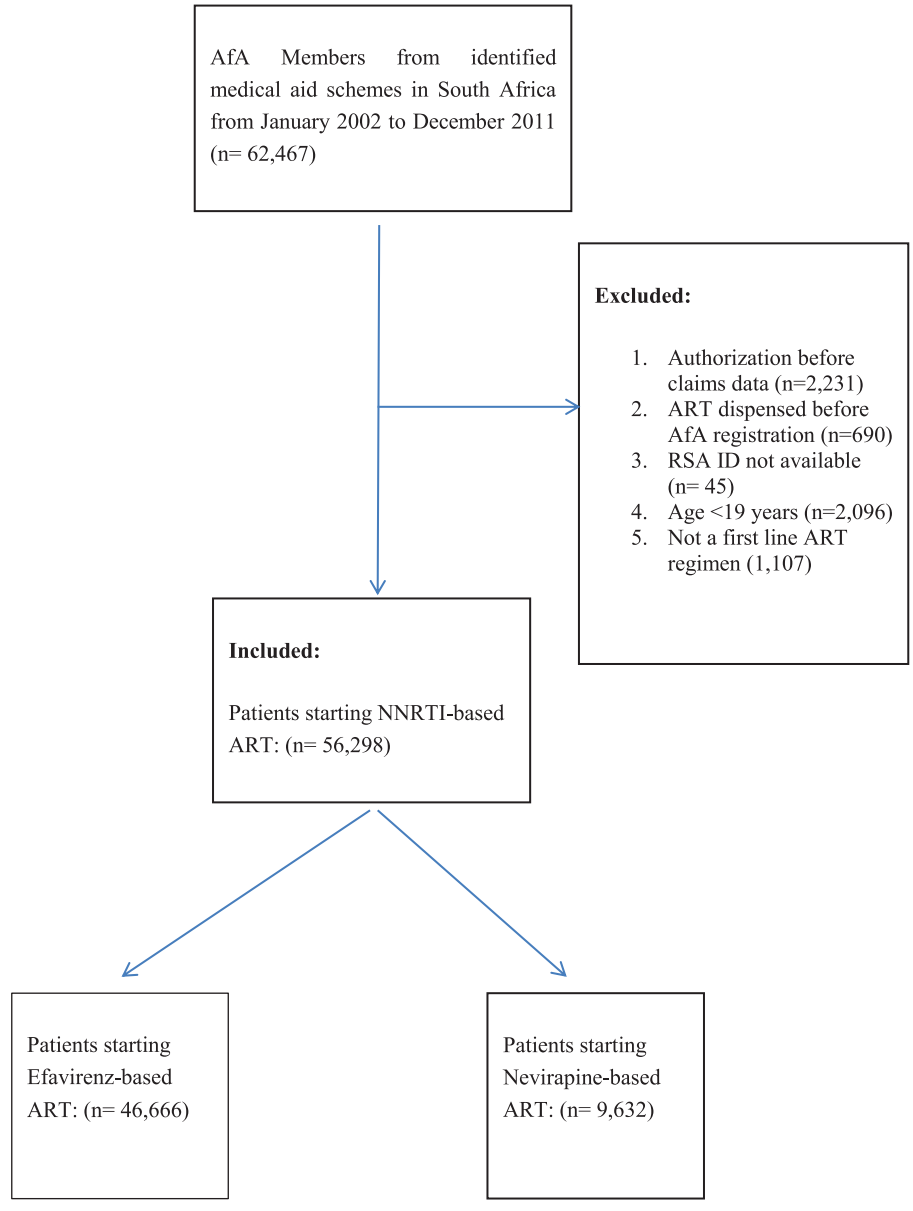

FIGURE 1. Participant selection and exclusion criteria.

regression model with updated regimen (censoring at the point of change to second line regimens). Adding calendar year to the model did not change the associations observed (Supplementary Table 2, http://links.lww.com/MD/A736). Findings did not change when we accounted for competing risk (Supplementary Table 3, http://links.lww.com/MD/A736). Excluding patients with suppressed baseline VLs attenuated the effect of efavirenz on incident diabetes somewhat: adjusted HR 1.16; 95% CI: 0.99–1.36 (Supplementary Table 4, http://links.lww.com/MD/A736).

DISCUSSION

We found efavirenz use to be associated with a significantly higher incidence of diabetes than nevirapine in a large cohort of South African patients. To the best of our knowledge, this is the first cohort study to show an increased risk of diabetes from efavirenz use in first-line ART. We also found that the NRTIs stavudine and zidovudine were associated with an increased incidence of diabetes. These findings have important implications for LMICs, which are facing a burgeoning diabetes epidemic, as efavirenz is the preferred NNRTI in first-line ART, zidovudine is recommended in second line ART, and many people are still taking stavudine, even though it is no longer recommended by the WHO.[8]

HIV-infected patients have an estimated 4-fold greater relative risk of developing diabetes than the HIV-uninfected population.[26] We found a crude incidence of diabetes of 13.24 per 1000 PYFU, which is at the upper end of the range reported from cohort studies in high income countries (4.2–14.1 per 1000 PYFU).[2,26–28] Factors contributing to the increased risk of diabetes in people with HIV include insulin resistance due to

TABLE 2. Results of Imputation for Baseline Variables With Missing Data

	Total Cohort (n = 56,298)	Efavirenz-Containing ART (n = 46,666)	Nevirapine-Containing ART (n = 9632)
Variable	**Mean (95% CI)**	**Mean (95% CI)**	**Mean (95% CI)**
Height (m)			
Before imputation	164.93 (164.84–165.03)	165.25 (165.18–165.31)	163.14 (162.92–163.35)
After imputation	164.74 (164.64–164.85)	165.14 (165.05–165.23)	163.07 (162.88–163.26)
Weight (kg)			
Before imputation	70.34 (70.18–70.51)	69.88 (69.72–70.03)	72.81 (72.39–73.23)
After imputation	70.14 (69.97–70.30)	69.80 (69.62–69.97)	71.59 (71.18–72.00)
CD4 count (cells/μl)			
Before imputation	215.82 (214.27–217.38)	208.77 (207.64–209.90)	254.44 (251.66–257.22)
After imputation	215.39 (214.41–216.38)	207.18 (206.12–208.25)	250.26 (247.83–252.69)
Viral load (copies/ml)			
Before imputation	4.31 (4.30–4.33)	4.39 (4.38–4.40)	3.88 (3.86–3.90)
After imputation	4.32 (4.31–4.33)	4.38 (4.37–4.39)	3.92 (3.90–3.94)
Body mass index (kg/m^2)			
Before imputation	25.95 (25.89–26.01)	25.67 (25.62–25.72)	27.44 (27.31–27.56)
After imputation	25.93 (25.87–25.99)	25.68 (25.61–25.75)	27.00 (26.86–27.14)

ART = antiretroviral therapy, CI = confidence interval.

the chronic inflammatory response to HIV infection,[29] which persists despite effective ART[29,30] and the effects of certain antiretroviral drugs. Our finding that the NRTIs stavudine and zidovudine were both associated with an increased incidence of diabetes has previously been reported.[2,31] NRTI's inhibit the enzyme DNA polymerase-γ, responsible for mitochondrial replication. The dysregulation of mitochondrial function in different compartments of the body results in various clinical manifestations of NRTI toxicity, including insulin resistance and diabetes.[32–34] In a prior cross sectional study we found an increased risk of dysglycemia in South African patients taking efavirenz compared with those taking nevirapine, but there were insufficient numbers of cases of diabetes for analysis.[14] A small case–control study from Botswana suggested an association between efavirenz use and diabetes.[35] Randomized controlled trials showed significantly higher serum glucose concentrations in participants in the efavirenz arms than the following comparator antiretroviral drugs: nevirapine,[13] abacavir,[13] atazanavir,[11] atazanavir–ritonavir,[9] and raltegravir.[12]

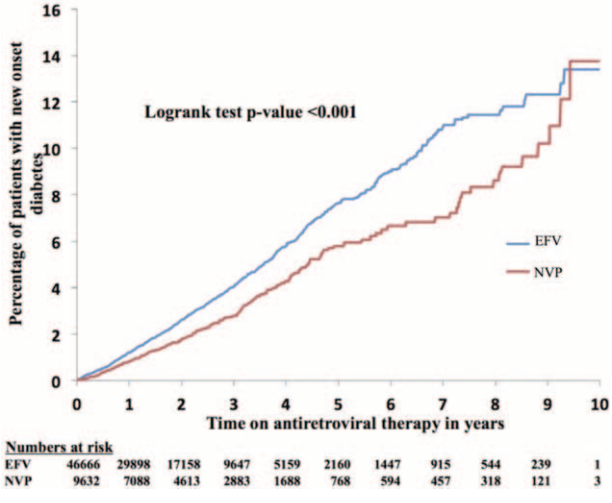

FIGURE 2. Kaplan–Meier analysis of incident diabetes.

The mechanism by which efavirenz mediates insulin resistance and diabetes is unknown. Possible mechanisms include mitochondrial toxicity[36] and toxic effects on adipocytes and increased rates of lipoatrophy.[37,38] Efavirenz causes hepatic mitochondrial toxicity[36] and induces hepatocyte endoplasmic reticulum stress leading to activation of the unfolded protein response, and apoptosis.[39,40] Efavirenz mediates mitochondrial toxicity via various mechanisms. Firstly, efavirenz directly inhibits Complex I of the electron transport chain, resulting in a markedly reduced mitochondrial transmembrane potential, thus compromising oxidative phosphorylation and ATP generation.[41–43] Secondly, efavirenz reduces complex IV (COIV) mRNA (a marker gene of mitochondrial function), and impairs mitochondrial function in adipocytes.[44] Furthermore, efavirenz-associated mitochondrial dysregulation in adipose tissue causes impaired adipogenesis, increased lipolysis, and release of free fatty acids and inflammatory cytokines.[44] The increased release of fatty acids due to adipocyte mitochondrial toxicity are thought to impair muscle and liver insulin sensitivity, leading to insulin resistance and diabetes mellitus.[45–51] In addition to its mitochondrial toxicity, efavirenz has been shown to reduce the secretion of adiponectin (an insulin-sensitizing, antidiabetic adipokine) by adipocytes.[44] We hypothesize that impairment of mitochondrial bioenergetics and toxicity to adipocytes contributes to the development of diabetes in patients on efavirenz. By contrast, nevirapine does not appear to exert mitochondrial toxicity.[37,44]

Our group have demonstrated a positive correlation between plasma efavirenz concentrations and both fasting and 2-hour glucose concentrations after oral glucose tolerance tests in South African patients.[52] People of African origin are more likely to be genotypic ''slow metabolizers'' of efavirenz, which results in elevated efavirenz plasma concentrations, than people of European descent (20% and 3%, respectively).[53] Therefore efavirenz may have a larger diabetogenic effect in

TABLE 3. Univariate, Multivariate, and Model Selection Results for the Cox Regression Model of Associations With Incident Diabetes

		Univariate		Multivariate		
Variable	**Category**	**HR (95% CI)**	***P***	**HR (95% CI)**	***P***	**Model Selection (AIC)**
Nonnucleoside reverse transcriptase inhibitor	Efavirenz	1.40 (1.22–1.60)	<0.001	1.27 (1.10–1.47)	0.001	1.27 (1.10–1.46)
	Nevirapine	Referent		Referent		Referent
Nucleoside reverse transcriptase inhibitor	Zidovudine	1.30 (1.15–1.46)	<0.001	1.35 (1.19–1.52)	<0.001	1.37 (1.21–1.54)
	Stavudine	1.53 (1.32–1.78)	<0.001	1.60 (1.38–1.87)	<0.001	1.64 (1.41–1.91)
	Other	Referent		Referent		Referent
Exposure to other diabetogenic drugs		1.68 (1.51–1.86)	<0.001	1.53 (1.37–1.70)	<0.001	1.53 (1.38–1.71)
Baseline age (yr)	19–24	0.35 (0.20–0.60)	<0.001	0.47 (0.27–0.81)	0.007	0.46 (0.27–0.80)
	25–34	0.64 (0.56–0.73)	<0.001	0.71 (0.62–0.82)	<0.001	0.71 (0.62–0.81)
	35–44	Referent		Referent		Referent
	45–54	1.50 (1.33–1.70)	<0.001	1.38 (1.21–1.56)	<0.001	1.36 (1.20–1.54)
	≥55	1.86 (1.50–2.31)	<0.001	1.64 (1.32–2.04)	<0.001	1.57 (1.26–1.95)
Sex	Male	1.41 (1.27–1.56)	<0.001	1.47 (1.32–1.64)	<0.001	1.44 (1.29–1.61)
	Female	Referent		Referent		Referent
Baseline body mass index (BMI) quartile (kg/m/m)	10–17	0.38 (0.23–0.63)	0.001	0.33 (0.19–0.56)	0.001	0.32 (0.19–0.55)
	18–24	0.65 (0.58–0.74)	<0.001	0.61 (0.53–0.69)	<0.001	0.60 (0.53–0.69)
	25–34	Referent		Referent		Referent
	35+	1.45 (1.16–1.81)	0.002	1.58 (1.26–1.97)	<0.001	1.58 (1.27–1.97)
Baseline CD4 count (cells/μl)	0–199	1.04 (0.92–1.17)	0.534	1.08 (0.95–1.23)	0.220	
	200–349	Referent		Referent		Excluded by AIC
	350+	1.25 (1.06–1.47)	0.007	1.10 (0.91–1.34)	0.324	
Baseline viral load (copies/ml)	0–999	1.31 (1.14–1.51)	<0.001	1.24 (1.05–1.47)	0.011	1.28 (1.11–1.47)
	1000–99,999	Referent		Referent		Referent
	100,000–999,999	1.07 (0.95–1.21)	0.261	1.03 (0.91–1.16)	0.674	1.03 (0.91–1.17)
	≥1,000,000	1.15 (0.89–1.48)	0.285	1.15 (0.89–1.49)	0.278	1.14 (0.89–1.48)

Drug switches within first-line regimen included in the model, with censoring at switch to second line therapy. All results are based on multiple imputations.

AIC = Akaike Information Criterion, CI = confidence interval, HR = hazard ratio.

Africans, which may explain why, in contrast with our findings, studies from high income countries have not found an association between efavirenz and diabetes.

Our finding that increasing age, and male sex were associated with an increased risk of diabetes is consistent with findings of other studies.[2,29,54–59] We could not show any association between baseline CD4 count and diabetes, which is similar to that reported by a French cohort,[27] but other studies have found an increased risk of diabetes with lower CD4 counts.[29,54] We found an association between the lowest stratum of baseline VL and an increased relative risk for developing diabetes, but no association with higher VL strata. The French cohort reported no association between VL and diabetes.[27] In contrast, other studies have found an association between high VLs and diabetes.[29,58] The association we observed between low baseline VLs and increased risk of diabetes may be attributed to the inclusion of patients already on undisclosed ART on entry to the AfA program.

Our study has limitations. We had missing baseline data, notably of BMI. However, we imputed missing data, which are known to be superior to using complete case analysis.[60,61] ART exposure may have occurred before commencing ART within the AfA program and there were 18.8% of patients with a suppressed VL at baseline, which is likely due to undisclosed ART use. We identified incident diabetes based on initiation of diabetes therapy as the results of plasma glucose or glycated hemoglobin are not captured in the database. We will therefore have missed cases of diabetes that were only treated with lifestyle modifications. As patients were not routinely screened for diabetes at the time of ART initiation, some patients may have entered the cohort with undiagnosed diabetes. We could not adjust for weight changes during follow-up in our analyses, as weight is only recorded at baseline. A strength of our study is that we adjusted for the concurrent use of diabetogenic medication in the multivariate model, which was associated with a 53% increase in the relative risk of diabetes.

In conclusion, we found that exposure to efavirenz, stavudine, and zidovudine was associated with an increased incidence of diabetes. Although the increased risk of diabetes with these antiretrovirals was relatively modest, the large African patient population exposed to ART for prolonged periods means that our finding has important public health implications. While screening for diabetes should be increased in people on long-term ART, consideration should be given to using antiretrovirals with less risk of metabolic complications. Further studies to confirm the association of efavirenz and risk of diabetes should be conducted in LMICs, and the molecular mechanisms of efavirenz-induced dysglycemia need further investigation.

TABLE 4. Univariate, Multivariate, and Model Selection Results for the Cox Regression Model of Associations With Incident Diabetes (Censored at First Ever Drug Switch)

		Univariate		Multivariate (Rubins Rule)		Model Selection (AIC)
Variable	**Category**	**HR (95% CI)**	***P***	**HR (95% CI)**	***P***	**HR (95% CI)**
Nonnucleoside reverse transcriptase inhibitor	Efavirenz	1.38 (1.18–1.60)	<0.001	1.26 (1.07–1.49)	0.005	1.33 (1.13–1.56)
	Nevirapine	Referent		Referent		Referent
Nucleoside reverse transcriptase inhibitor	Zidovudine	1.34 (1.17–1.54)	<0.001	1.39 (1.21–1.60)	<0.001	1.57 (1.37–1.80)
	Stavudine	1.62 (1.37–1.90)	<0.001	1.67 (1.41–1.98)	<0.001	1.96 (1.66–2.32)
	Other	Referent		Referent		Referent
Exposure to other diabetogenic drugs		1.68 (1.50–1.88)	<0.001	1.52 (1.36–1.71)	<0.001	1.50 (1.34–1.69)
Baseline age (yr)	19–24	0.39 (0.22–0.69)	0.001	0.53 (0.30–0.94)	0.031	0.52 (0.29–0.93)
	25–34	0.63 (0.55–0.73)	<0.001	0.71 (0.61–0.82)	<0.001	0.70 (0.60–0.81)
	35–44	Referent		Referent		Referent
	45–54	1.49 (1.30–1.70)	<0.001	1.37 (1.20–1.57)	<0.001	1.36 (1.19–1.56)
	≥55	1.89 (1.50–2.37)	<0.001	1.69 (1.34–2.13)	<0.001	1.61 (1.28–2.03)
Sex	Male	1.40 (1.25–1.56)	<0.001	1.48 (1.31–1.67)	<0.001	1.47 (1.30–1.66)
	Female	Referent		Referent		Referent
Baseline body mass index (kg/m^2)	10–17	0.40 (0.23–0.71)	0.004	0.35 (0.19–0.63)	0.002	0.35 (0.19–0.62)
	18–24	0.65 (0.53–0.81)	0.001	0.61 (0.48–0.76)	0.001	0.61 (0.48–0.76)
	25–34	Referent		Referent		Referent
	35+	1.44 (1.14–1.82)	0.004	1.57 (1.24–1.98)	<0.001	1.58 (1.25–1.99)
						Excluded by AIC
Baseline CD4 count (cells/μl)	0–199	1.06 (0.93–1.21)	0.415	1.11 (0.97–1.27)	0.136	
	200–349	Referent		Referent		
	350+	1.32 (1.11–1.57)	0.001	1.11 (0.91–1.36)	0.307	
Baseline viral load (copies/ml)	0–999	1.35 (1.17–1.57)	<0.001	1.27 (1.06–1.53)	0.010	1.27 (1.09–1.48)
	1000–99,999	Referent		Referent		Referent
	100,000–999,999	1.03 (0.90–1.18)	0.653	0.99 (0.86–1.13)	0.841	1.00 (0.87–1.14)
	≥1,000,000	1.15 (0.88–1.50)	0.311	1.15 (0.88–1.50)	0.320	1.15 (0.88–1.50)

All results are based on multiple imputations.
AIC = Akaike Information Criterion, CI = confidence interval, HR = hazard ratio.

REFERENCES

1. Samaras K. Prevalence and pathogenesis of diabetes mellitus in HIV-1 infection treated with combined antiretroviral therapy. *J Acquir Immune Defic Syndr.* 2009;50:499–505.
2. De Wit S, Sabin CA, Weber R, et al. Incidence and risk factors for new-onset diabetes in HIV-infected patients: the Data Collection on Adverse Events of Anti-HIV Drugs (D:A:D) study. *Diabetes Care.* 2008;31:1224–1229.
3. Mbanya JC, Motala AA, Sobngwi E, et al. Diabetes in sub-Saharan Africa. *Lancet.* 2010;375:2254–2266.
4. Levitt NS, Steyn K, Dave J, et al. Chronic noncommunicable diseases and HIV-AIDS on a collision course: relevance for health care delivery, particularly in low-resource settings—insights from South Africa. *Am J Clin Nutr.* 2011;94:1690S–1696S.
5. Murata H, Hruz PW, Mueckler M. Indinavir inhibits the glucose transporter isoform Glut4 at physiologic concentrations. *AIDS.* 2002;16:859–863.
6. Taylor SA, Lee GA, Pao VY, et al. Boosting dose ritonavir does not alter peripheral insulin sensitivity in healthy HIV-seronegative volunteers. *J Acquir Immune Defic Syndr.* 2010;55:361–364.
7. Vyas AK, Koster JC, Tzekov A, et al. Effects of the HIV protease inhibitor ritonavir on GLUT4 knock-out mice. *J Biol Chem.* 2010;285:36395–36400.
8. Consolidated Guidelines on the Use of Antiretroviral Drugs for Treating and Preventing HIV Infection: Recommendations for a Public Health Approach. Geneva, Switzerland: World Health Organization (WHO); 2013. ISBN 9789241505727.
9. Erlandson KM, Kitch D, Tierney C, et al. Impact of randomized antiretroviral therapy initiation on glucose metabolism. *AIDS.* 2014;28:1451–1461.
10. Fisac C, Fumero E, Crespo M, et al. Metabolic benefits 24 months after replacing a protease inhibitor with abacavir, efavirenz or nevirapine. *AIDS.* 2005;19:917–925.
11. Jemsek JG, Arathoon E, Arlotti M, et al. Body fat and other metabolic effects of atazanavir and efavirenz, each administered in combination with zidovudine plus lamivudine, in antiretroviral-naive HIV-infected patients. *Clin Infect Dis.* 2006;42:273–280.
12. Lennox JL, Dejesus E, Berger DS, et al. Raltegravir versus efavirenz regimens in treatment-naive HIV-1-infected patients: 96-week efficacy, durability, subgroup, safety, and metabolic analyses. *J Acquir Immune Defic Syndr.* 2010;55:39–48.
13. Martinez E, Arnaiz JA, Podzamczer D, et al. Substitution of nevirapine, efavirenz, or abacavir for protease inhibitors in patients with human immunodeficiency virus infection. *N Engl J Med.* 2003;349:1036–1046.
14. Dave JA, Lambert EV, Badri M, et al. Effect of nonnucleoside reverse transcriptase inhibitor-based antiretroviral therapy on

dysglycemia and insulin sensitivity in South African HIV-infected patients. *J Acquir Immune Defic Syndr.* 2011;57:284–289.

15. Boulle A, Van Cutsem G, Hilderbrand K, et al. Seven-year experience of a primary care antiretroviral treatment programme in Khayelitsha, South Africa. *AIDS.* 2010;24:563–572.
16. Schomaker M, Gsponer T, Estill J, et al. Non-ignorable loss to follow-up: correcting mortality estimates based on additional outcome ascertainment. *Stat Med.* 2014;33:129–142.
17. Davis S. Diabetogenic drugs: treating chronic conditions to minimise new onset diabetes. *SAPJ.* 2010:22–27.
18. Brunton L, Lazo J, Parker K (Eds): *Goodman & Gilman's the Pharmacological Basis of Therapeutics*. New York: McGraw-Hill; 2005.
19. Schomaker M, Heumann C. Model selection and model averaging after multiple imputation. *Comput Stat Data An.* 2014;71:758–770.
20. Honaker J, King G, Blackwell M. Amelia II: a program for missing data. *J Stat Softw.* 2011;45:1–47.
21. R Core Team (2013). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL http://www.R-project.org/.
22. Bozdogan H. Model selection and Akaike's Information Criterion (AIC): the general theory and its analytical extensions. *Psychometrika.* 1987;52:345–370.
23. Schomaker M, Wan ATK, Heumann C. Frequentist model averaging with missing observations. *Comput Stat Data An.* 2010;54:3336–3347.
24. Volinsky CT, Madigan D, Raftery AE, et al. Bayesian model averaging in proportional hazard models. Assessing the risk of a stroke. *Appl Stat.* 1997;46:433–448.
25. Burnham K, Anderson D. Model Selection and Multimodel Inference. A Practical Information-Theoretic Approach. New York: Springer; 2002
26. Brown TT, Li X, Cole SR, et al. Cumulative exposure to nucleoside analogue reverse transcriptase inhibitors is associated with insulin resistance markers in the Multicenter AIDS Cohort Study. *AIDS.* 2005;19:1375–1383.
27. Capeau J, Bouteloup V, Katlama C, et al. Ten-year diabetes incidence in 1046 HIV-infected patients started on a combination antiretroviral treatment. *AIDS.* 2012;26:303–314.
28. Petoumenos K, Worm SW, Fontas E, et al. Predicting the short-term risk of diabetes in HIV-positive patients: the Data Collection on Adverse Events of Anti-HIV Drugs (D:A:D) study. *J Int AIDS Soc.* 2012;15:17426.
29. Brown TT, Tassiopoulos K, Bosch RJ, et al. Association between systemic inflammation and incident diabetes in HIV-infected patients after initiation of antiretroviral therapy. *Diabetes Care.* 2010;33:2244–2249.
30. Brigham EP, Patil SP, Jacobson LP, et al. Association between systemic inflammation and obstructive sleep apnea in men with or at risk for HIV infection. *Antivir Ther.* 2014;19:725–733.
31. Brown TT, Cole SR, Li X, et al. Antiretroviral therapy and the prevalence and incidence of diabetes mellitus in the multicenter AIDS cohort study. *Archiv Intern Med.* 2005;165:1179–1184.
32. Blumer RM, van Vonderen MG, Sutinen J, et al. Zidovudine/lamivudine contributes to insulin resistance within 3 months of starting combination antiretroviral therapy. *AIDS.* 2008;22:227–236.
33. Brinkman K, Kakuda TN. Mitochondrial toxicity of nucleoside analogue reverse transcriptase inhibitors: a looming obstacle for long-term antiretroviral therapy? *Curr Opin Infect Dis.* 2000;13:5–11.
34. Kakuda TN. Pharmacology of nucleoside and nucleotide reverse transcriptase inhibitor-induced mitochondrial toxicity. *Clin Ther.* 2000;22:685–708.
35. Moyo D, Tanthuma G, Cary MS, et al. Cohort study of diabetes in HIV-infected adult patients: evaluating the effect of diabetes mellitus on immune reconstitution. *Diabetes Res Clin Pract.* 2014;103:e34–e36.
36. Blas-Garcia A, Apostolova N, Ballesteros D, et al. Inhibition of mitochondrial function by efavirenz increases lipid content in hepatic cells. *Hepatology.* 2010;52:115–125.
37. El Hadri K, Glorian M, Monsempes C, et al. In vitro suppression of the lipogenic pathway by the nonnucleoside reverse transcriptase inhibitor efavirenz in 3T3 and human preadipocytes or adipocytes. *J Biol Chem.* 2004;279:15130–15141.
38. Perez-Molina JA, Domingo P, Martinez E, et al. The role of efavirenz compared with protease inhibitors in the body fat changes associated with highly active antiretroviral therapy. *J Antimicrob Chemother.* 2008;62:234–245.
39. Apostolova N, Gomez-Sucerquia LJ, Alegre F, et al. ER stress in human hepatic cells treated with efavirenz: mitochondria again. *J Hepatol.* 2013;59:780–789.
40. Polo M, Alegre F, Funes HA, et al. Mitochondrial (dys)function—a factor underlying the variability of efavirenz-induced hepatotoxicity? *Br J Pharmacol.* 2015;172:1713–1727.
41. Apostolova N, Gomez-Sucerquia LJ, Moran A, et al. Enhanced oxidative stress and increased mitochondrial mass during efavirenz-induced apoptosis in human hepatic cells. *Br J Pharmacol.* 2010;160:2069–2084.
42. Karamchand L, Dawood H, Chuturgoon AA. Lymphocyte mitochondrial depolarization and apoptosis in HIV-1-infected HAART patients. *J Acquir Immune Defic Syndr.* 2008;48:381–388.
43. Pilon AA, Lum JJ, Sanchez-Dardon J, et al. Induction of apoptosis by a nonnucleoside human immunodeficiency virus type 1 reverse transcriptase inhibitor. *Antimicrob Agents Chemother.* 2002;46:2687–2691.
44. Gallego-Escuredo JM, Del Mar Gutierrez M, Diaz-Delfin J, et al. Differential effects of efavirenz and lopinavir/ritonavir on human adipocyte differentiation, gene expression and release of adipokines and pro-inflammatory cytokines. *Curr HIV Res.* 2010;8:545–553.
45. Arner P. Insulin resistance in type 2 diabetes: role of fatty acids. *Diabetes Metab Res Rev.* 2002;18(Suppl. 2):S5–S9.
46. Hadigan C, Borgonha S, Rabe J, et al. Increased rates of lipolysis among human immunodeficiency virus-infected men receiving highly active antiretroviral therapy. *Metab Clin Exp.* 2002;51:1143–1147.
47. Hruz PW. Molecular mechanisms for altered glucose homeostasis in HIV infection. *Am J Infect Dis.* 2006;2:187–192.
48. Johnson JA, Albu JB, Engelson ES, et al. Increased systemic and adipose tissue cytokines in patients with HIV-associated lipodystrophy. *Am J Physiol Endocrinol Metab.* 2004;286:E261–E271.
49. Kim JA, Wei Y, Sowers JR. Role of mitochondrial dysfunction in insulin resistance. *Circ Res.* 2008;102:401–414.
50. Krebs M, Roden M. Molecular mechanisms of lipid-induced insulin resistance in muscle, liver and vasculature. *Diabetes Obes Metab.* 2005;7:621–632.
51. Sivitz WI, Yorek MA. Mitochondrial dysfunction in diabetes: from molecular mechanisms to functional significance and therapeutic opportunities. *Antioxid Redox Signal.* 2010;12:537–577.
52. Sinxadi P, McIlleron H, Dave JA. Plasma Efavirenz Concentrations are associated with Lipid and Glucose Concentrations. *Medicine (Baltimore).* 2016;95:e2385.
53. Haas DW, Ribaudo HJ, Kim RB, et al. Pharmacogenetics of efavirenz and central nervous system side effects: an Adult AIDS Clinical Trials Group study. *AIDS.* 2004;18:2391–2400.
54. Galli L, Salpietro S, Pellicciotta G, et al. Risk of type 2 diabetes among HIV-infected and healthy subjects in Italy. *Eur J Epidemiol.* 2012;27:657–665.

55. Brambilla AM, Novati R, Calori G, et al. Stavudine or indinavir-containing regimens are associated with an increased risk of diabetes mellitus in HIV-infected individuals. *AIDS.* 2003;17: 1993–1995.

56. Butt AA, McGinnis K, Rodriguez-Barradas MC, et al. HIV infection and the risk of diabetes mellitus. *AIDS.* 2009;23:1227–1234.

57. Tien PC, Schneider MF, Cole SR, et al. Antiretroviral therapy exposure and incidence of diabetes mellitus in the Women's Interagency HIV Study. *AIDS.* 2007;21:1739–1745.

58. Ledergerber B, Furrer H, Rickenbach M, et al. Factors associated with the incidence of type 2 diabetes mellitus in HIV-infected participants in the Swiss HIV Cohort Study. *Clin Infect Dis.* 2007;45:111–119.

59. El-Sadr WM, Mullin CM, Carr A, et al. Effects of HIV disease on lipid, glucose and insulin levels: results from a large antiretroviral-naive cohort. *HIV Med.* 2005;6:114–121.

60. Rubin DB. Multiple imputation after 18+ years. *J Am Stat Assoc.* 1996;91:473–489.

61. van der Heijden GJ, Donders AR, Stijnen T, et al. Imputation of missing values is superior to complete case analysis and the missing-indicator method in multivariable diagnostic research: a clinical example. *J Clin Epidemiol.* 2006;59:1102–1109.