

The ASSURE study: HIV-1 suppression is maintained with bone and renal biomarker improvement 48 weeks after ritonavir discontinuation and randomized switch to abacavir/lamivudine + atazanavir*

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Objectives

HIV treatment guidelines endorse switching or simplification of antiretroviral therapy in therapy-experienced patients with suppressed viraemia; ritonavir discontinuation may also enhance tolerability and reduce long-term adverse events (AEs). This open-label, multicentre, noninferiority study enrolled HIV-1-infected, treatment-experienced adults with confirmed HIV-1 RNA ≤ 75 HIV-1 RNA copies/mL currently receiving tenofovir/emtricitabine + atazanavir/ritonavir (TDF/FTC + ATV/r) for ≥ 6 months with no reported history of virological failure.

Methods

Participants were randomized 1:2 to continue current treatment or switch to abacavir/lamivudine + atazanavir (ABC/3TC + ATV). Endpoints included the proportion of participants with HIV-1 RNA < 50 copies/mL by time to loss of virological response (TLOVR), AEs, fasting lipids, and inflammatory, coagulation, bone and renal biomarkers.

Results

After 48 weeks, 76% (152 of 199) of ABC/3TC + ATV-treated and 79% (77 of 97) of TDF/FTC + ATV/r-treated participants had HIV-1 RNA < 50 copies/mL (TLOVR; $P = 0.564$). Other efficacy analyses yielded similar results. Rates of new grade 2–4 AEs were 45% in both groups, but an excess of hyperbilirubinaemia made the rate of treatment-emergent grade 3–4 laboratory abnormalities higher with TDF/FTC + ATV/r (36%) compared with ABC/3TC + ATV (19%). Most fasting lipid levels remained stable over time; high-density lipoprotein (HDL) cholesterol increased modestly in ABC/3TC + ATV-treated participants. Bone and renal biomarkers improved significantly between baseline and week 48 in participants taking ABC/3TC + ATV and were stable in participants taking TDF/FTC + ATV/r. No significant changes occurred in any inflammatory or coagulation biomarker within or between treatment groups.

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Conclusions

The ABC/3TC + ATV treatment-switch group had similar viral suppression rates up to 48 weeks to the TDF/FTC + ATV/r comparator group, with lower rates of moderate- to high-grade hyperbilirubinaemia and improvements in bone and renal biomarkers.

Keywords: abacavir, bone biomarkers, HIV, renal biomarker, tenofovir

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Introduction

HIV treatment guidelines from the US Department of Health and Human Services (DHHS) have endorsed regimen simplification or switching in treatment-experienced patients with suppressed viraemia [1]. One simplification strategy is the discontinuation of low-dose ritonavir once viral suppression has been achieved, which has been shown in some studies to improve tolerability and reduce toxicities [2–9]. Depending on the other regimen components, ritonavir discontinuation may also eliminate concern about cytochrome P4503A (CYP3A)-mediated drug interactions with other drugs [2–10]. While atazanavir is a generally well-tolerated protease inhibitor (PI) whose plasma concentration and genetic barrier to resistance are improved by coadministration with ritonavir, the combination is also associated with an increased risk of indirect hyperbilirubinaemia [11–13], which, when accompanied by scleral icterus, can prompt treatment discontinuation [14].

This study, ASSURE (A Simplification Study of Unboosted R_{eyataz} with E_{pzicom}), investigated the efficacy and safety of discontinuing ritonavir in virologically suppressed participants receiving a regimen of tenofovir/emtricitabine + atazanavir/ritonavir (TDF/FTC + ATV/r), one of the recommended first-line regimens in the DHHS guidelines [1]. As TDF/FTC is not recommended for use with unboosted ATV because of a drug–drug interaction that results in decreased concentrations of ATV [11], participants in this trial who were randomized to discontinue ritonavir simultaneously switched to a nucleoside reverse transcriptase inhibitor (NRTI) backbone of abacavir/lamivudine (ABC/3TC). This paper presents the 48-week results of this study, including virological efficacy, immunological response, safety, tolerability, and HIV-1 genotypic and phenotypic resistance patterns in participants who experienced confirmed virological failure. Changes from baseline in fasting lipids and in biomarkers associated with cardiovascular, bone and renal health were also evaluated.

Methods

The prospective, randomized, multicentre, open-label, phase IV ASSURE study (EPZ113734; NCT01102972) enrolled HIV-

1-infected, antiretroviral (ART)-experienced adults (≥18 years of age) who were receiving a once-daily regimen of TDF/FTC (300 mg/200 mg) + ATV/r (300 mg/100 mg) for at least 6 months prior to the first day of screening. Eligible participants were virologically suppressed (defined as HIV-1 RNA ≤ 75 HIV-1 RNA copies/mL) at two consecutive time-points (including the screening visit and one additional visit at least 28 days prior to screening). TDF/FTC + ATV/r could be a participant's initial regimen or first or second switch regimen (defined as any change in ART components) as long as there was no documented history of virological failure. All previous ART regimens consisted of two NRTIs (either TDF/FTC or zidovudine/lamivudine) plus either a Food and Drug Administration (FDA)-licensed nonnucleoside reverse transcriptase inhibitor (NNRTI) or a ritonavir-boosted PI, although alternative prior regimens were allowed on a case-by-case basis.

Participants were excluded if they were human leucocyte antigen (*HLA*)-*B*5701*-positive, or had prior abacavir exposure, active Centers for Disease Control and Prevention (CDC) clinical category C disease, ongoing clinically relevant hepatitis and/or chronic hepatitis B virus (HBV) infection [HBV surface antigen positive (HBsAg+)] or a creatinine clearance < 50 mL/min via the Cockcroft – Gault method. While a baseline genotype was not required, participants were deemed ineligible if they had known HIV genotyping results that indicated the presence of any NRTI or PI mutation associated with resistance to any study drug. There were no CD4 cell count restrictions. Women of child-bearing potential were eligible if they had negative pregnancy tests at screening and baseline and agreed to use protocol-prescribed contraception methods throughout the study. Additional information on inclusion/exclusion criteria, including a link to the study protocol, is available in a previous publication [15]. All participants provided written informed consent to participate in the study, and the protocol was approved by the institutional review board for each study site.

Design and intervention

After stratification by prior ART experience (TDF/FTC + ATV/r as initial regimen or as first/second switch regimen), eligible participants were randomized 2:1 to sim-

plify their regimen to once-daily ABC/3TC 600 mg/300 mg (ViiV Healthcare, Research Triangle Park, NC) plus 400 mg ATV once daily (two 200 mg tablets; Bristol-Myers Squibb, Princeton, NJ) or to remain on once-daily TDF/FTC (Gilead Sciences, Foster City, CA) plus one 300 mg ATV tablet boosted with a 100 mg ritonavir tablet (AbbVie, Chicago, IL). Randomization and study drug provisioning were performed by GlaxoSmithKline's randomization and medication ordering system.

Procedures and assessments

Participants were evaluated at screening, baseline, and weeks 2, 4, 12, 24, 36 and 48; visits included routine chemistry, haematology, HIV-1 RNA and immunology assessments. The concentrations of specific biomarkers [bone alkaline phosphatase (BAP), parathyroid hormone (PTH), C-terminal telopeptide (C-telopeptide), osteocalcin, urine β 2 microglobulin (β 2m):creatinine ratio, high-sensitivity C-reactive protein (hs-CRP), interleukin-6 and D-dimer] were evaluated at baseline, week 24 and week 48 (as were vitamin D levels).

HIV-1 RNA concentrations were measured using the RealTime HIV-1 Assay (Abbott Molecular, Inc., Des Plaines, IL). Adverse events and laboratory toxicities were graded using the 2004 Division of AIDS Toxicity Grading Scale. All suspected ABC hypersensitivity events were reported as serious adverse events (SAEs). Glomerular filtration rates (GFRs) were estimated using the modification of diet in renal disease (MDRD) formula [16]. Smoking status and presence of diabetes were assessed at baseline, week 24 and week 48 for Framingham risk score calculation [17]; treatment-emergent changes in fasting total, high-density lipoprotein (HDL) and low-density lipoprotein (LDL) cholesterol and triglycerides were assessed using the Division of AIDS lipid toxicity grading system [18]. Viral genotypes and phenotypes were determined by Monogram Biosciences (South San Francisco, CA); mutations were defined according to International AIDS Society–USA Guidelines [19]. All other laboratory tests were performed centrally by Quest Diagnostics (Van Nuys, CA).

Study populations

The intent-to-treat (ITT) population consisted of all enrolled participants randomized in the study. The primary population for efficacy analyses included all participants in the ITT population who received at least one dose of study drug [ITT-exposed (ITT-E)]. The safety population consisted of all randomized participants who consumed any investigational product. The virological failure population included all participants who met the protocol-defined

criteria for confirmed virological failure, defined as plasma HIV-1 RNA \geq 400 copies/mL on two consecutive occasions. The observed data set contained all data collected while participants were in the study; no missing values were imputed.

Outcome measures

The primary efficacy endpoint was the proportion of participants with HIV-1 RNA $<$ 50 copies/mL at week 24 by the time to loss of virological response (TLOVR) algorithm [15]. The proportion of participants with HIV-1 RNA $<$ 50 copies/mL at week 48 was a secondary endpoint. Other secondary endpoints included the proportion of participants with HIV-1 RNA $<$ 400 copies/mL, the change from baseline in CD4 cell count, time to virological failure, detection of genotypic and phenotypic resistance at the time of virological failure, change from baseline in fasting lipid profiles (total, LDL and HDL cholesterol and triglycerides), grade 2–4 AEs and all SAEs. There were two exploratory endpoints: (1) change from baseline in neurocognition scores, to be reported in a separate paper; and (2) change from baseline in eight cardiovascular/inflammation, bone and renal biomarkers, included in this publication.

Statistical analyses

Per protocol, virological response rates at week 48 were compared using a Cochran – Mantel–Haenszel test stratified by initial ART regimen (TDF/FTC + ATV/r as initial regimen or as first/second switch regimen). All statistical comparisons were based on a two-sided significance level of 0.05. Continuous variables were assessed using the Wilcoxon rank-sum test, and binary responses were compared using Fisher's exact test. Biomarker data were log-transformed prior to analysis, and the change from baseline was assessed using geometric mean ratios with 95% confidence intervals. The statistical analyses were performed using SAS Version 9.1 (SAS Institute Inc., Cary, NC).

Results

Characteristics and accountability

This study enrolled 297 participants from 43 clinical sites in the USA and Puerto Rico. One participant randomized to TDF/FTC + ATV/r was withdrawn because of a protocol violation prior to receiving any study medication, so the ITT-E population included 199 participants simplifying to ABC/3TC + ATV and 97 continuing on TDF/FTC + ATV/r. Participants were recruited between April 2010 and December 2011, and the last 48-week analysis visit occurred in

Table 1 Baseline participant demographics and characteristics

	ABC/3TC + ATV (n = 199)	TDF/FTC + ATV/r (n = 97)	Total
Age (years) [median (range)]	44 (21–66)	42 (20–68)	43.5 (20–68)
Male [n (%)]	155 (78)	79 (81)	234 (79)
Race [n (%)]			
African-American/African heritage	65 (33)	37 (38)	102 (34)
White/Caucasian/European heritage	122 (61)	55 (57)	177 (60)
Other	12 (6)	5 (5)	17 (6)
Hispanic or Latino ethnicity [n (%)]	51 (26)	26 (27)	77 (26)
Plasma HIV-1 RNA (log ₁₀ copies/mL) (median)	1.59	1.59	1.59
<50 copies/mL [n (%)]	192 (96)	93 (96)	285 (96)
50 to 74 copies/mL [n (%)]	2 (1)	2 (2)	4 (1)
≥ 75 copies/mL [n (%)]	5 (3)	2 (2)	7 (2)
CD4 cell count (cells/μL) [median (range)]	492 (77–1196)	480 (108–1479)	292 (77–1479)
<200 cells/μL [n (%)]	14 (7)	6 (6)	20 (7)
CDC HIV infection classification [n (%)]			
Category A	136 (68)	67 (69)	203 (69)
Category B	26 (13)	13 (13)	39 (13)
Category C	37 (19)	17 (18)	54 (18)
Hepatitis C virus coinfection [n (%)]	18 (9)	8 (8)	26 (9)
Time on prior ART (days) [median (range)]	978 (177–4830)	1106 (199–7078)	998 (177–7078)

ABC/3TC, abacavir/lamivudine; ART, antiretroviral therapy; ATV, atazanavir; ATV/r, atazanavir/ritonavir; CDC, Centers for Disease Control and Prevention; TDF/FTC, tenofovir/emtricitabine.

December 2012. Most participants (79%) were male (Table 1), 34% were of African-American/African heritage, and 26% self-identified as Hispanic/Latino. The median baseline CD4 cell count was 492 cells/μL. Median time on ART prior to enrolment was 978 days for the ABC/3TC + ATV group and 1106 days for the TDF/FTC + ATV/r group.

Eighty-five per cent (253) of participants in the ITT-E population completed 48 weeks on study (Fig. 1). The most common reasons for study withdrawal were lost to follow-up (5%; 15 participants), consent withdrawn (4%; 12 participants), and AEs (3%; 10 participants).

Efficacy results

As previously reported [15], 86.9% (173 of 199) of participants in the ABC/3TC + ATV treatment group successfully maintained HIV-1 RNA < 50 copies/mL by the TLOVR analysis at week 24 compared with 86.6% (84 of 97) in the TDF/FTC + ATV/r group. The adjusted treatment difference was 0.33%, and the two-sided 95% confidence interval (CI) (–7.97 to 8.64%) stratified by prior ART regimen excluded the predefined noninferiority margin of –12%, demonstrating the noninferiority of the ABC/3TC + ATV simplification regimen to the TDF/FTC + ATV/r continuation regimen.

At week 48, the efficacy results were also similar between treatment groups ($P = 0.564$), with 76% (152 of 199) of participants taking ABC/3TC + ATV and 79% (77 of 97) taking TDF/FTC + ATV/r having HIV-1 RNA < 50 copies/mL by TLOVR analysis. In the observed analysis, there was no

significant difference ($P = 0.134$) in HIV-1 RNA < 50 copies/mL at week 48 between treatment groups, with response rates of 91% (154 of 169) in the ABC/3TC + ATV-treated group versus 96% (79 of 82) in the TDF/FTC + ATV/r-treated group. There was no significant difference between treatment groups by TLOVR analysis in the HIV-1 RNA < 50 copies/mL response rate when adjusted by gender ($P = 0.672$), race ($P = 0.645$) or age ($P = 0.652$).

At week 48, median CD4 cell counts were 603 and 590 cells/μL for ABC/3TC + ATV and TDF/FTC + ATV/r, respectively. However, the median increase in CD4 cell count from baseline to week 48 was significantly larger ($P = 0.026$) in the ABC/3TC + ATV group (+90 cells/μL) compared with the TDF/FTC + ATV/r group (+47 cells/μL).

Virology results

Confirmed virological failure occurred in four of 199 (2%) participants switched to ABC/3TC + ATV and one of 97 (1%) participants continuing on TDF/FTC + ATV/r. At virological failure, HIV-1 from three participants (two receiving ABC/3TC + ATV; one receiving TDF/FTC + ATV/r) was fully susceptible to all drugs, with no reverse transcriptase (RT) mutations and a few minor PI mutations (including L10I, G16G/E, D60E, I62V, V77I and I93L) detected, and confirmed virological failure was associated with site reports of noncompliance and/or therapy interruption. HIV-1 from the remaining two participants had reduced study drug susceptibility at failure. One ABC/3TC + ATV-receiving participant experienced initial rebound at day 14, con-

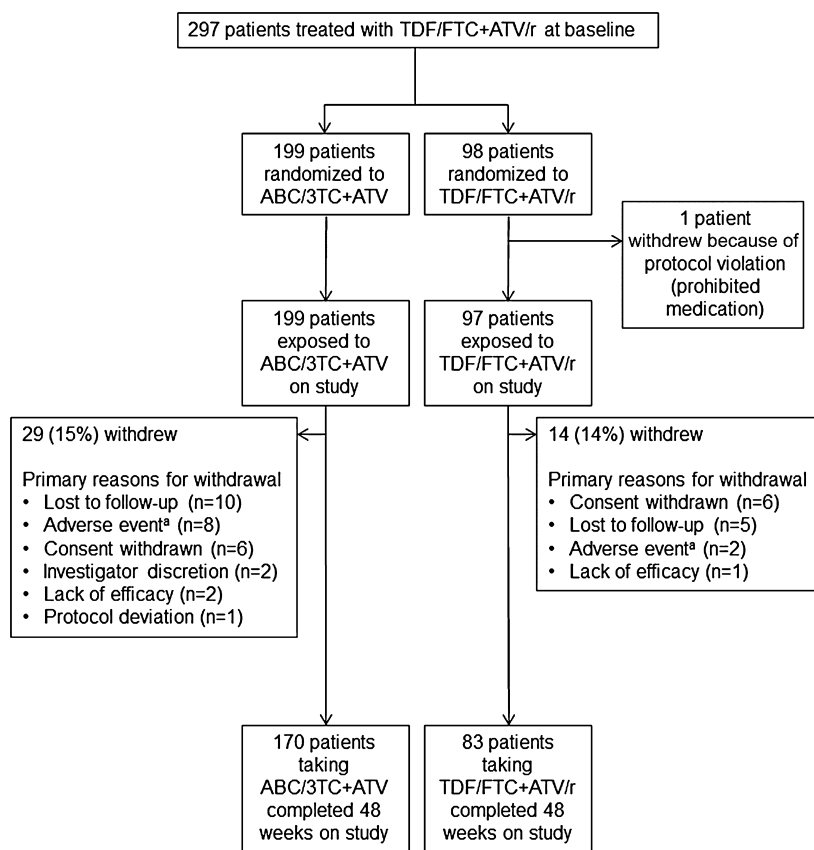


Fig. 1 Participant disposition. Primary reasons for withdrawal were listed by the investigator on the case report form. ^aAll adverse events leading to study discontinuation were grade 1 or 2, with the exception of a grade 3 lipase increase in one participant continuing on tenofovir/emtricitabine + atazanavir/ritonavir (TDF/FTC + ATV/r). ABC/3TC, abacavir/lamivudine; ATV, atazanavir; ATV/r, atazanavir/ritonavir; TDF/FTC, tenofovir/emtricitabine.

comitant with site-reported treatment compliance issues. No prior HIV-1 genotypes or comprehensive treatment records were available, but the participant reported receiving nevirapine plus TDF/FTC for >8 years before TDF/FTC + ATV/r. Numerous HIV-1 RT mutations (M41L, L74V, K103N, M184V, L210W and T215Y) were detected, including thymidine analogue mutations (TAMs), despite no prior reported zidovudine- or stavudine-containing treatment. Multiple PI mutations (L10F, K20I, M46I, I54V, I62V, L63P, A71T, G73T, I84V and L90M) were detected. The participant's HIV-1 had reduced phenotypic susceptibility to multiple drugs, including ABC, 3TC and ATV. The final ABC/3TC + ATV-receiving participant experienced viral rebound at week 36. No prior genotypes were available. Several years of prior treatment with TDF/FTC and efavirenz before switching to a TDF/FTC + ATV/r regimen were reported. The M41L and T215Y TAMs and the M184V mutation were detected at failure, despite no prior reported zidovudine or stavudine treatment, as were numerous PI

mutations (L10I, K20R, L24I, M36I, K43T, M46L, I62V, V82A and L89M); the virus from this participant remained susceptible to both ABC and TDF but had reduced susceptibility to ATV and 3TC/FTC.

Safety and tolerability results

Rates of AEs of moderate or greater severity (grade 2–4) were similar between the two groups (45% for both groups; Table 2). There were few grade 2–4 treatment-related AEs in either group (9% for ABC/3TC + ATV and 6% for TDF/FTC + ATV/r). Five participants experienced grade 2–4 cardiac disorders during the course of the study; none were considered to be drug related. In the TDF/FTC + ATV/r group, one participant had grade 2 palpitations. In the ABC/3TC + ATV group, two participants were diagnosed with grade 2 cardiomyopathy; the third had a prior history of congestive heart failure and developed grade 2 coronary artery disease, and the fourth participant, a former smoker

Table 2 Adverse events and laboratory abnormalities

	ABC/3TC + ATV (n = 199) n (%)	TDF/FTC + ATV/r (n = 97) n (%)
Grade 2–4 AEs		
Any	90 (45)	44 (45)
Occurring in ≥ 5% of participants in either treatment group		
Upper respiratory tract infection	11 (6)	7 (7)
Treatment related [†]	17 (9)	6 (6)
Nausea	4 (2)	0
Abnormal dreams	2 (1)	0
Dizziness	2 (1)	0
Severe or grade 3–4 AEs		
Any [†]	29 (15)	10 (10)
Treatment related	0	2 (2)
Blood bilirubin increased	0	1 (1)
Lipase increased	0	1 (1)
Serious AEs		
Any [†]	21 (11)	6 (6)
Treatment related	1 (< 1)	0
Drug hypersensitivity	1 (< 1)	0
AEs leading to study withdrawal		
Any	8 (4)	2 (2)
AEs occurring in > 1 participant and leading to study withdrawal		
Nausea	3 (2)	0
Vomiting	2 (1)	0
Rash	2 (1)	0
Treatment-emergent laboratory abnormalities		
Any grade 2 – 4 event [‡]	99 (50)	66 (69)
Any grade 3 – 4 event [‡]	37 (19)	35 (36)
Total bilirubin	12 (6)	28 (29)
Creatine kinase	7 (4)	4 (4)
Lipase	5 (3)	3 (3)
Inorganic phosphorus	5 (3)	1 (1)
Glucose	5 (3)	1 (1)
LDL cholesterol calculation	3 (2)	2 (2)
Aspartate aminotransferase	1 (< 1)	1 (1)
Alanine aminotransferase	1 (< 1)	1 (1)
Cholesterol	1 (< 1)	0

[†]Listing includes all events occurring in more than one participant.

[‡]No single event occurred in more than three participants.

[§] $P = 0.0026$ using Fisher's exact test.

[¶] $P = 0.0014$ using Fisher's exact test.

ABC/3TC, abacavir/lamivudine; AE, adverse event; ATV, atazanavir; ATV/r, atazanavir/ritonavir; LDL, low-density lipoprotein; TDF/FTC, tenofovir/emtricitabine.

with diabetes, hyperlipidaemia, and a family history of cardiovascular disease, had a grade 4 acute inferior myocardial infarction.

Adverse events leading to study withdrawal affected 4% of participants receiving ABC/3TC + ATV and 2% of participants continuing on TDF/FTC + ATV/r; of these, only nausea, vomiting and rash were reported in more than one participant. Suspected ABC hypersensitivity was reported for one participant during the course of the study; the participant was not withdrawn but restarted his original TDF/FTC + ATV/r regimen with resolution of signs and symptoms.

The rate of treatment-emergent or worsening grade 2–4 ($P = 0.0026$) and grade 3–4 ($P = 0.0014$) laboratory abnormalities was significantly higher in the TDF/FTC + ATV/r group compared with the ABC/3TC + ATV group. The dif-

ference was primarily driven by change from baseline in total bilirubin levels. Other treatment-emergent grade 3–4 laboratory abnormalities were infrequent and similar between groups (Table 2). The fasting lipid levels for participants with paired baseline and week 48 results (Fig. 2) were similar between treatment groups and varied little between baseline and week 48, except for a small but statistically significant increase in median HDL cholesterol levels (3.0 mg/dL; $P = 0.013$) for the ABC/3TC + ATV group.

At baseline, the median estimated glomerular filtration rate by the MDRD equation was 93 mL/min/1.73 m² for both treatment groups. At week 48, there was a small median increase of 0.8 mL/min/1.73 m² for the ABC/3TC + ATV group and a small median decrease of -1.3 mL/min/1.73 m² in the TDF/FTC + ATV/r group, but this effect was not statistically significant.

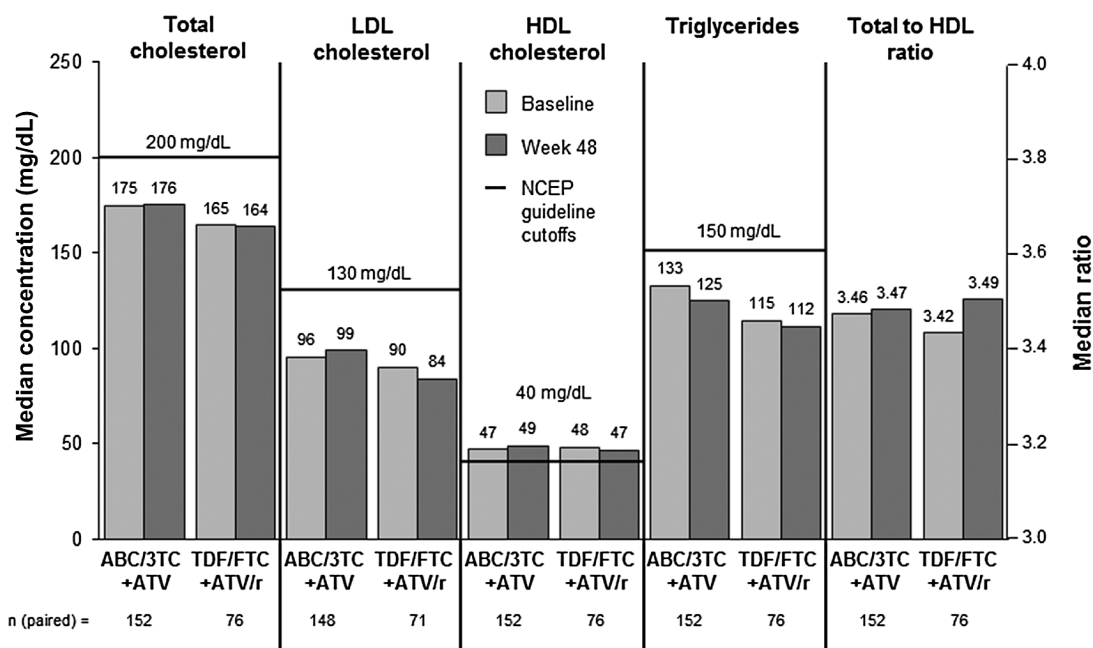


Fig. 2 Median fasting lipid parameters with guideline cut-offs from the National Cholesterol Education Program (NCEP) [17] for participants with paired baseline and week 48 values. A significant increase ($P < 0.0001$) in median high-density lipoprotein (HDL) cholesterol levels for the change from baseline (CFB) to week 48 was seen for the abacavir/lamivudine + atazanavir (ABC/3TC + ATV) group with no significant increase in the tenofovir/emtricitabine + atazanavir/ritonavir (TDF/FTC + ATV/r) treatment group ($P = 0.8994$); a significant difference ($P = 0.013$) between treatment groups was also observed. ABC/3TC, abacavir/lamivudine; ATV, atazanavir; ATV/r, atazanavir/ritonavir; LDL, low-density lipoprotein; TDF/FTC, tenofovir/emtricitabine.

The median 10-year risk of coronary heart disease as measured by the Framingham equation at baseline and week 48 was 2.0% and 2.0%, respectively, for the ABC/3TC + ATV group and 1.5% and 2.0% for the TDF/FTC + ATV/r group, respectively.

Biomarkers

Between baseline and week 48, there was a significant ($P < 0.001$) decrease resulting in improvement for the measured bone biomarkers BAP, PTH, C-telopeptide and osteocalcin in the ABC/3TC + ATV group (Fig. 3a–d); these biomarkers remained relatively unchanged in the TDF/FTC + ATV/r group over the same time period. Comparing the two treatment groups, the decrease from baseline to week 48 in all four of these bone biomarkers was significantly larger ($P < 0.001$) for the ABC/3TC + ATV group than for the TDF/FTC + ATV/r group. Serum calcium levels remained similar within and between groups, with baseline mean values of 9.33 and 9.35 mg/dL for the ABC/3TC + ATV and TDF/FTC + ATV/r groups, respectively, and mean changes of 0.05 and -0.02 mg/dL, respectively, at week 48. Vitamin D (25 OH) geometric mean values were in the low but normal range at baseline (28.3 ng/mL in the

ABC/3TC + ATV group and 26.4 ng/mL in the TDF/FTC + ATV/r group). At week 48, these values were 24.6 ng/mL in the ABC/3TC + ATV group and 29.3 ng/mL in the TDF/FTC + ATV/r group, representing a decline from baseline for the ABC/3TC + ATV group ($P < 0.001$) and an increase for the TDF/FTC + ATV/r group ($P = 0.005$).

The renal biomarker $\beta 2M$:creatinine ratio declined by 56% in the ABC/3TC + ATV group ($P < 0.001$) but increased by 14% in the TDF/FTC + ATV/r group (Fig. 3e); the change from baseline to week 48 was significantly different ($P < 0.001$) between treatment groups.

Conversely, there were no significant changes from baseline to week 48 in any of the biomarkers associated with cardiovascular disease, inflammation or thrombogenesis that were evaluated (hs-CRP, interleukin-6 and D-dimer) either within or between treatment groups (Fig. 3f–h).

Discussion

In this randomized, multicentre ASSURE study, the proportion of participants maintaining suppression of HIV-1 RNA < 50 copies/mL by TLOVR at week 48 was similar between participants switching to ABC/3TC + ATV (76%) and those maintaining on TDF/FTC + ATV/r (79%). The

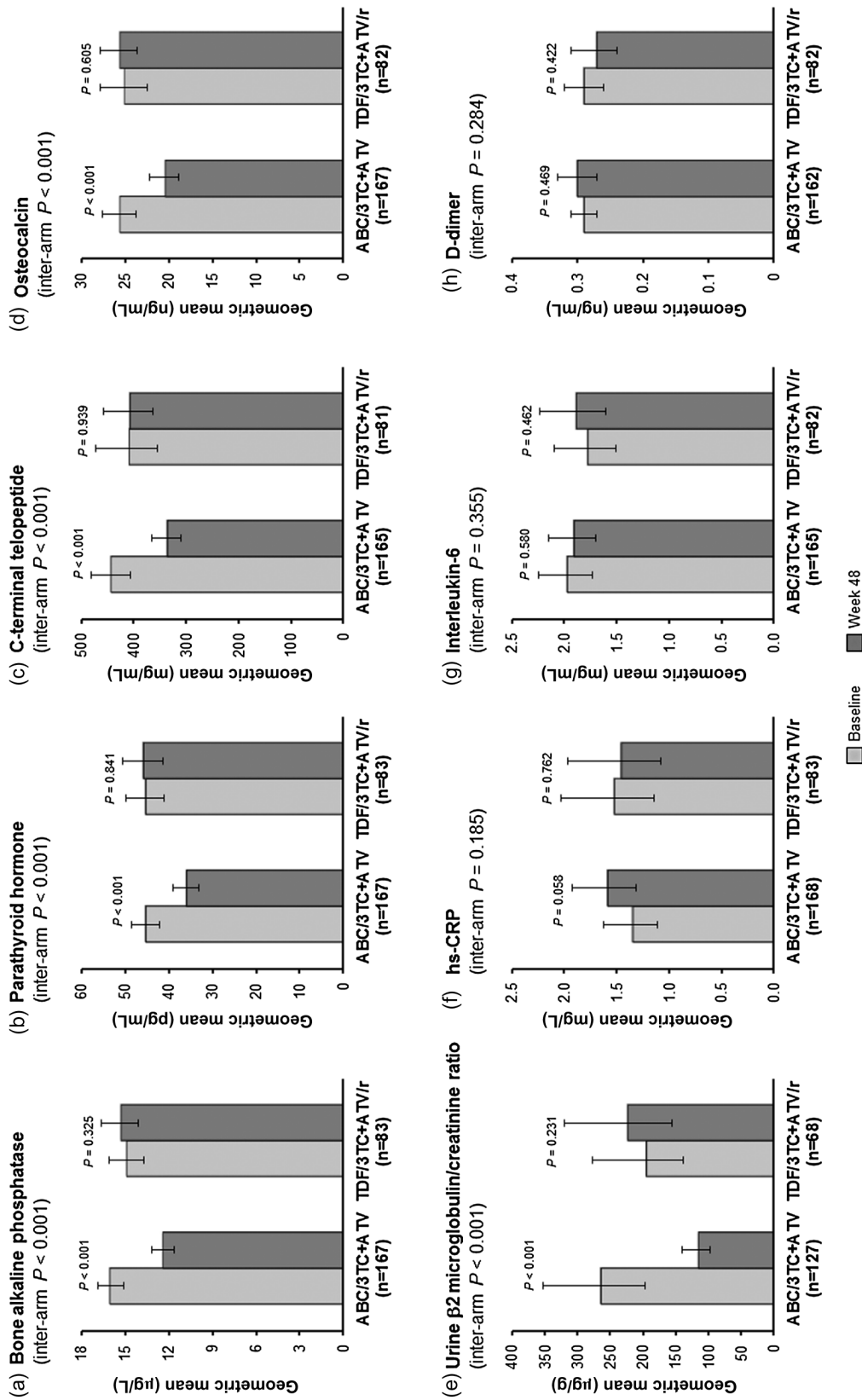


Fig. 3 Biomarkers. Geometric means and 95% confidence intervals for the bone biomarkers (a) bone alkaline phosphatase, (b) parathyroid hormone, (c) C-terminal telopeptide and (d) osteocalcin; (e) the renal biomarker urine β_2 microglobulin:creatinine ratio; and the inflammatory biomarkers (f) high-sensitivity C-reactive protein (hs-CRP), (g) interleukin-6 and (h) D-dimer in participants from both treatment groups with data at baseline and week 48. Intra-arm P -values were calculated using a paired t -test on the ratio of the geometric mean concentration at baseline and the geometric mean concentration at week 48. Inter-arm P -values were calculated using a two-sample t -test on the ratio of the geometric mean change from baseline to week 48 in the abacavir/lamivudine + atazanavir (ABC/3TC + ATV) versus tenofovir/emtricitabine + atazanavir/ritonavir (TDF/FIC + ATV/r) groups. ABC/3TC, abacavir/lamivudine; ATV, atazanavir; ATV/r, atazanavir/ritonavir; TDF/FIC, tenofovir/emtricitabine.

efficacy and tolerability of ABC/3TC + ATV demonstrated in this study complement those observed in the ARIES trial, where ART-naïve patients initiated treatment with ABC/3TC + ATV/r, and after 36 weeks the virologically suppressed population was randomized to continue on the same regimen or switch to a regimen of ABC/3TC + ATV. Viral suppression was maintained 48 weeks post-randomization in 86% of patients in the ABC/3TC + ATV-treated group and in 81% of those taking ABC/3TC + ATV/r, accompanied by lower rates of treatment-related grade 2–4 AEs and a decrease in hyperbilirubinaemia from 14% to 4% for the ABC/3TC + ATV-treated group [9]. In ASSURE, switching to ABC/3TC + ATV was also accompanied by fewer treatment-emergent or worsening AEs, driven largely by a lower rate of moderate to severe hyperbilirubinaemia and by a modest but significant increase in median CD4 cell count. Similarly, in the INDUMA study, previously antiretroviral-naïve patients received two NRTIs plus ATV/r for 26 to 30 weeks, and virologically suppressed patients were randomized to unboosted ATV or maintained ATV/r. After 48 weeks there was no significant difference in virological suppression between the groups (78% for ATV and 75% for ATV/r), while hyperbilirubinaemia decreased by 50% (from 32 to 16%) in the unboosted ATV group [4].

In the ASSURE study, significant improvements in the bone turnover markers PTH, BAP, C-telopeptide and osteocalcin were observed following the switch to ABC/3TC + ATV, with little change over time in the TDF/FTC + ATV/r group. Initiation of ART is often associated with decreases in bone mineral density that slow or stabilize over 24 to 48 weeks of therapy; these losses tend to be larger with TDF-containing regimens compared with non-TDF-containing regimens [20–23]. Bone biomarkers have been studied in treatment-naïve patients initiating therapy [24–27], but few randomized trials have included treatment-experienced adults [28–30]; results from these treatment-experienced studies were generally consistent with our findings that bone biomarkers were more favourable in patients taking ABC/3TC compared with TDF/FTC and probably reflect clinically relevant differences between these regimens in bone turnover. The improvement in bone markers observed after switching from TDF in our study was not explained by changes in levels of calcium or vitamin D. Vitamin D levels at baseline and at week 48 remained within the lower end of the normal range for both treatment groups, and a small decline was noted for these levels in the ABC/3TC + ATV treatment group.

Switching to ABC/3TC + ATV also resulted in improvement in the β 2M:creatinine ratio, a validated marker of combined glomerular and tubular health. While the association between TDF and renal impairment is well studied

(for example [31–34]), only one randomized trial, ASSERT, which compared ABC/3TC with TDF/FTC both in combination with efavirenz in treatment-naïve patients, has included the β 2M:creatinine ratio [32]. After 48 weeks on therapy, the β 2M:creatinine ratio increased by 24% in the TDF/FTC group and decreased by 47% in the ABC/3TC group ($P < 0.0001$), a result consistent with the results observed in ASSURE (albeit in treatment-naïve patients).

In this study, switching to ABC/3TC + ATV was not associated with significant changes in the markers of inflammation and coagulation, interleukin-6, hs-CRP and D-dimer. Data regarding these markers following administration of ABC have been mixed. Several studies found no increase in markers of cardiovascular disease with initiation of ABC compared with other NRTIs [28,29,34–39], while study A5202 reported a significant increase from baseline in hs-CRP after 96 weeks in treatment-naïve patients randomized to ABC/3TC, with no significant change in patients randomized to TDF/FTC, although interleukin-6 levels decreased significantly in both the ABC/3TC- and TDF/FTC-treated groups [24]. Four other studies have examined cardiovascular markers in treatment-experienced patients: STEAL [28], SWAP [29], BICOMBO [36], and SWIFT [40]. As in our study, none of these trials reported a difference between ABC/3TC and TDF/FTC in hs-CRP, interleukin-6 or D-dimer over 48 weeks of post-randomization treatment.

Virological failure was rare in both study arms. Wild-type virus at virological failure was detected in three of five participants with confirmed viraemia > 400 copies/mL and was associated with therapy interruption/noncompliance. Two additional participants, both receiving ABC/3TC + ATV, experienced virological failure, one shortly after therapy switch (day 14) and one at week 36. Multiple viral resistance mutations, including TAMs, were detected in virus from both participants, and the K103N NNRTI mutation was detected in one participant. As TAMs are not selected by treatment with either TDF/FTC or ABC/3TC, these results are suggestive of these participants having previously experienced treatment failure on either zidovudine- or stavudine-containing regimens, although no prior exposure was reported for either participant.

There are several limitations of this trial that should be considered when interpreting results from this study. This simplification treatment strategy is restricted to certain NRTI backbones such as ABC/3TC that can be used in combination with unboosted ATV. The study enrolled virologically suppressed, therapy-experienced patients and was designed to exclude those patients with prior virological failure that could have resulted in selection for drug resistance-associated viral mutations affecting resistance to the investigational products. Therefore, extrapolating

these findings to other patient populations may be inappropriate. Plasma concentrations of ATV in the absence of ritonavir boosting are lower, and therefore this regimen may be more susceptible to patient adherence issues, especially if archived resistance-associated mutations are present. While the markers of bone density examined are considered accepted markers of bone turnover, this study did not directly measure bone density.

Conclusions

Over 48 weeks, discontinuation of ritonavir after switching from a regimen of TDF/FTC + ATV/r to ABC/3TC + ATV maintained viral suppression in study participants and led to improvements in CD4 cell count, the bone biomarkers PTH, BAP, C-telopeptide and osteocalcin, and the renal β 2M:creatinine ratio. The ABC/3TC + ATV treatment group also had significantly fewer laboratory abnormalities than the TDF/FTC + ATV/r treatment group without increasing other fasting lipid levels or cardiovascular biomarkers of inflammation and thrombogenesis.

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Contributions to authorship

The sponsor developed the study design with input from prospective investigators. Substantial contributions to study conception and design were made by DAW, DAM, HHZ, LLR and MSS. Substantial contributions to the analysis and interpretation of the data were made by HHZ, DAW, LLR, DAM and MSS. Substantial contributions to acquisition of patient clinical study data were made by DAW, LB, CBS, HE, ED and GWG. All authors (DAW, DAM, HHZ, LLR, MSS, LB, CBS, HE, ED and GWG) had full access to the data and vouch for the accuracy and completeness of the data and analyses. The manuscript was written and approved by all of the authors, each of whom contributed to the drafts and revisions.

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