Title

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Journal

JAIDS Journal of Acquired Immune Deficiency Syndromes, 74(5)

ISSN

1525-4135

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Publication Date

2017-04-15

DOI

10.1097/qai.0000000000001200

Peer reviewed

HHS Public Access

Author manuscript

J Acquir Immune Defic Syndr. Author manuscript; available in PMC 2018 April 15.

Published in final edited form as:

J Acquir Immune Defic Syndr. 2017 April 15; 74(5): e138–e145. doi:10.1097/QAI.0000000000001200.

Changes in Urinary Biomarkers over 10 Years is Associated with Viral Suppression in a Prospective Cohort of Women Living with HIV

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AUTHOR CONTRIBUTIONS: Research idea and study design: SMB, RS, VJ, MGS; data acquisition: SMB, RS, MGS; data analysis/interpretation: SMB, RS, MME, AGA MGS; statistical analysis: RS; supervision or mentorship: MGS. Each author contributed important intellectual content during manuscript drafting or revision and accepts accountability for the overall work by ensuring that questions pertaining to the accuracy or integrity of any portion of the work are appropriately investigated and resolved. MGS takes responsibility that this study has been reported honestly, accurately, and transparently; that no important aspects of the study have been omitted; and that any discrepancies from the study as planned have been explained.

Disclosures and conflicts of interest: R.S. received an honorarium from Merck for participating in a Renal Expert Input Forum in June 2014; this honorarium was donated to NCIRE to support kidney research. S.M.B, V.J., M.M.E., A.G.A, C.R.P., M.R.B., M.H.C., M.J.N., D.R.G., A.S., M.A.Y. and M.G.S. all individually report no relevant disclosures or conflicts of interest.

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Abstract

Background—Urine biomarkers have helped identify persons at risk for progressing to kidney disease in the setting of HIV infection. We explored factors associated with changes in three urine biomarkers over 10 years among women living with HIV.

Methods—Prospective cohort of 294 HIV-infected women from the multicenter Women's Interagency HIV Study (WIHS). Predictors included HIV viral and immunological parameters, co-morbid conditions and health-related behaviors. Outcomes were patterns of changes of urine interleukin-18 (IL-18), albumin-to-creatinine ratio (ACR) and alpha-1-microglobulin (α1m) over 10 years. We used quantile regression to examine patterns of change in each urine biomarker during follow-up and multivariable analysis of variance (MANOVA) regression to identify predictors of biomarker changes.

Results—Over 10 years, the median concentrations of IL-18 declined from 120 to 64 pg/mL, α1m rose from 0.7 to 1.5 ng/mL, and ACR remained stable (9 to 8 mg/g). In multivariate analyses, the strongest predictors of increases in IL-18 were higher baseline BMI, increase in waist circumference, higher follow-up HIV viral load, lower follow-up CD4 cell count, HCV co-infection and higher follow-up HDL cholesterol. Predictors of increasing concentration of α1m were lower CD4 cell counts, higher diastolic blood pressure, HCV co-infection and smoking. Finally, determinants of ACR increases during follow-up were higher follow-up diastolic blood pressure, HCV co-infection, higher follow-up HIV viral load and triglyceride concentration.

Conclusions—Over 10 years, HIV disease status had different associations with each urine biomarker under study. Overall, the associations with changes in each biomarker support research into their use for longitudinal monitoring of kidney health.

Short Summary

In a 10 year longitudinal study, the authors evaluated how changes in HIV clinical and immunological factors were related to changes in distinct urine biomarkers. The patterns of changes in urine biomarkers as well as the risk factors that were associated with these changes were distinct. Further research is warranted in large, longitudinal cohorts, to understand how these changes may predict subsequent changes in kidney function.

Index words

biomarkers; chronic renal insufficiency; HIV; WIHS; women

INTRODUCTION

Human immunodeficiency virus (HIV) infection is associated with increased risk of developing chronic kidney disease (CKD)[1], and among persons with HIV infection, the development of CKD is associated with increased morbidity and mortality[1–3]. Urine biomarkers have previously been shown to capture subclinical kidney injury that is not detectable by standard clinical measures such as serum creatinine-based estimated

glomerular filtration rate (eGFR) or urine protein concentrations[4–10]. In addition, several urine biomarkers have shown the ability to distinguish the contributions of specific risk factors for subclinical kidney disease. This attribute is particularly important in HIV-infected persons because CKD can arise as a consequence of multiple different etiologies including chronic antiretroviral therapy, HIV infection itself, hepatitis C virus (HCV) co-infection and co-morbid chronic illnesses such as diabetes mellitus and hypertension[1,2,11–17]. Furthermore, the presence of two abnormal alleles for the *APOL1* gene predisposes a subset of African-Americans to more extensive glomerular injury and faster CKD progression[18–21].

Several distinct urine biomarkers have been evaluated that are predictive of progressive kidney decline and incident CKD. Our prior work in the Women's Interagency HIV Study (WIHS) identified biomarkers that reflect different pathophysiologic aspects of kidney injury, which could aid in early detection[4,6,22–25]. The strongest associations with declines in kidney function have been observed with the urine albumin to creatinine ratio (ACR), alpha-1-microglobulin (α1m) and interleukin-18 (IL-18) concentrations[4,22,23,25]. Elevations of the ACR have been associated with declining kidney function in a variety of diseases, including HIV, and is indicative of glomerular injury of the kidney[4,25]. The α1m is a low molecular weight protein that reflects proximal tubular dysfunction when elevated in urine samples[4,22]. IL-18 is a pro-inflammatory cytokine involved in cell-mediated immunity, and its urine levels correspond to the extent of proximal tubular injury[4,25]. In multivariable analyses with comprehensive covariate adjustment, each of these three markers has been independently associated with faster decline in kidney function and higher mortality risk[4,22–24].

A major step in understanding the clinical utility of a kidney biomarker involves establishing whether or not serial measures capture longitudinal changes in kidney health. Biomarkers are of lesser value if they cannot capture dynamic changes in risk over time. Therefore, biomarker research in HIV-infected persons needs to evaluate the long-term changes in novel urine biomarkers and to understand whether they are responsive to changes in important health status measures, including immune function. In this study, to improve on prior studies that have utilized biomarker measures from a single point in time, we present data investigating 10-year changes of three urine biomarkers among women living with HIV. Our primary hypothesis was that improvements in systemic immune status over 10 years would lead to favorable changes in the urine biomarker profile of kidney injury.

SUBJECTS AND METHODS

Study design and population

The WIHS is a large, multicenter, prospective cohort study of HIV-infected women and at-risk HIV-uninfected women in the United States[26], ongoing since 1993. The WIHS has been described elsewhere[26,27]. At semi-annual visits, participants are interviewed and examined, and serum specimens are collected and stored in a −80°C freezer. During the time of this study, women were enrolled in six U.S. sites (Bronx, Brooklyn, Chicago, Los Angeles, San Francisco, and Washington, DC). The enrolled women are representative of U.S. women living with HIV in terms of demographic and clinical parameters[26]. The specific sub-study of this analysis, known as the WIHS HIV Kidney Aging study, is a nested cohort study

designed to evaluate incident kidney disease in the setting of HIV infection[4,22,28,29]. Baseline measures of urine biomarkers were conducted on stored urine samples that were collected between October 1999 and March 2000 (year 0). Follow-up measures were made in a subsequent collection in 2010 (year 10). Among 908 women with baseline measures of urine ACR, IL-18 and α1m, we selected a random sample among those women with available stored urine at year 10 to include in this study (N=294). WIHS was approved by institutional review boards at all study sites. This study of kidney injury was also approved by the University of California, San Francisco, San Francisco Veterans Affairs Medical Center, Johns Hopkins University and Yale University committees on human research. All WIHS participants provided written and informed consent. The procedures were consistent with the Helsinki Declaration.

Predictors

Candidate covariates were selected from the baseline and year 10 visits, with the exception of CD4 cell count and HIV viral load, which were modeled in several ways including baseline, peak, nadir and time-averaged values. Candidate variables that were considered as potential predictors of biomarker changes included: time-averaged HIV viral load (in copies/mL), peak HIV viral load (in copies/mL), most recent HIV viral load (copies/mL), time-averaged CD4 cell count (cells/μL), nadir CD4 cell count during study period (cells/μL), nadir CD4 cell count prior to year 0 (cells/μL), most recent CD4 cell count (cells/μL), diastolic blood pressure (in mm Hg), systolic blood pressure (in mm Hg), history of diabetes mellitus (yes or no, by laboratory confirmation or self-report), history of hypertension (yes or no, by self-report or confirmed on examination or use of anti-hypertensive medication), HCV co-infection (yes or no, by presence of HCV antibody or RNA), current smoking status (yes or no), past smoking history (yes or no), body mass index (in kg/m^2), waist circumference (in centimeters), age (in years), serum LDL cholesterol (in mg/dL), serum triglyceride (in mg/dL), serum HDL cholesterol (in mg/dL), history of AIDS (yes or no) and race (African-American, Caucasian or other). Viral suppression was defined as < 80 copies/mL. We also considered changes from baseline for each continuous variable as candidate predictors.

Outcome

The primary outcome was the change in three separate urine biomarkers, ACR, α1m and IL-18, which were measured at year 0 and at year 10 of this study. All urinary kidney injury biomarkers were measured at the Cincinnati Children's Hospital Medical Center Biomarker Laboratory. Urine albumin was measured by immunoturbidimetry using a Siemens Dimension Xpand plus HM clinical analyzer (Siemens, Munich, Germany). Urine α1m was measured by a commercially available assay (Siemens BN II Nephelometer; Siemens, Munich, Germany). Urine IL-18 was measured using a commercially available ELISA kit (Medical & Biological Laboratories Co., Nagoya, Japan). All urine specimens were in continuous storage without prior freeze-thaw. Laboratory personnel were blinded to clinical information about the participants and specimens were evaluated in random order. Assays were performed in 2010 for baseline samples and in 2015 for year 10 samples. We repeated 50 baseline measurements on specimens in 2015 and found no evidence for assay drift or bias in any of the assays. For this repeat testing, the Kendall coefficients of concordance W

test statistic (and p-value) were as follows: 0.94 (P<0.001) for IL-18, 0.94 (P<0.001) for urine albumin, 1.0 (P<0.001) for urine creatinine and 0.96 (P<0.001) for α1m. The intra-assay coefficients of variation for the urine measures were: albumin, 5.9%; α1m, 5.2% and IL-18, 5.2%.

Statistical analysis

Demographic and clinical characteristics were summarized descriptively at each study visit (year 0 and year 10). We used quantile regression to examine the patterns and distributions of change in each urine biomarker over 10 years of follow-up. This method enables comparison of biomarker concentrations at the median, quartiles and other percentiles from baseline to year 10 and is particularly helpful in exploring differences in the tails of distributions.

We identified patterns of changes in biomarker levels using k-means clustering to partition subjects into distinct groups. Our cluster construction was informed solely by the baseline and year 10 levels of each biomarker. We used the SAS FASTCLUS procedure to identify outliers and reduce their effect on cluster centers, using the strict option and cubic clustering criterion to determine the final number of clusters. For each biomarker, clusters were compared over the 10-year study period by plotting percentiles in order to compare relative differences within and between clusters. We used multinomial logistic regression to identify factors associated with cluster membership, using cluster 1 as the reference group. Finally, multivariable analysis of variance (MANOVA) was used to model predictors and patterns of change in all three biomarkers simultaneously. We standardized each biomarker (to a mean of zero and variance of one) so that measures with a larger variance would not have a greater influence on model estimates. Models were constructed using the SAS SYSLIN procedure for seemingly unrelated regression (SUR)[30] using the year 10 biomarker level as the dependent variable and the year 0 level as a covariate along with other factors of interest. The SUR model uses the correlations among the errors in different equations to improve efficiency of the regression estimates while enabling potentially different predictors for each dependent variable. Because many of the candidate covariates were inter-correlated, we used the least absolute shrinkage and selection operator (LASSO) method for variable selection[31] (R package glmnet). We then applied Bayesian Model Averaging (BMA) to the LASSO-selected predictors, retaining variables with a posterior probability of greater than 35%. We used the mlogitBMA package for R[32] to perform BMA for multinomial logit models. All other analyses were conducted using SAS (version 9.4, Cary, NC).

RESULTS

Demographic and clinical characteristics of the 294 women at baseline (year 0) and follow-up (year 10) are summarized in Table 1. Of note, over 10 years, the proportion of women diagnosed with hypertension nearly doubled, with an increase in the prevalence of anti-hypertensive use during this time. The prevalence of diabetes mellitus and HCV co-infection remained stable, and all serum cholesterol parameters (LDL, HDL and triglycerides) were on average, improved after 10 years. Use of ART increased over 10 years, with correspondingly large increases in CD4 cell count and the proportion of women with viral

suppression. There was very little change in eGFR by creatinine over 10 years (97 to 95 mL/min/1.73 m^2).

Figure 1 shows distributions of urine biomarker levels at baseline and year 10. Median IL-18 concentration declined by nearly 50% over 10 years; in contrast, median α1m concentration rose by approximately 50%. Although median ACR levels were similar at baseline and year 10, the distribution of values (10^{th}, 90^{th} percentiles) was somewhat wider at year 10 (2 to 92 mg/g) compared with baseline (4 to 43 mg/g). The cluster analysis identified four distinct patterns of biomarker change (Figure 2). Cluster 1 appeared to be the healthiest subset with the lowest levels of IL-18 and α1m throughout the follow-up period. In contrast, cluster 2 had the highest levels of both IL-18 and α1m. Cluster 3 was distinguished by having the highest urinary ACR levels at both time-points and α1m levels that rose during follow-up. Cluster 4 was notable for high baseline IL-18 levels that declined sharply during follow-up.

Next, we performed multivariable regression analysis to identify predictors of biomarker changes. Figure 3 depicts the relative importance of each candidate variable, which represents the posterior probability that each variable has a non-zero association with each biomarker outcome. Dominant factors for IL-18 changes included higher year 10 HIV viral load, higher baseline BMI, higher change in waist circumference, lower year 10 CD4 cell count and higher year 10 HDL cholesterol. The primary determinants of increases in α1m levels were the nadir CD4 cell count both prior to baseline and during the follow-up period. The most important determinant of increasing ACR values was the year 10 diastolic blood pressure.

Using variables selected by the LASSO and BMA procedures, we performed a multivariable-adjusted MANOVA regression analysis of changes from baseline to year 10 in all three biomarker levels (parsimonious model shown in Table 2, full model available in Supplemental Table 1). The selected predictors showed distinct associations with changes in each biomarker. Factors associated with increases in IL-18 were higher BMI at baseline, increasing waist circumference, lower HDL cholesterol at year 10, HCV co-infection, lower CD4 cell count and higher HIV viral load, both at year 10. Factors associated with increases in α1m included decreasing BMI over the study period, smoking tobacco at baseline, higher DBP at baseline, HCV co-infection, higher nadir CD4 cell count prior to baseline and decreasing nadir CD4 between year 0 and year 10. Finally, factors associated with increases in ACR included higher triglycerides at year 10, higher DBP at year 10, HCV infection and higher HIV viral load at year 10. Finally, factors associated with increases in ACR included higher triglycerides at year 10, higher DBP at year 10, HCV infection and higher HIV viral load at year 10. In sensitivity analyses, the findings were similar after indexing to urine creatinine. For α1m, the rise over 10 years appeared to be modestly stronger when evaluated using measures indexed for creatinine (see Supplemental Figure 1 and Supplemental Table 3).

We then used multinomial logistic regression analyses to identify factors that were independently associated with each of the clusters depicted in Figure 2 (with model shown in Supplemental Table 2), using the healthiest cluster (cluster 1) as the reference group. Characteristics significantly associated with greater odds of cluster 2 membership were

current smoking (OR=4.55, $P<0.001$) and a history of AIDS (OR=2.71, $P=0.006$), while higher time-averaged CD4 cell count was associated with a lower odds of cluster 2 membership (OR=0.51 per doubling of CD4, $P=0.006$). Current smoking was the only independent predictor of cluster 3 membership (OR=2.23, $P=0.03$), while other factors showed little association. Compared with cluster 1, higher time-averaged CD4 cell count was associated with lower likelihood of being in cluster four (OR=0.56 per doubling of CD4, $P=0.01$) compared with cluster one.

DISCUSSION

In this prospective cohort of women living with HIV, we found that the urine IL-18 concentration is a dynamic biomarker that decreased over 10 years of follow-up and appeared to be strongly influenced by changes in viral load and CD4 cell count in cluster analysis. In contrast, α1m levels rose over time, while ACR showed little change on average over the course of 10 years of follow-up. This is a novel finding in that each of these biomarkers demonstrated a distinct trajectory in these HIV-infected women over the same time period. Although IL-18 levels declined substantially over 10 years, particularly in those women with controlled viremia, this beneficial effect was mitigated by higher baseline BMI, increases in waist circumference, lower HDL cholesterol, higher year 10 viral load and lower year 10 CD4 cell count, and HCV co-infection. Levels of α1m increased on average over 10 years, and similarly, appeared to be influenced by HCV co-infection, and also higher baseline diastolic blood pressure, smoking and lower nadir CD4 cell count at both baseline and during the study period. Lower BMI at follow-up was also associated with increasing α1m, but the change in median BMI from year 0 to 10 was from 27 to 28 kg/m^2. Finally, although ACR levels were stable on average and remained low in most patients, they were more likely to increase over time in those with higher triglycerides, higher diastolic blood pressure, higher HIV viral load, and in those with HCV co-infection.

The decline in IL-18 paralleled improvements in viral control, and the mechanisms by which this occurs are not established. Since urine IL-18 concentrations have been found to be associated with both the incidence of CKD and mortality in the setting of HIV infection, they have been hypothesized to reflect both ongoing subclinical kidney injury and potentially to be a marker of systemic inflammation. It is noteworthy that IL-18 concentrations decreased substantially during the follow-up period, whereas limited changes were seen with ACR and eGFR[25]. Since HIV replication is known to occur in renal tubular cells[8], the findings of this study may support the proposed mechanism whereby HIV treatment leads to reductions in viral replication and lower local inflammation within the kidney.

The increases in α1m at 10 years of follow-up were an unexpected finding. Since CD4 cell counts and viral suppression dramatically improved over the 10-year study period, a pattern for α1m similar to IL-18 might have been expected. The divergent findings for IL-18 and α1m warrant additional investigation. In future studies we plan to measure additional time-points for the biomarkers in order to evaluate changes related to ART agents, such as tenofovir. ACR is a strong prognostic marker for morbidity and mortality in WIHS[25,33–38], yet in this study, it remained relatively stable over 10 years despite significant improvements

in control of HIV infection. Kidney function, as measured by eGFR, remained relatively constant over this time period as well, allowing us to explore those factors that affect markers of tubular and glomerular function and injury independent of established clinical measures of kidney disease. Over the same period, there was a significant increase in diagnosis and treatment of hypertension, and of note, we found that higher DBP was associated with increases in ACR. However, the lack of change in ACR over the decade of follow-up may reflect that HIV disease has a greater impact on tubular injury than on glomerular injury in this contemporary cohort of HIV-infected women. Prior research in WIHS has shown that all three biomarkers are strongly predictive of kidney function decline longitudinally, so their prognostic relevance has been established.

All of the changes in biomarkers in this study should be considered in the setting of relative improvements in HIV control. As patients were increasingly treated for HIV during this decade of observation, residual inflammation may have persisted due to ART-related toxicities or complications, and improved survival may have led to longer periods of at-risk time in order to develop chronic diseases that are pro-inflammatory. In contrast to prior work, where IL-18 was primarily driven by viral status[28], here we see an effect of metabolic disease on inflammation. These markers have been helpful in identifying evidence for kidney disease[4,6,22–25], and this work provides further evidence that the biomarkers could be valuable tools to monitor preclinical kidney disease status. In the evolution of a biomarker toward clinical practice, it must be shown to be not only prognostic, but also to be responsive to changes in health status.

There are some limitations to the interpretation of these data. We only measured three biomarkers out of many potential measures that could be indicative of kidney disease onset or progression. These biomarkers were chosen because they were associated with CKD and mortality in prior WIHS studies. This study only included women and therefore may not be readily generalizable to men. The first urine samples were collected over a decade before measurement; some degradation may lead to a non-differential error in quantification of urine biomarkers, but this is unlikely given the concordance noted on repeat testing from a sample of 50 specimens. Importantly, there is a possibility of residual confounding not accounted for in the modeling as well as a survival bias in the cohort and therefore the study sample. We did not focus on the relative impact of tenofovir exposure in this study, due to the limited exposure to tenofovir in this subset of participants at the time of biomarker measurement. Additionally, most participants in this cohort did not initiate treatment with TDF until 2005–2008. To more concretely understand the association of tenofovir exposure with these urine biomarkers of kidney injury, we plan to conduct measures of the biomarkers after additional years of follow-up to allow longer cumulative durations of tenofovir use.

There are several important strengths to this study. To our knowledge, this is the largest study to date of urine biomarker changes in HIV-infected women. Moreover, we had a long duration of follow-up from an ethnically and clinically diverse cohort. This is particularly important in the setting of understanding disease progression among different genetic susceptibilities for chronic kidney disease. In addition, we were able to consider a large number of possible predictors in helping to understand what factors contributed most to changes in biomarker levels over time, including status of HIV control and comorbidities.

In summary, we present data supporting that IL-18 is a dynamic urine biomarker that changes in tandem with control of HIV disease and that α1m may be a biomarker that reflects inflammation from a number of pathophysiological mechanisms. In contrast, ACR was relatively static over 10 years in this longitudinal cohort. Although further study is required to understand the effects of antiretroviral associated nephrotoxicity on changes in urine biomarkers, this work opens the door for additional research to understand patterns of biomarker change that may predict incidence and progression of CKD.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

We thank the Women's Interagency HIV Study (WIHS) participants who contributed data to this study. Data were collected by the WIHS Collaborative Study Group with centers (Principal Investigators at the time of data collection) at New York City/Bronx Consortium (Kathryn Anastos, MD); Brooklyn, NY (Howard Minkoff, MD); Washington DC, Metropolitan Consortium (Mary Young, MD); The Connie Wofsy Study Consortium of Northern California (Ruth Greenblatt, MD, Phyllis Tien, MD, and Bradley Aouizerat, PhD, MAS); Los Angeles County/ Southern California Consortium (Alexandra Levine, MD); Chicago Consortium (Mardge Cohen, MD); and Data Coordinating Center (Stephen J. Gange, PhD). The contents of this publication are solely the responsibility of the authors and do not necessarily represent the official views of the US National Institutes of Health.

Support: The WIHS Kidney Aging Study is funded by grant 1 R01 AG034853-01A2 (PI, Shlipak), which was administered by the Northern California Institute for Research and Education, and with resources of the Veterans Affairs Medical Center, San Francisco, California. Data in this manuscript were collected by the Women's Interagency HIV Study (WIHS). The contents of this publication are solely the responsibility of the authors and do not represent the official views of the National Institutes of Health (NIH). WIHS (Principal Investigators): UAB-MS WIHS (Michael Saag, Mirjam-Colette Kempf, and Deborah Konkle-Parker), U01-AI-103401; Atlanta WIHS (Ighovwerha Ofotokun and Gina Wingood), U01-AI-103408; Bronx WIHS (Kathryn Anastos), U01-AI-035004; Brooklyn WIHS (Howard Minkoff and Deborah Gustafson), U01-AI-031834; Chicago WIHS (Mardge Cohen and Audrey French), U01-AI-034993; Metropolitan Washington WIHS (Mary Young and Seble Kassaye), U01-AI-034994; Miami WIHS (Margaret Fischl and Lisa Metsch), U01-AI-103397; UNC WIHS (Adaora Adimora), U01-AI-103390; Connie Wofsy Women's HIV Study, Northern California (Ruth Greenblatt, Bradley Aouizerat, and Phyllis Tien), U01-AI-034989; WIHS Data Management and Analysis Center (Stephen Gange and Elizabeth Golub), U01-AI-042590; Southern California WIHS (Alexandra Levine and Marek Nowicki), U01-HD-032632 (WIHS I – WIHS IV). The WIHS is funded primarily by the National Institute of Allergy and Infectious Diseases (NIAID), with additional co-funding from the Eunice Kennedy Shriver National Institute of Child Health and Human Development (NICHD), the National Cancer Institute (NCI), the National Institute on Drug Abuse (NIDA), and the National Institute on Mental Health (NIMH). Targeted supplemental funding for specific projects is also provided by the National Institute of Dental and Craniofacial Research (NIDCR), the National Institute on Alcohol Abuse and Alcoholism (NIAAA), the National Institute on Deafness and other Communication Disorders (NIDCD), and the NIH Office of Research on Women's Health. WIHS data collection is also supported by UL1-TR000004 (UCSF CTSA) and UL1-TR000454 (Atlanta CTSA). S.M.B. is supported by the UCSF Traineeship in AIDS Prevention Studies (US National Institutes of Health (NIH) T32 MH-19105). This research was also supported by a grant from the National Institutes of Health, University of California, San Francisco-Gladstone Institute of Virology & Immunology Center for AIDS Research (CFAR), P30-AI027763 (Principal Investigators Paul Volberding and Warner Greene). The funders did not play a role in study design, collection, analysis, interpretation of data, writing the report or the decision to submit the report for publication.

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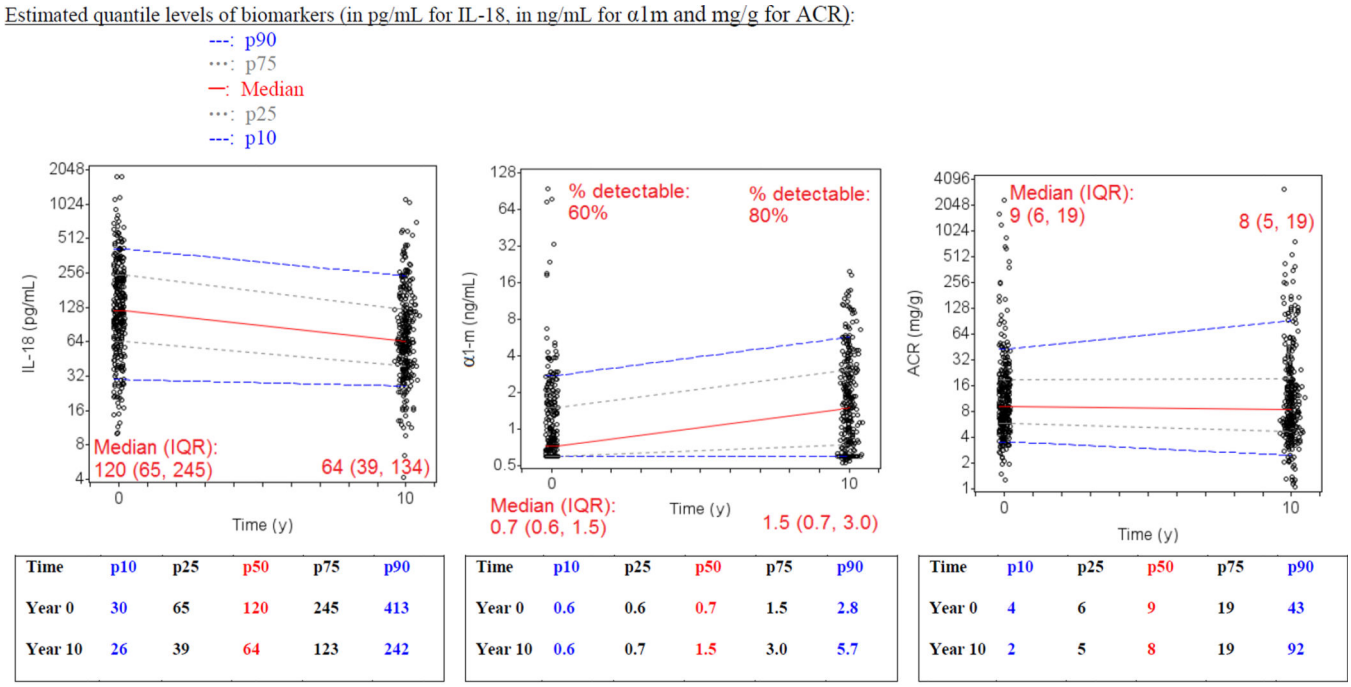


Time	p10	p25	p50	p75	p90
Year 0	30	65	120	245	413
Year 10	26	39	64	123	242

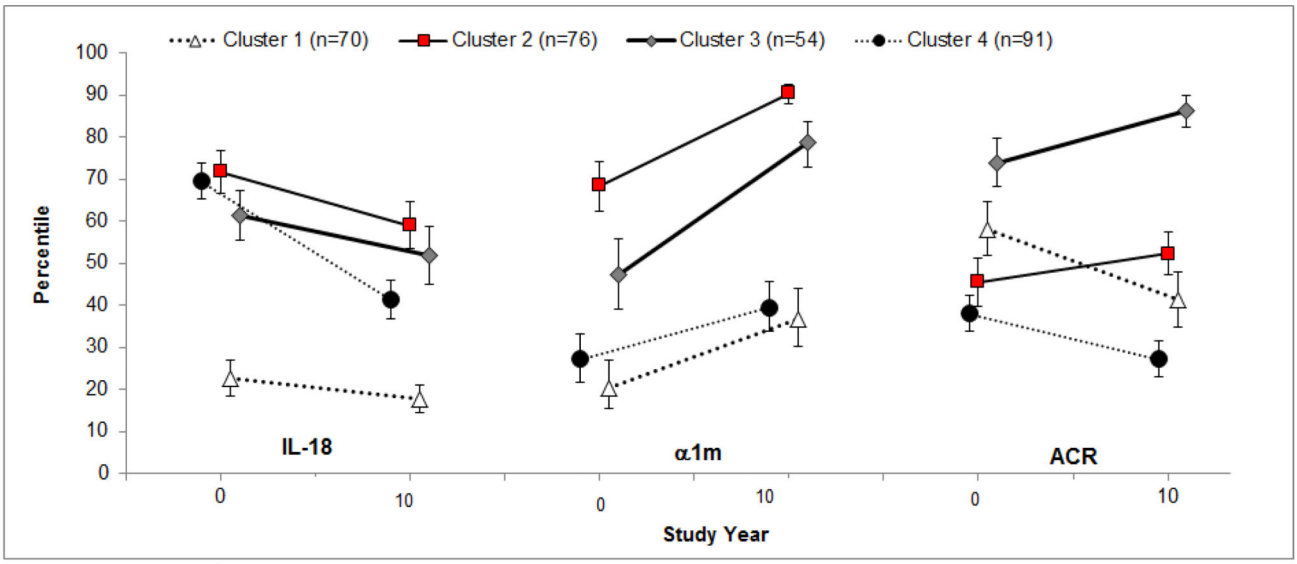
Time	p10	p25	p50	p75	p90
Year 0	0.6	0.6	0.7	1.5	2.8
Year 10	0.6	0.7	1.5	3.0	5.7

Time	p10	p25	p50	p75	p90
Year 0	4	6	9	19	43
Year 10	2	5	8	19	92

Figure 1.

Comparison of urine biomarker level changes in HIV-infected women at baseline and 10 year follow up.

Abbreviations: IL-18, urine interleukin 18; α1m, urine alpha-1-microglobulin; ACR, urine albumin:creatinine ratio; y, year; IQR, interquartile range



Actual values (median, IQR):

	IL-18 (pg/mL)		α1m (ng/mL)		ACR (mg/g)	
	Year 0	Year 10	Year 0	Year 10	Year 0	Year 10
Δ Cluster 1 (N = 70)	40 (23, 60)	35 (25, 52)	0.6 (0.6, 0.8)	0.8 (0.5, 1.3)	14 (7, 24)	7 (4, 16)
■ Cluster 2 (N = 76)	207 (111, 349)	111 (66, 229)	1.7 (1.0, 2.7)	4.2 (2.8, 6.2)	8 (5, 14)	10 (6, 16)
♦ Cluster 3 (N = 54)	112 (92, 221)	91 (57, 195)	0.8 (0.6, 1.5)	2.2 (1.4, 3.3)	25 (13, 52)	88 (26, 162)
● Cluster 4 (N = 91)	170 (115, 293)	63 (43, 117)	0.6 (0.6, 0.8)	0.8 (0.5, 1.3)	6 (5, 10)	5 (3, 7)

Figure 2.

Estimates (95% CI) plotted represent percentiles of change for each urine biomarker over 10 years of follow-up

Abbreviations: IL-18, urine interleukin 18; α1m, urine alpha-1-microglobulin; ACR, urine albumin:creatinine ratio; IQR, interquartile range

Percentiles were computed within the full sample using all available measurements at year 0 and 10, then plotted separately within each cluster. For IL-18 the median change (95% CI in pg/mL) for clusters 1, 2, 3 and 4 were −3 (−29, 17), −60 (−204, 33), −37 (−100, 17) and −94 (−207, −37), respectively. For α1m the median change (95% CI in ng/mL) for clusters 1, 2, 3 and 4 were 0.02 (0.00, 0.53), 2.2 (0.6, 4.3), 0.98 (0.27, 2.5) and 0.04 (0.00, 0.50), respectively. For ACR the median change (95% CI in mg/g) for clusters 1, 2, 3 and 4 were −3.6 (−16, 2.6), 0.8 (−4.2, 8.5), 44 (−20, 115) and −1.5 (−4.1, 0.9), respectively.

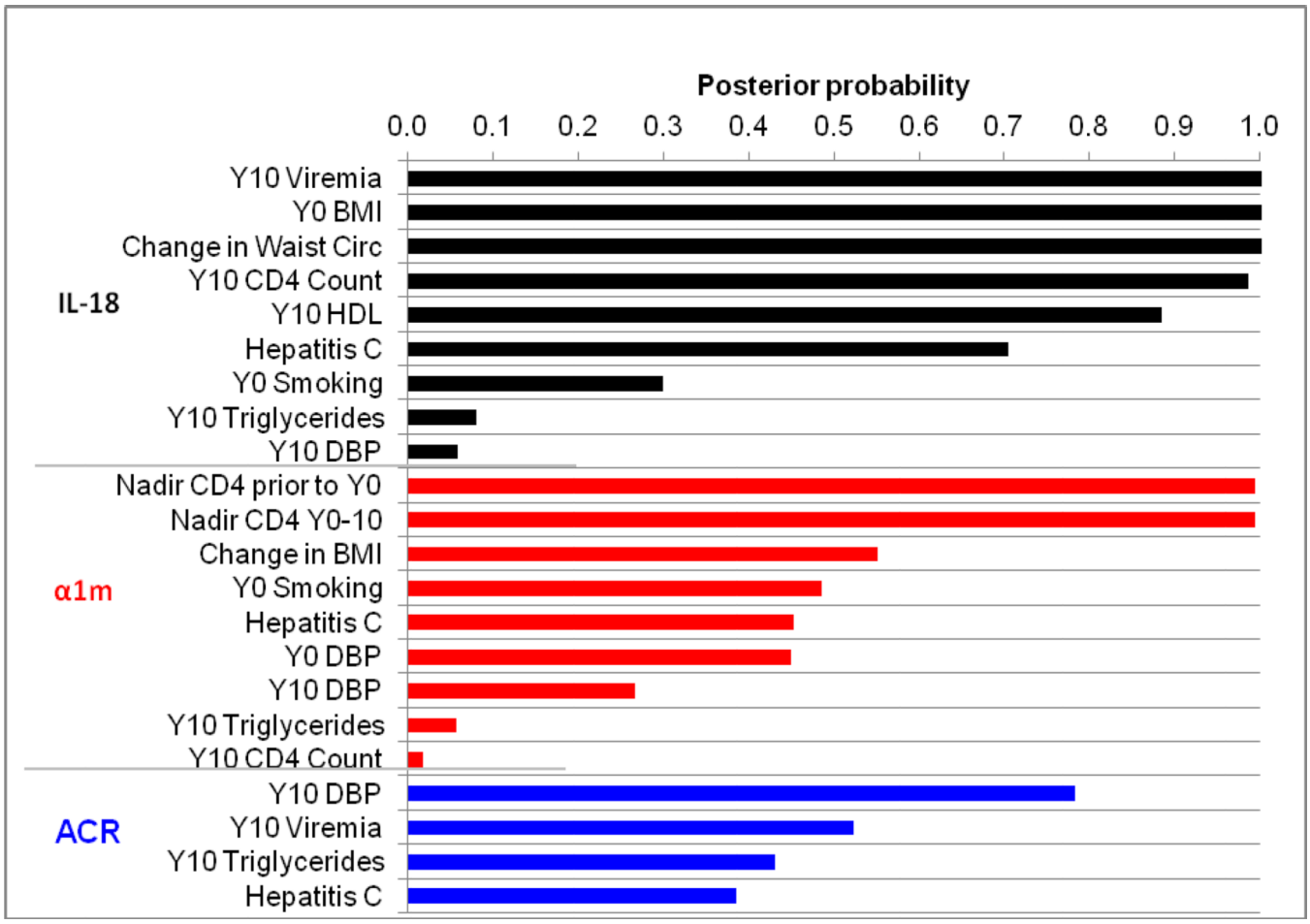


Figure 3.

Relative importance of variables selected by LASSO and Bayesian Model Averaging. Abbreviations: IL-18, urine interleukin 18; α1m, urine alpha-1-microglobulin; ACR, urine albumin:creatinine ratio; Y0, baseline year 0; Y10, year 10 follow-up; BMI, body mass index; DBP, diastolic blood pressure

Table 1

Demographic and clinical characteristics of HIV-infected women in biomarker follow-up study at baseline (year 0) and follow-up at year 10.

Parameter	Year 0 N = 294	Year 10 N = 294
Age (years)	41 (36, 45)	50 (46, 54)
Race		
African-American	176 (60%)	176 (60%)
Caucasian	66 (22%)	66 (22%)
Other	52 (18%)	52 (18%)
Menopausal	50 (17%)	133 (47%)
Cigarette smoking history		
Current	146 (50%)	110 (37%)
Past	81 (28%)	121 (41%)
Never	67 (23%)	63 (21%)
Diagnosis of diabetes mellitus	84 (29%)	84 (29%)
Systolic blood pressure (mm Hg)	120 (110, 129)	119 (109, 133)
Diastolic blood pressure (mm Hg)	72 (68, 80)	74 (67, 82)
Diagnosis of hypertension	71 (24%)	131 (45%)
Antihypertensive use	32 (11%)	104 (35%)
Serum LDL cholesterol (mg/dL)	116 (85, 143)	99 (77, 124)
Serum HDL cholesterol (mg/dL)	45 (36, 55)	53 (41, 64)
Serum triglyceride (mg/dL)	126 (87, 174)	110 (86, 156)
Body mass index (kg/m^2)	27 (23, 32)	28 (24, 33)
Waist circumference (centimeters)	88 (80, 100)	94 (85, 107)
Current HAART use	186 (64%)	264 (90%)
History of AIDS	149 (51%)	181 (62%)
Hepatitis C virus infection	79 (27%)	79 (27%)
Current CD4 cell count (cells/microliter)	394 (269, 587)	520 (326, 698)
Lifetime nadir CD4 cell count (cells/microliter)	240 (133, 337)	163 (79, 258)
Current HIV RNA < 80 copies/mL	95 (32%)	196 (67%)
Lifetime peak HIV RNA > 10,000 copies/mL	221 (75%)	255 (87%)
eGFRcr ($mL/min/1.73\ m^2$)	97 (82, 112)	95 (76, 109)

Data are presented as Median (IQR) or numbers (percent).

Abbreviations: IQR, interquartile range; eGFRcr, estimated glomerular filtration rate by serum creatinine; HAART, highly active antiretroviral therapy

Table 2

Characteristics associated with change from baseline to year 10 in urine biomarker levels using multivariable-adjusted MANOVA.

Variable	IL-18 % Estimate (95%CI)	α1m % Estimate (95%CI)	ACR % Estimate (95%CI)
Parsimonious model, non-significant factors dropped			
Y0 body mass index (kg/m^2)	**2.5 (1.48, 3.5) *P*<0.001**		
Change from baseline body mass index (kg/m^2)		**−2.1 (−3.6, −0.55) *P*=0.008**	
Change from baseline waist circumference (per 10 cm)	**15.3 (7.8, 23.2) *P*<0.001**		
Y10 HDL (per 10 mg/dL)	**−5.6 (−9.2, −1.81) *P*=0.004**		
Y10 triglycerides (per doubling)			**11.5 (0.90, 23.1) *P*=0.03**
Y0 smoking		**22.3 (5.2, 42.1) *P*=0.009**	
Y0 diastolic blood pressure (per 10 mm Hg)		**7.3 (0.80, 14.2) *P*=0.03**	
Y10 diastolic blood pressure (per 10 mm Hg)			**9.0 (1.95, 16.6) p=0.01**
Hepatitis C virus infection	**27.3 (9.7, 47.7) *P*=0.002**	**25.6 (6.4, 48.4) *P*=0.007**	**22.7 (4.1, 44.7) *P*=0.02**
Nadir CD4 prior to baseline (per doubling)		**9.5 (3.8, 15.6) *P*=0.001**	
Nadir CD4 between Y0 and Y10 (per doubling)		**−10.1 (−14.2, −5.7) *P*<0.001**	
Y10 CD4 Count (per doubling)	**−10.7 (−16.5, −4.5) *P*=0.001**		
Y10 HIVRNA (per 10 fold increase)	**19.2 (10.1, 29.0) *P*<0.001**		**9.7 (1.37, 18.7) *P*=0.02**
Adjusted R^2	0.33	0.23	0.18

Abbreviations: IL-18, urine interleukin 18; α1m, urine alpha-1-microglobulin; ACR, urine albumin:creatinine ratio; Y0, baseline year 0; Y10, year 10 follow-up