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Early Changes in T-Cell Activation Predict Antiretroviral Success in Salvage Therapy of HIV Infection

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Abstract

Objective—Because effective antiretroviral therapy (ART) reduces immune activation, we hypothesize that early changes in immune activation are associated with subsequent virologic response to therapy.

Design—Observational cohort study.

Setting—Institutional HIV clinic.

Subjects—Thirty-four adult HIV patients with virologic failure on their current antiretroviral regimen.

Intervention—Change to salvage regimen selected by patient's physician.

Main Outcome Measures—Measures of immune activation at baseline and at 2, 4, 8, and 24 weeks after enrollment. Data were analyzed by proportional hazards (PH) models.

Results—PH models showed that reductions between baseline and week 2 in expression of CD38 ($P = 0.02$) or CD95 ($P = 0.02$) on $CD4^+$ T cells were associated with increased likelihood of achieving virologic suppression. Kaplan-Meier analysis demonstrated that patients who had reductions within the first 2 weeks of therapy in $CD4^+$ T-cell expression of CD38 ($P = 0.003$) or CD95 ($P = 0.08$) were more likely to achieve viral suppression than those who did not.

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AUTHOR CONTRIBUTIONS

A. D. Badley, M. R. Loutfy, F. Mandy, C. M. Kovacs, and M. Bergeron participated in the design and implementation of the study; the collection, compilation, and interpretation of data; and the preparation of the manuscript. V. Govan and J. B. Angel participated in the implementation of the study; the collection, compilation, and interpretation of data; and the preparation of the manuscript. B. D. Shepard and J. Raboud participated in the compilation and interpretation of data and the preparation of the manuscript. C. Diong and S. A. Rizza participated in the data interpretation and manuscript preparation.

A. D. Badley had full access to all the data in the study and had final responsibility for the decision to submit for publication. None of the authors have a commercial association or other association that might pose a conflict of interest. The funding sources had no involvement in the study design; in the collection, analysis, or interpretation of data; in the writing of the manuscript; or in the decision to submit the manuscript for publication.

Conclusions—Reduced $CD4^+$ T-cell expression of CD38 and CD95 occurring within 2 weeks of salvage therapy is associated with subsequent viral suppression. Monitoring CD38 and CD95 may allow earlier assessment of the response to ART.

Keywords

antiretroviral therapy; clinical trial; HIV; immune activation; prognosis; virologic failure

The management of HIV-infected patients is guided largely by 2 key measures: the amount of viral replication, which predicts the rate of disease progression, and the degree of immune system dysfunction as measured by absolute $CD4^+$ T-cell number.[1] The continuing development of effective anti-retroviral therapies (ARTs) has allowed pharmacologic suppression of HIV replication in many infected patients, often leading to quantitative and qualitative reconstitution of the immune system. Ongoing viral replication in the presence of incompletely suppressive drugs, when coupled with the labile genetic nature of HIV, may lead to resistance mutations, loss of virologic control,[2–5] decreased $CD4^+$ T-cell number, and significant morbidity and mortality, however.[6,7] Moreover, once resistance occurs, the likelihood of extended resuppression of viral replication with subsequent therapies is substantially lower.[8,9] With the current practice of monitoring only viral replication and $CD4^+$ T-cell number, it may take several months to determine whether or not a new antiretroviral regimen is effective.[1] Therefore, identifying markers that accurately predict at early time points whether there is likely to be eventual viral suppression, particularly in situations in which salvage drug regimens are required, would be of great clinical utility.

There has been considerable interest in evaluating markers of immune function to aid in the management of patients infected with HIV. Several immunologic markers have been proposed as predictors of disease outcome, including serum p24 antigen levels, serum tumor necrosis factor (TNF)-α and TNFγ levels, CD38 antigen intensity on $CD8^+$ T cells, and a variety of other T-cell activation markers (eg, CD95, human leukocyte antigen D-related [HLA-DR], CD28).[10–21] Not all the previous studies evaluated patients who had previously received ART,[11,13,16] however, and, often, only late time points (eg, ≥24 weeks after therapy) were analyzed.[10,12,14,15,17,18,20] Although a study that analyzed earlier time points (beginning at 12 weeks) showed that the $CD8^+CD38^+$ T-cell count is a prognostic marker of therapeutic failure in HIV-infected children,[22] no studies have evaluated the prognostic value of early changes in these immune markers in adult HIV-positive patients over time. Thus, it is unknown whether early changes in such markers in adult patients add prognostic information to the current standard clinical practice of monitoring $CD4^+$ T-cell counts and HIV viral load at intervals of every few months.

With the goal of identifying whether changes in immunologic markers that occur early after initiation of salvage therapy are associated with subsequent virologic response to therapy, a single-center cohort study was initiated. Multivariate flow cytometry was used to assess immune activation and T-cell subsets before and during salvage therapy to identify immunologic markers that predict within the first few weeks the subsequent restoration of virologic control.

METHODS

Study Population and Intervention

Patients were enrolled from September 1, 2001 through December 1, 2001 from the Maple Leaf Medical Clinic (Toronto, Ontario, Canada). The study (Clinical Trials Registry NCT00440206) was reviewed and approved by the respective Institutional Review Board (IRB), IRB Services, and the Ottawa Hospital. Patients providing written informed consent

and meeting the following inclusion criteria were enrolled in the study: (1) previously documented HIV infection, (2) age ≥18 years, (3) treatment with the same anti-retroviral regimen (including ≥3 medications) for at least 3 months, (4) virologic failure defined as viral load ≥50 copies/mL on 2 occasions at least 2 weeks apart, and (5) plans by the patient and patient's attending physician to switch from the current antiretroviral regimen to a salvage regimen (ie, ART initiated after failure of a prior regimen). Exclusion criteria included patients with (1) hemoglobin <100 g/L, (2) platelet count <20,000 cells/μL, (3) international normalized ratio (INR) ≥3.5, or (4) partial thromboplastin time (PTT) ≥60 IU. For each patient, the study concluded when either of the following was met: (1) 12 months had elapsed since the date of enrollment or (2) the salvage antiretroviral regimen was discontinued or changed at the discretion of the patient and/or the patient's physician.

Laboratory Measurements and Clinical Evaluation

Viral load assessment was determined by measuring the plasma HIV-1 RNA level using the branched-DNA Chiron 3.0 assay (Chiron, Emeryville, CA) with a lower limit of detection of 50 copies/mL. For each patient, viral load measurements were made at the time of the study enrollment (before initiation of salvage regimen, week 0) and at 2, 4, 8, 24, and 48 weeks after initiation of the new regimen. A clinical evaluation and safety and toxicity laboratory evaluations for monitoring ART were performed by the patient's physician according to standard clinical care guidelines at the time of the study.

Immunophenotypic Assays

Immunophenotypic analysis was performed using 2 4-color immunophenotyping panels at baseline and at 2, 4, 8, and 24 weeks of follow-up. The first panel (panel A) consisted of the following commercially premixed antibodies: CD45-fluorescein isothiocyanate (FITC)/CD4-RD1/CD8-ECD/CD3-PC5 and CD45-FITC/CD56-RD1/CD19-PE-TxRed (ECD)/CD3-PC5 (Beckman Coulter, Fullerton, CA). The second panel (panel B) was prepared with the following antibodies: CD45RA-FITC, HLA-DR-FITC (BD Biosciences, Fullerton, CA) and CD4-ECD, CD8-PC5, CD45RO-phycoerythrin (PE), CD28-FITC, CD62L-PE, CD69-PE, CD38-PE, and CD45RO-PE (Beckman Coulter).

Samples were prepared by mixing each venous blood sample with antibodies from panel A or panel B and incubating for 10 minutes at room temperature before lysing with the automated ImmunoPrep reagent system (Beckman Coulter). Samples were fixed with 0.5 mL of 2% paraformaldehyde solution. For measurements with panel A, 100 μL of Flow-Count (Beckman Coulter) absolute count beads was added before analysis to allow for measurement of absolute cell counts as a single-platform procedure.

Flow cytometry was performed with Coulter Epics XL-MCL (Beckman Coulter). The instrument was prepared using QC3 reference bead standards (Bangs Laboratories, Fishers, IN) to establish target channels and standardize the fluorescence intensity analysis range throughout the study. Lymphocytes gates were set in panel A using the universal template CD45 and CD4 double-anchor gate.[23] Absolute and relative cell values were measured. The $CD4^+$ and $CD8^+$ T-cell gates in panel B were defined with the heterogeneous gating strategy, where CD4 and CD8 bright fluorescence expression was expressed against the side-scattered light. All results were expressed as percentages of CD4 and bright CD8 cells. In addition, the mean fluorescence intensity of each of the activation markers HLA-DR, CD38, CD28, CD95, and CD69 was measured.

Statistical Analysis

Associations between baseline characteristics and subsequent virologic suppression were calculated using the χ^2 test or Fisher exact test for categoric covariates and the Wilcoxon

rank sum test for continuous covariates. Virologic suppression was defined to be a viral load <50 copies/mL at any time during the study. Patients were classified as virologic "responders" if they achieved virologic suppression during the study period; otherwise, they were classified as "nonresponders." Cox proportional hazards (PR) models with fixed covariates and time-dependent covariates and Kaplan-Meier analyses were used to determine the associations of covariates with time to achieve virologic suppression.

RESULTS

Thirty-four patients were enrolled in the study. Two patients did not complete follow-up. All patients were male, 27 (84%) of the 32 patients were white, and the median age was 43 years (interquartile range: 39 to 48 years). Nineteen (59%) of the 32 patients achieved virologic suppression during the study. Of these 19, 14 had evaluable viral load data at week 2, and 5 of 14 achieved viral suppression by that time point. Nine (64%) of 14 patients who ultimately achieved virologic suppression did not achieve virologic suppression by week 2. The baseline demographics and clinical characteristics of the study population are shown in Table 1. Patients who achieved virologic suppression were less likely to have a history of an AIDS-defining illness ($P < 0.01$), had a lower baseline viral load ($P < 0.01$), and had a higher baseline $CD4^+$ T-cell count ($P = 0.03$). Higher baseline expression of CD38 on CD4 ($P = 0.04$) and CD8 ($P = 0.01$), higher expression of CD38 mean fluorescence intensity on CD8 ($P = 0.03$), and lower expression of CD28 on CD4 ($P = 0.02$) were associated with a decreased likelihood of virologic suppression. Other baseline demographic and clinical covariates were not different between groups.

Univariate PH models were used to determine the associations of baseline demographic and clinical characteristics with time to virologic suppression (Table 2). Patients with a history of an AIDS-defining illness (hazard ratio [HR] = 0.32; $P = 0.01$) and those with a higher baseline viral load (HR = 0.56 per $\log_{10}$ copies/mL; $P < 0.01$) were less likely to achieve virologic suppression. Patients with a $CD4^+$ T-cell count ≥100 cells/mm^3 at baseline were more likely to achieve virologic suppression (HR = 5.00; $P = 0.01$) than patients with a $CD4^+$ T-cell count <100 cells/mm^3. Univariate PH models with time-dependent covariates were used to estimate the HR of virologic suppression associated with immune markers (Table 3). Continued high expression of CD38 on $CD4^+$ and $CD8^+$ T cells (HR = 0.73 per 10 cells/mm^3; $P = 0.01$ and HR = 0.80 per 10 cells/mm^3; $P = 0.02$, respectively) and increased expression of CD95 on $CD4^+$ T cells (as measured by mean fluorescence intensity, HR = 0.81 per cell/mm^3; $P = 0.05$) during the course of the study were associated with a decreased likelihood of virologic suppression.

Univariate PH models with covariates for changes in immunologic markers from baseline to week 2 identified associations between changes within the first 2 weeks of salvage therapy and virologic suppression (Table 4). Decreased expression of HLA-DR and CD95 (the latter measured by mean fluorescence intensity) on $CD4^+$ T cells during the first 2 weeks of salvage therapy was associated with increased likelihood of virologic suppression (HR = 0.33 per 10 cells/mm^3; $P = 0.02$ and HR = 0.78 per 1 cell/mm^3; $P = 0.02$, respectively). As at baseline, decreased levels of CD38 on $CD4^+$ T cells (as measured by mean fluorescence intensity) continued to be associated with an increased likelihood of achieving virologic suppression (HR = 0.66 per 10 cells/mm^3; $P = 0.02$).

In a multivariate PH model, patients with baseline $CD4^+$ T cells ≥200 cells/mm^3 remained much more likely to achieve virologic suppression (HR = 11.93; $P = 0.003$). After controlling for baseline $CD4^+$ T-cell counts in multivariate modeling, decreases in expression of CD38 on $CD4^+$ T cells between baseline and week 2 were associated with an increased likelihood of virologic suppression (HR = 2.13 per 10 cells/mm^3; $P = 0.007$).

Moreover, a decrease in expression of CD95 on $CD4^+$ T cells between baseline and 2 weeks of salvage therapy was associated with an increased likelihood of subsequent virologic suppression (HR = 1.54 per cell/mm^3; $P = 0.007$). Kaplan-Meier analysis confirmed that changes in CD38 on $CD4^+$ T cells within the first 2 weeks of salvage ART are associated with subsequent viral suppression ($P = 0.003$; Fig. 1A). Although Kaplan-Meier analysis demonstrated a similar trend for changes in CD95 on $CD4^+$ T cells within the first 2 weeks of salvage ART, the trend was not statistically significant ($P = 0.08$; see Fig. 1B).

DISCUSSION

Although other studies have highlighted an association between decreased immune activation and virologic suppression and/or immune reconstitution, it has been unknown how early such changes occur and whether they can be used to predict antiviral response. The present study is the first to show that decreased immune activation within the first 2 weeks of salvage therapy is associated with, and predictive of, subsequent virologic suppression. We show in a multivariate model controlling for baseline $CD4^+$ T-cell count that decreases in CD38 expression on $CD4^+$ T cells (with a similar trend for decreased expression of CD95 on $CD4^+$ T cells) after 2 weeks of salvage therapy were predictive of an increased likelihood of achieving virologic suppression. Importantly, these changes in activation occurred independent of viral suppression, because, at week 2, when measures of immune activation were improved, 64% of patients still had detectable viral replication. These data support the hypothesis that early measures of immune activation can be used to predict patients who ultimately suppress viral replication after initiation of salvage therapy. When compared with measures of $CD4^+$ T-cell count and viral load, which are measured at much later time points after initiation of salvage therapy (commonly 1 to 3 months later), the prognostic value of early measures of CD38 and CD95 expression by $CD4^+$ T cells may provide a distinct clinical advantage by allowing earlier assessment of the response to changes in ART.

The results of this study further highlight the relation between HIV disease progression, immune dysfunction, and T-cell activation, because we show that persistent immune activation predicts ongoing viral replication and progressive dysfunction of the $CD4^+$ T-cell population.[19,24–26] In a study of $CD4^+$ T-cell recovery in HIV-1–infected patients after 12 months of highly active antiretroviral therapy (HAART), incomplete $CD4^+$ T-cell recovery, despite suppression of viral replication with HAART, was associated with ongoing $CD4^+$ T-cell activation.[27] Persistent $CD4^+$ but not $CD8^+$ T-cell activation was associated with decreased $CD4^+$ T-cell recovery during the first 3 months of HAART.[28] Similarly, several recent studies have further confirmed the relation between limited immune restoration and higher levels of $CD4^+$ T-cell activation.[11,17,29,30] Each of these studies examined the relation between T-cell activation and subsequent immunologic recovery, however, without specifically examining direct relations between T-cell activation and virologic suppression.

Our data, and those of previous reports, suggest a link between ongoing viremia and persistent T-cell activation. Several studies have highlighted mechanisms mediating this link. A recent study identified bacterial translocation as a mechanism of chronic immune activation in the context of a compromised gastrointestinal mucosal surface during chronic HIV infection.[31] It is also likely that the association between ongoing viremia and persistent T-cell activation occurs in response to virally produced proteins and/or host-produced cytokines that are elaborated during virus-host interactions. Consistent with that hypothesis, CD38- and CD95-mediated T-cell activation can be induced by Tat,[32–34] Nef,[35,36] and gp120[34] as well as by altered production of a wide variety of cytokines, such as TNFα, interferon (IFN)-γ, interleukin (IL)-1β, IL-2, IL-6, IL-10, and IL-12, produced by chronically activated $CD8^+$ T cells and macrophages during HIV infection.[37–42]

Consequently, as levels of these immunomodulators decrease in conjunction with effective therapy, the levels of CD38 and CD95 decrease.[18] Taken together, these data demonstrate the potential clinical utility of early measures of activation (changes in CD95 and/or CD38 between baseline and week 2) as predictors of virologic suppression after initiation of salvage therapy.

Similarly, different levels of $CD4^+$ T-cell activation at baseline were recently reported in another study. Baseline proportions of $CD4^+$ T-cell activation were not significantly different among immunologically concordant versus discordant patients before initiation of ART.[17] When immune cell subsets were measured by absolute cell counts, however, discordant patients showed increased activation of $CD4^+$ and $CD8^+$ T cells at baseline, suggesting that baseline measurements of activation may prove clinically useful in predicting $CD4^+$ T-cell recovery.[17] Follow-up immunophenotypic data were measured after 48 weeks of successful (HIV viral load ≤50 copies/mL) ART,[17] and there was no comparison of those patients who achieved virologic suppression versus those who did not. Similarly, AIDS Clinical Trials Group (ACTG) 384 showed that persistent T-cell activation was associated with decreased T-cell recovery after ART in treatment-naive patients.[43] The authors acknowledge that because $CD4^+$ T-cell subsets were first assessed 6 months after treatment initiation, they “may have missed early changes”.[43]

Several limitations must be considered in appropriate interpretation of the data, some of which suggest further areas of research. Two of 34 patients did not complete any follow-up evaluations during the study and were thus excluded from the analysis. The study population was homogeneous, including only middle-aged, primarily white, men. Nonetheless, the results of the study are consistent with those reported in a pediatric study, which showed that the $CD8^+CD38^+$ T-cell count is a prognostic marker of therapeutic failure in HIV-infected children.[22] There was a trend toward more frequent use of protease inhibitors (PIs) in those therapies that ultimately failed to achieve virologic suppression. Although the present study demonstrated a decline in immune activation markers during early effective salvage therapy, further studies should be initiated to define the generalizability of these findings in diverse patient populations, who are treated with a range of salvage therapies, and to determine whether the predictive ability of these changes are present in naive patients initiating therapy

The present study adds to the body of data that highlights the importance of persistent T-cell activation in HIV pathogenesis.[28,44,45] Three features make the present study unique. First, although other studies have focused on the impact of persistent T-cell activation on eventual T-cell recovery, the present study investigated the relation between T-cell activation and eventual virologic suppression. Second, the present study focused on patients undergoing salvage therapy, whereas other studies of T-cell activation and HIV disease progression have typically included only treatment-naive patients. Third, the present study examined early time points (2, 4, 8, 24, and 48 weeks) after initiation of salvage therapy, whereas other studies have generally focused on time points beginning several months after the initiation of an initial anti-retroviral regimen. Based on these early data points, the present study reports that measures comparing T-cell activation between baseline and 2 weeks of salvage ART are sufficient for predicting those individuals who eventually achieve control of HIV replication, which is the true measure of disease progression or control.

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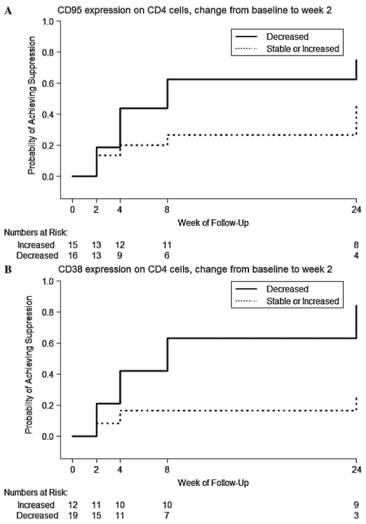

FIGURE 1.

A, Probability of viral suppression stratified by change in CD38 expression on $CD4^+$ T cells between baseline and week 2. B, Probability of viral suppression stratified by change in CD95 expression on $CD4^+$ T cells between baseline and week 2.

TABLE 1

Comparison of Demographic and Clinical Characteristics by Virologic Responder Status

Characteristics	Nonresponder (n = 13)	Responder (n = 19)	Total (n = 32)	P
Male	13 (100.0%)	19 (100.0%)	32 (100.0%)	—
Age, y	40 (39 to 42)	45 (41 to 53)	43 (39 to 48)	0.05
Race				0.85
White	11 (84.6%)	16 (84.2%)	27 (84.4%)	
Black	1 (7.7%)	2 (10.5%)	3 (9.4%)	
Asian	1 (7.7%)	0 (0.0%)	1 (3.1%)	
Other	0 (0.0%)	1 (5.3%)	1 (3.1%)	
HIV risk factor				0.46
MSM	11 (91.7%)	15 (83.3%)	26 (86.7%)	
Heterosexual	1 (8.3%)	0 (0.0%)	1 (3.3%)	
Blood transfusion	0 (0.0%)	1 (5.6%)	1 (3.3%)	
MSM + IVDU	0 (0.0%)	2 (11.1%)	2 (6.7%)	
Missing	1	1		
AIDS-defining illness	12 (92.3%)	8 (42.1%)	20 (62.5%)	0.01
Hepatitis B	1 (7.7%)	1 (5.3%)	2 (6.3%)	0.99
Hepatitis C	1 (7.7%)	3 (15.8%)	4 (12.5%)	0.63
No. years using ART	10 (8 to 14)	11.5 (8 to 13)	11 (8 to 13)	0.95
No. previous antiretroviral drugs	11 (10 to 12)	8 (7 to 11)	10 (7 to 12)	0.07
No. previous regimens	7 (6 to 11)	6.0 (4.0 to 7.0)	6.0 (5.0 to 9.5)	0.12
Present regimen includes PI	13 (100.0%)	13 (68.4%)	26 (81.3%)	0.06
Present regimen includes NNRTI	8 (61.5%)	11 (57.9%)	19 (59.4%)	0.84
Baseline viral load, $\log_{10}$ copies/mL	5.0 (4.9 to 5.5)	3.7 (2.7 to 4.7)	4.3 (3.2 to 5.0)	<0.01
Baseline CD4 count, cells/mm^3	43 (20 to 181)	230 (132 to 350)	191 (50 to 276)	0.03
Baseline CD8 count, cells/mm^3	560 (421 to 861)	748 (420 to 1240)	710 (421 to 965)	0.39
$CD3^+$ mean	75.5 (74.4 to 81.6)	75.8 (69.4 to 82.9)	75.7 (72.2 to 82.3)	0.89
$CD38^+$ on CD4	85.6 (66.8 to 87.0)	64.6 (48.4 to 78.8)	71.4 (55.7 to 85.0)	0.04
HLA-DR on CD4	12.7 (7.2 to 16.7)	7.8 (6.4 to 13.5)	10.4 (6.9 to 16.2)	0.39
$CD38^+$ on CD8	94.1 (89.1 to 95.6)	70.0 (46.7 to 84.1)	77.4 (48.2 to 94.3)	0.01
HLA-DR on CD8	22.6 (19.9 to 32.5)	18.6 (13.4 to 27.9)	21.1 (16.4 to 28.0)	0.15
$CD28^+$ on CD4	51.9 (48.5 to 65.6)	76.9 (57.5 to 89.4)	65.2 (50.1 to 86.5)	0.02
$CD95^+$ on CD4	62.1 (58.5 to 70.3)	63.5 (56.1 to 83.6)	62.8 (57.2 to 76.2)	0.82
$CD95^+$ on CD8	60.1 (40.6 to 67.4)	56.4 (34.4 to 61.3)	56.8 (36.9 to 63.6)	0.37
$CD69^+$ on CD4	12.2 (6.7 to 14.9)	15.4 (7.3 to 21.8)	13.9 (6.8 to 19.5)	0.35
$CD69^+$ on CD8	21.8 (13.4 to 26.2)	17.8 (12.5 to 20.6)	18.2 (12.8 to 23.8)	0.29
CD38 MnI on CD4	21.8 (15.6 to 26.0)	16.9 (8.7 to 22.5)	18.5 (12.8 to 24.6)	0.22
CD38 MnI on CD8	36.6 (18.6 to 43.1)	14.4 (5.3 to 24.2)	20.5 (9.2 to 36.9)	0.03
CD95 MnI on CD4	6.0 (5.2 to 7.9)	6.1 (4.9 to 6.9)	6.0 (4.9 to 7.4)	0.70

Quantities in N (%) or median.

Interquartile ranges are reported with medians.

Percentages are calculated based on valid data only.

IVDU indicates intravenous drug user; MnI mean intensity; MSM, men who have sex with men; NNRTI, nonnucleoside reverse transcriptase inhibitor.

TABLE 2

Univariate PH Models of Time to Virologic Suppression

Covariate	HR	*P*
Age, per y	1.04 (0.99 to 1.10)	0.13
White race	1.00 (0.29 to 3.43)	0.99
Men who have sex with men	0.61 (0.17 to 2.14)	0.44
AIDS-defining illness	0.32 (0.13 to 0.79)	0.01
Hepatitis B	0.87 (0.12 to 6.51)	0.89
Hepatitis C	1.60 (0.46 to 5.56)	0.46
Baseline viral load, $\log_{10}$ copies/mL	0.56 (0.39 to 0.80)	<0.01
Baseline CD4 count, per 100 cells/mm^3	1.16 (0.96 to 1.40)	0.13
Baseline CD4 count >200 cells/mm^3	3.47 (1.34 to 8.96)	0.01
Baseline CD4 count >100 cells/mm^3	5.00 (1.45 to 17.27)	0.01
No. years using ART	0.97 (0.84 to 1.12)	0.66
No. previous antiretroviral drugs	0.81 (0.67 to 0.98)	0.03
No. previous regimens	0.91 (0.79 to 1.05)	0.18
Present regimen includes PI	0.41 (0.16 to 1.09)	0.07
Present regimen includes NNRTI	0.91 (0.36 to 2.26)	0.83

NNRTI indicates nonnucleoside reverse transcriptase inhibitor.

TABLE 3

Univariate PH Models of Time to Virologic Suppression With Time-Dependent Covariates

Covariate	Unit (Cells/mm^3)	HR	*P*
CD4 count	100	1.17 (0.90 to 1.52)	0.23
CD8 count	100	1.06 (0.97 to 1.16)	0.16
$CD3^+$ mean	10	1.32 (0.82 to 2.12)	0.26
$CD38^+$ on CD4	10	0.73 (0.57 to 0.94)	0.01
HLA-DR on CD4	10	0.68 (0.37 to 1.25)	0.21
$CD38^+$ on CD8	10	0.80 (0.67 to 0.96)	0.02
HLA-DR on CD8	10	0.76 (0.49 to 1.16)	0.20
$CD28^+$ on CD4	10	1.17 (0.92 to 1.48)	0.20
$CD95^+$ on CD4	10	0.84 (0.61 to 1.16)	0.29
$CD95^+$ on CD8	10	0.84 (0.64 to 1.08)	0.18
$CD69^+$ on CD4	10	0.85 (0.56 to 1.30)	0.46
$CD69^+$ on CD8	10	0.68 (0.43 to 1.09)	0.11
CD38 MnI on CD4	10	0.65 (0.40 to 1.06)	0.08
CD38 MnI on CD8	10	0.68 (0.47 to 0.98)	0.04
CD95 MnI on CD4	1	0.81 (0.66 to 1.00)	0.05

MnI indicates mean intensity.

TABLE 4

Univariate PH Models of Time to Virologic Suppression, With Immune Marker Changes from Baseline to Week 2 as Covariates

Covariate	Unit (Cells/mm^3)	HR	*P*
Change in CD4 count	100	0.51 (0.19 to 1.40)	0.19
Change in CD8 count	100	0.87 (0.69 to 1.12)	0.28
Change in CD3$^+$ mean	10	2.30 (0.63 to 8.39)	0.21
Change in CD38$^+$ on CD4	10	0.83 (0.46 to 1.50)	0.55
Change in HLA-DR on CD4	10	0.33 (0.14 to 0.82)	0.02
Change in CD38$^+$ on CD8	10	1.00 (0.46 to 2.16)	1.00
Change in HLA-DR on CD8	10	0.98 (0.52 to 1.86)	0.95
Change in CD28$^+$ on CD4	10	0.91 (0.71 to 1.15)	0.42
Change in CD95$^+$ on CD4	10	0.73 (0.47 to 1.12)	0.15
Change in CD95$^+$ on CD8	10	0.95 (0.68 to 1.32)	0.76
Change in CD69$^+$ on CD4	10	0.75 (0.54 to 1.04)	0.08
Change in CD69$^+$ on CD8	10	0.70 (0.36 to 1.35)	0.29
Change in CD38 MnI on CD4	10	0.66 (0.46 to 0.95)	0.02
Change in CD38 MnI on CD8	10	1.35 (0.66 to 2.77)	0.41
Change in CD95 MnI on CD4	1	0.78 (0.63 to 0.97)	0.02