Predictive Value of Plasma HIV RNA Level on Rate of CD4 T-Cell Decline in Untreated HIV Infection

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D EPLETION OF CD4 CELLS IS A hallmark of progressive human immunodeficiency virus (HIV) disease[1] and a powerful predictor of the short-term risk of progression to AIDS.[2] Untreated HIV infection leads to AIDS in approximately half of untreated patients within 10 years,[3] but the rate at which AIDS occurs in each individual varies greatly. Both CD4 cell count and plasma HIV RNA level discriminate independently between rapid and slow progressors[4]; thus, the 9-year probability of AIDS ranges from 3.6% among patients with low plasma HIV RNA levels and high CD4 cell counts to 100% among patients at the opposite end of

Context Plasma human immunodeficiency virus (HIV) RNA level predicts HIV disease progression, but the extent to which it explains the variability in rate of CD4 cell depletion is poorly characterized.

Objective To estimate the proportion of variability in rate of CD4 cell loss predicted by presenting plasma HIV RNA levels in untreated HIV-infected persons.

Design Repeated-measures analyses of 2 multicenter cohorts, comprising observations beginning on May 12, 1984, and ending on August 26, 2004. Analyses were conducted between August 2004 and March 2006.

Setting Two cohorts of HIV-infected persons: patients followed up at 4 US teaching medical institutions or participating in either the Research in Access to Care for the Homeless Cohort (REACH) or the San Francisco Men's Health Study (SFMHS) cohorts and participants in the Multicenter AIDS Cohort Study (MACS) cohort.

Participants Antiretroviral treatment–naive, chronically HIV-infected persons (n=1289 and n=1512 for each of the 2 cohorts) untreated during the observation period (≥6 months) and with at least 1 HIV RNA level and 2 CD4 cell counts available. Approximately 35% were nonwhite, and 35% had risk factors other than male-to-male sexual contact.

Main Outcome Measures The extent to which presenting plasma HIV RNA level could explain the rate of model-derived yearly CD4 cell loss, as estimated by the coefficient of determination (R^2).

Results In both cohorts, higher presenting HIV RNA levels were associated with greater subsequent CD4 cell decline. In the study cohort, median model–estimated CD4 cell decrease among participants with HIV RNA levels of 500 or less, 501 to 2000, 2001 to 10 000, 10 001 to 40 000, and more than 40 000 copies/mL were 20, 39, 48, 56, and 78 cells/µL, respectively. Despite this trend across broad categories of HIV RNA levels, only a small proportion of CD4 cell loss variability (4%-6%) could be explained by presenting plasma HIV RNA level. Analyses using multiple HIV RNA measurements or restricting to participants with high HIV RNA levels improved this correlation minimally (R^2, 0.09), and measurement error was estimated to attenuate these associations only marginally (deattenuated R^2 in the 2 cohorts, 0.05 and 0.08, respectively).

Conclusions Presenting HIV RNA level predicts the rate of CD4 cell decline only minimally in untreated persons. Other factors, as yet undefined, likely drive CD4 cell losses in HIV infection. These findings have implications for treatment decisions in HIV infection and for understanding the pathogenesis of progressive immune deficiency.

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the spectrum.[5] In addition to their role as predictors of the clinical outcomes of HIV infection, CD4 cell count and plasma HIV RNA level are commonly used as markers of the success of highly active antiretroviral therapy (HAART). Large clinical trials using currently recommended antiretroviral combinations suggest that over three fourths of individuals starting HAART will achieve a plasma HIV RNA level below the limit of detection of clinically available assays, and most of them, but not all, will experience concomitant CD4 cell increases.[6-8]

Although the emphasis on plasma HIV RNA level as a determinant of therapeutic decisions has diminished in current treatment guidelines, presenting HIV RNA level continues to be recommended as one of the elements on which the timing of initiation of antiretroviral therapy is based.[9-11] The rationale for this practice has chiefly arisen from large cohort studies that have demonstrated an increasing risk of disease progression and AIDS among groups of patients defined by broad ranges of increasing plasma HIV RNA level, even after controlling for CD4 cell count.[5] To translate these observations into rules to guide the decision of initiating antiretroviral therapy in an individual patient, however, it is necessary to assess the extent to which the rate of CD4 cell decline can be predicted on the basis of an initial plasma HIV RNA value at the individual level. This question is intimately related, but not equivalent, to that addressed by previous studies[5] and has not been evaluated in a study population with a diversity of demographic characteristics representative of those seen in clinical practice.

Both cross-sectional analyses[5,12,13] and longitudinal studies[4,14,15] suggest that plasma HIV RNA level, categorized in broad strata, is inversely associated with rate of CD4 cell depletion, suggesting that HIV replication within CD4 cells leads to their accelerated destruction and eventual depletion. Yet, observations on the proportion and phenotype of infected cells[16] suggest a much more complex scenario, in which preferential infection of HIV-reactive cells,[17] widespread immune activation,[18] and selective inhibition of HIV-specific immune responses[19] each play a role in the pathogenesis of HIV-related immune deficiency. This more complex model of HIV pathogenesis is consistent with clinical experience and published reports[20] showing that patients with low HIV RNA levels can experience rapid and profound CD4 cell losses, while others with much greater levels of viremia maintain stable CD4 cell counts.

We sought to estimate the extent to which presenting plasma HIV RNA levels can account for interindividual variability in CD4 cell depletion rate among chronically HIV-infected individuals in the absence of antiretroviral therapy by providing a quantitative estimate of the coefficient of determination between plasma HIV RNA level and CD4 cell loss rate in a broad population of patients, including larger proportions of women and ethnic minorities. We report that plasma HIV RNA level can account for only a small proportion of the variability in rate of CD4 cell loss in chronic, untreated HIV infection.

METHODS
Study Population

The study population (henceforth referred to as the "study cohort") included individuals from 1 of 3 sources: (1) patients in a subset of participating sites (Case Western Reserve University, Cleveland, Ohio [n=172]; University of Washington, Seattle [n=141]; University of California–San Diego [n=445]; and Harvard University, Boston, Mass [n=133]) in the Centers for AIDS Research Network of Integrated Clinical Systems (CNICS), a collaborative database of real-time clinical care and laboratory data that encompasses 7 centers of excellence in HIV care throughout the United States; (2) participants in the San Francisco Men's Health Study (SFMHS)[21] [n=312]; and (3) participants in the Research in Access to Care for the Homeless Cohort (REACH)[22] [n=89]. Observations spanned from May 12, 1984, to August 26, 2004; analyses were conducted between August 2004 and March 2006.

Participants from each of the listed teaching institutions were consecutive patients followed up at each institution's HIV clinic, all of whom are routinely offered the opportunity to participate in observational research involving their deidentified clinical, demographic, and laboratory data. At each institution, the enrollment rate is greater than 95% of all patients receiving care at the clinic. Three institutions currently participating in CNICS did not contribute data to this analysis because they either could not provide their complete data set at the time of this analysis or, in 1 case, joined the CNICS consortium after the cohort for this study had been assembled.

Both REACH and SFMHS are based on probabilistic sampling of their respective target populations; the design of both surveys has been described.[21,22] Briefly, SFMHS surveyed single men 25 to 54 years of age in San Francisco,[21] whereas REACH surveyed homeless and marginally housed adults throughout the city and county of San Francisco,[22] and both relied on study-mandated follow-up visits at regularly spaced intervals. Enrollment rates in SFMHS and REACH were approximately 64% and 80%, respectively. Nonwhites accounted for 34% of the clinic-based cohort, 10% of SFMHS, and 56% of REACH. In the clinic-based cohort and in REACH, 16% and 13.5% were women and 57% and 20% had risk factors other than male-to-male sexual contact, respectively. All participants in SFMHS were men who had sex with men.

Seroconverters in SFMHS (n=49, those with a negative HIV antibody test result on accrual who acquired HIV while under observation) were excluded from the analysis. Individuals enrolled from other sources did not have uniformly available data on seroconversion date, but were presumed to have chronic HIV infection.

To exclude the possibility that some of these participants may have been enrolled during the acute infection pe-

riod, however, we computed the observed change in plasma HIV RNA level during the first 6 months after the baseline observation for participants who had multiple HIV RNA measurements available and targeted for detailed review those persons found to have experienced a decrease in HIV RNA of 1 log or greater during that period. Since all participants were unexposed to antiretroviral therapy, a significant decrease in plasma HIV RNA level during the early observation period was considered to be a possible indicator of recent infection because HIV RNA levels are expected to decrease spontaneously toward the viral "set point" during that period. Based on review of source medical records, 3 participants were considered to represent probable acute or recent infections and were excluded from further analysis.

For all sources of participants, eligible study participants included those with at least 6 months of follow-up who had at least 1 plasma HIV RNA and 2 CD4 cell count measurements, the first concurrent with the initial plasma HIV RNA measurement (designated as the baseline observation). While the requirement of at least 6 months of follow-up resulted in exclusion of a large number of otherwise potentially eligible participants, estimation of CD4 cell decline rate in those with less than 6 months of available observations was imprecise and lacked comparability to the validation cohort (see below), which included observations 6 months apart.

All patients provided institutional review board–approved written informed consent to have their clinical information used for research purposes, including at a minimum the type of information to be collected, alternatives to participation, and possible uses of the compiled data. Race/ethnicity data were recorded routinely for program evaluation and as possible explanatory variables in future analyses and were derived from each person's self-identification.

To validate the findings in a population followed using uniform procedures, a parallel analysis was carried out using the Multicenter AIDS Cohort Study (MACS) public data set (US Department of Commerce, release P12, December 16, 2003; available from the National Technical Information Service [www.ntis.gov]), using an identical analytical approach. MACS is a prospective study of the natural and treated histories of HIV-1 infection in homosexual and bisexual men, including sites in Baltimore, Chicago, Pittsburgh, and Los Angeles. A total of 6973 men have been enrolled over 3 separate recruitment periods beginning in 1984, 1987, and 2001. Median age is 34 years.[23,24]

The baseline observation for the analyses on the MACS cohort was required to be as follows: (1) either the third or fourth visit available in the data set, whichever included both a CD4 cell count and a plasma HIV RNA measurement and (2) at least 1 visit after the first time the participant was known to be HIV positive. Since visits in MACS occurred every 6 months, this ensured that the initial observation for participants who became infected after enrollment into the cohort occurred at least 6 months after initial infection. For both cohorts, CD4 cell count measurements up to the time of the first documented exposure to antiretroviral therapy or the last follow-up visit were included in the analyses.

Measurements

Plasma HIV-1 RNA levels were measured using either the Amplicor HIV-1 Monitor assay (Roche Diagnostics, Indianapolis, Ind) or the branched-DNA assay (Chiron Corp, Emeryville, Calif), and all values were standardized to their branched DNA equivalents.[5] The CD4 cell counts were measured by flow cytometry using standardized methods; specific instruments varied according to the institution.

Statistical Analysis

Analyses were conducted primarily by 5 of the authors (R.J.B., V.K.C., W.M., B.R., and A.K.S.; S.Y. contributed in developing an initial algorithm to calculate average observed CD4 cell slopes for an early analysis in this project). All available CD4 cell count and plasma HIV RNA data were used. Missing demographic data were not imputed. Descriptive and other basic statistics were computed using Intercooled Stata, version 8.0 (Stata Corp, College Station, Tex). To assess the association between plasma HIV RNA concentration and CD4 cell change, we used random-effects linear models (PROC MIXED, SAS software v.8.2; SAS Institute Inc, Cary, NC) assuming an unstructured correlation structure.

We used $\log_{10}$-transformed values of plasma HIV RNA in all analyses. First, we created 5 strata by dividing the study population into quartiles of presenting plasma HIV RNA level and subdividing the lowest quartile into 2 groups (above or below 500 copies/mL) to account for the likely unique nature of participants who, in the absence of antiretroviral therapy, maintain a very low level of HIV RNA (ie, <500 copies/mL). We then fitted separate models to produce estimates and confidence intervals (CIs) of mean CD4 cell decline for each HIV RNA stratum. No implausible CD4 cell or HIV RNA values were observed, and therefore no attempt was made to manipulate outlier values in any way.

To determine the extent to which interindividual variability in CD4 cell change was explained by presenting HIV RNA measurement, we fitted a model using all study participants and computed model–estimated CD4 cell change per individual using deviations from the average CD4 cell decline for the study population. We then performed linear regression of model-estimated CD4 cell count slope on presenting HIV RNA level and obtained the coefficient of determination (R^2). We calculated the precision of the R^2 estimates by computing 95% CIs using Fisher Z-transformation for the corresponding (unsquared) correlation.

We estimated that a sample size of 1250 individuals would be needed for the analysis to obtain a 95% CI for R^2 values of ±0.03 or less. For all analyses, a *P* value of ≤.05 was considered significant. To determine if repeated measurements of HIV RNA (as ob-

tained for clinical follow-up) were better than a single value at explaining interindividual variability in CD4 T-cell change, we used a similar approach to generate model-based estimates of HIV RNA change per individual and regressed these estimates on model-derived individual rates of CD4 cell loss. To assess whether multiple measurements of plasma HIV RNA could explain a greater proportion of variability in CD4 cell decline rate, we computed the mean and median HIV RNA level among participants with multiple measurements available and substituted these values for the presenting HIV RNA level in the analyses described above.

To explore the possible effect of CD4 cell count and HIV RNA measurement error on the results, we first derived an attenuation factor[25,26] for HIV RNA based on the observed variability in $\log_{10}$ presenting HIV RNA relative to published estimates of measurement error.[27,28] Correlations tend to be attenuated toward zero (and corresponding R^2 values are attenuated downward) by measurement error; the attenuation factor quantifies the magnitude of this effect of measurement error on the R^2 values. Similarly, we derived an attenuation factor due to measurement error in CD4 cell count from the variability in estimated CD4 cell count slopes relative to the average standard error of these slopes as estimated from participant-specific linear regression models. We then generated deattenuated co-

Table. Baseline Characteristics of the Study Cohort and Estimates of CD4 Cell Loss Rate, by Stratum of Presenting Plasma HIV RNA Level*

	Presenting Plasma HIV RNA Level, Copies/mL					
	≤500	501-2000	2001-10 000	10 001-40 000	>40 000	Overall
Participants/observations, No.	176/1691	157/1455	308/2682	318/2603	330/2273	1289/10 704
Age, y†	36 (31-42)	36 (30-41)	34 (30-41)	36 (30-41)	36 (31-42)	36 (31-41)
Male sex, No. (%)‡	146 (83.4)	129 (82.2)	261 (84.7)	291 (91.5)	307 (93.3)	1134 (88.1)
Enrolled in or after 1995, No. (%)	154 (87.5)	126 (80.3)	245 (79.5)	215 (67.6)	237 (71.8)	977 (75.8)
Risk factor, No. (%) Male homosexual intercourse	89 (50.6)	99 (63.1)	198 (64.3)	224 (70.4)	227 (68.8)	837 (64.9)
Intravenous drug use	10 (5.7)	8 (5.1)	29 (9.4)	22 (6.9)	21 (6.4)	90 (7.0)
Male homosexual intercourse and intravenous drug use	21 (11.9)	10 (6.4)	20 (6.5)	21 (6.6)	31 (9.4)	103 (8.0)
Heterosexual intercourse	26 (14.8)	27 (17.2)	38 (12.3)	33 (10.4)	30 (9.1)	154 (11.9)
Hemophilia	1 (0.6)	0	1 (0.3)	1 (0.3)	0	3 (0.2)
Other/unknown	29 (16.5)	13 (8.3)	22 (7.1)	17 (5.3)	21 (6.4)	102 (7.9)
Race/ethnicity, No. (%) White	95 (54.0)	96 (61.1)	195 (63.3)	226 (71.1)	226 (68.5)	838 (65.0)
African American	48 (27.3)	35 (22.3)	77 (25.0)	47 (14.8)	47 (14.2)	254 (19.7)
Asian American/ Pacific Islander	0	1 (0.6)	4 (1.3)	1 (0.3)	3 (0.9)	9 (0.7)
Native American	0	0	0	0	1 (0.3)	1 (0.1)
Hispanic	13 (7.4)	16 (10.2)	22 (7.1)	27 (8.5)	42 (12.7)	120 (9.3)
Other/unknown	20 (11.4)	9 (5.7)	10 (3.2)	17 (5.3)	11 (3.3)	67 (5.2)
Previous history of AIDS, No, (%)§	3 (2.0)	4 (2.7)	20 (6.9)	18 (6.1)	41 (13.1)	86 (7.2)
No. of available CD4 cell count measurements per individual	7 (4-14)	8 (5-13)	7 (5-12)	7 (4-12)	5 (3-9)	7 (4-11)
Time of follow-up, y	3 (1.4-4.9)	2.7 (1.4-5.6)	2.5 (1.3-4.6)	2.7 (1.1-5.2)	1.5 (0.9-3.2)	2.3 (1.1-4.6)
Presenting plasma HIV RNA, copies/mL	84 (68-251)	1151 (774-1568)	5000 (3375-6874)	20 652 (14 929-28 121)	89 181 (52 970-169 478)	10 210 (1839-42 452)
Presenting CD4 cell count, cells/μL	654 (474-864)	587 (450-775)	544 (402-713)	507 (364-705)	433 (274-590)	525 (371-712)
Random-effects model estimate of absolute CD4 cell loss, cells/μL per y (95% CI)‖	20.2 (9-31.3)	39.3 (29.2-49.4)	47.7 (39.7-55.6)	55.9 (47.3-64.5)	77.7 (68.2-87.3)	50.5 (46.2-54.8)

Abbreviations: CI, confidence interval; HIV, human immunodeficiency virus.
*Data are presented as median (interquartile range), unless otherwise specified.
†Based on 1278 participants. No data available on 11 (0.8%) of the overall study population.
‡Based on 1287 participants. No data available on 2 (0.15%) of the overall study population.
§Based on 1199 participants. No data available on 90 (7%) of the overall study population.
‖As estimated per stratum.

Figure 1. Estimated Annual CD4 Cell Count Loss in Untreated HIV-Infected Patients According to Presenting Plasma HIV RNA Level
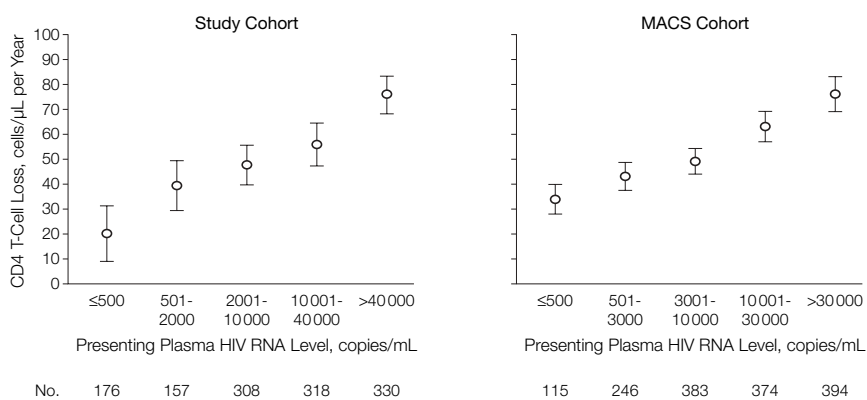
Data are presented as estimates of CD4 cell loss from random-effects models for each stratum with their associated 95% confidence intervals. HIV indicates human immunodeficiency virus; MACS, Multicenter AIDS Cohort Study.

efficients of determination, taking into account both of these attenuation factors, as previously described.[26] Because the analyses used all available data for a given individual, and because the timing of CD4 cell and HIV RNA testing was dictated by clinical indication (in the clinic-based cohorts), instead of a predetermined schedule, there are no missing data in the classic sense in the CD4 cell and HIV RNA measurements. Similarly, in the interval cohorts (REACH and SFMHS), all available patient visits were used.

RESULTS

We included 1289 patients, providing 10 704 observations. Mean follow-up by source was as follows: Case Western Reserve University (n=172), 2.2 years; University of Washington (n=141), 2.4 years; University of California–San Diego (n=444), 2.1 years; Harvard University (n=131), 2.3 years; SFMHS (n=312), 5.1 years; and REACH (n=89), 3.7 years.

Another 712 individuals were not included because of insufficient time of follow-up. Compared with those included, those who were excluded were more often male (65.6% vs 58.2%, P=.02), but their age was similar (median, 36 vs 38 years, P=.73). Excluded individuals also had lower initial CD4 cell counts (median, 275 cells/μL vs 525 cells/μL, $P<$.001) and higher plasma HIV RNA levels (median, 38 097 copies/mL vs 10 210 copies/mL, $P<$.001), reflecting earlier initiation of antiretroviral therapy among those with more advanced disease stages. Indeed, approximately 60% of the excluded persons initiated antiretroviral therapy within 6 months of their initial observation. By comparison, only 10% of the included participants received antiretroviral therapy within the first year after the initial observation.

The **TABLE** summarizes characteristics of the study cohort and model estimates of CD4 cell slopes for each of the 5 HIV RNA strata. Presenting HIV RNA level and CD4 cell count were within the expected range for a cohort of these characteristics and remarkably similar to those reported in other cohorts.[5] Average CD4 cell count decline was consistently greater with higher presenting HIV RNA levels, ranging from 20 cells per year in the less than 500 copies/mL stratum (95% CI, 9-31 cells per year; n=176) to 78 cells per year (95% CI, 68-87 cells per year; n=330) in the greater than 40 000 copies/mL stratum.

Fitting a regression of estimated yearly CD4 cell loss per person (n=1289) on their presenting plasma HIV RNA level yielded a coefficient of determination (R^2) of 0.04 (95% CI, 0.02-0.06), indicating that most of the variability in CD4 cell slope could not be accounted for by presenting plasma HIV RNA levels. Coefficients of determination were also small when analyzed within each of the HIV RNA strata (R^2 within each of the 5 HIV RNA strata, <0.01) or among those with very high levels of presenting HIV RNA (R^2=0.002 in participants with HIV RNA either >100 000 or >300 000 copies/mL, n=145 and n=42, respectively). Stratified analyses by sex and risk factor yielded similar conclusions (R^2 for men and women, 0.04 and 0.002, respectively; R^2 within each of the risk categories, <0.01).

To exclude the possibility of bias from the exclusion of a large group of potential participants due to short follow-up time, we analyzed individuals with initial CD4 cell count of 275 cells/μL or less (n=175, 13%), the median value among the excluded group. In this subgroup, the R^2 was 0.02, thus suggesting that the conclusions apply equally well to a subset of participants with similar CD4 cell counts and HIV RNA levels to those excluded due to short follow-up. Conclusions also did not change in stratified analyses according to whether the source of participants was an interval cohort such as SFMHS and REACH or a clinic-based cohort (R^2 for the clinic-based and cohort subgroups, 0.02 and 0.07, respectively; n=888 and n=401, respectively).

To address whether these conclusions held using a larger interval cohort in which all participants were followed up according to a uniform schema, we replicated the analyses above in the MACS (n=1512, contributing 12 529 observations) and found a similar monotonic increase in CD4 cell count decline with increasing categories of presenting plasma HIV RNA. The results are shown in **FIGURE 1**. The coefficient of determination of CD4 cell decline rate on presenting plasma HIV RNA measurement was 0.06 (95% CI, 0.04-0.08), closely resembling the findings in our primary data set. The low predictive value of presenting plasma HIV RNA level on subsequent CD4 cell decline reflects the large variability in

CD4 cell slopes within each HIV RNA stratum, indicating that individuals with similar presenting plasma HIV RNA levels can have grossly different rates of CD4 cell decline. This is illustrated in **FIGURE 2**, which depicts the considerable overlap of individual estimated CD4 cell decline rates across presenting HIV RNA strata in the study cohort.

We next considered the effect of measurement error in both CD4 cell counts and plasma HIV RNA levels by estimating the possible attenuation of the coefficient of determination due to this error[25,26] and found that assay variability could be expected to produce an attenuation of the R^2 of only 8% for plasma HIV RNA level and 16% for CD4 cell slope. Based on these estimates, we calculated the largest plausible coefficient of determination to be 0.052 (when using the coefficient obtained from our data set) to 0.078 (when using the MACS data set).

We were also interested in the possibility that an estimate of HIV RNA change over time could represent more accurately the overall trajectory of HIV viremia (for example, by reflecting large changes such as those seen in advanced HIV disease), thus enhancing the predictive value of HIV RNA levels on rate of CD4 cell decline. To examine this possibility, we estimated per-individual HIV RNA slopes, as described for CD4 cell slopes, and used these estimates as the explanatory variable in a linear regression model with estimated CD4 cell decline as the dependent variable among the 974 (75.5%) persons who had multiple available measurements of plasma HIV RNA. Despite a significant inverse correlation, the coefficient of determination in this analysis was only 0.05. The change in longitudinally measured plasma HIV RNA levels was as expected for a cohort of chronically infected individuals: median annual change was 0.06 $\log_{10}$ copies/mL (interquartile range: −0.01 to 0.14), and 841 (86%) of the observations were within −0.25 and 0.25 $\log_{10}$ copies/mL, suggesting a stable plasma HIV RNA level.

We then assessed whether a summary measure of several HIV RNA lev-els could enhance the predictability of CD4 cell decline rate. We computed the median of all available HIV RNA measurements during the entire observation period, the first year of observation or the first 6 months of observation, and used it as the explanatory variable of CD4 cell decline rate. Results are shown in **FIGURE 3**; whereas increasing the number of HIV RNA measurements used to predict the rate of CD4 cell decline increased the coefficient of determination, even using all available HIV RNA values resulted in an R^2 of 0.09.

Finally, we focused on the subgroup of patients whose initial CD4 cell count was between 200 and 350 cells/μL, which reflects the spectrum of patients that poses the greatest challenge to the clinician in deciding the timing of initiation of antiretroviral therapy. Among 175 such patients (median follow-up, 1.9 years; median HIV RNA level, 4.2 $\log_{10}$ copies/mL), the R^2 was 0.09, only slightly higher than in the overall study cohort.

COMMENT

Our findings confirm previous observations[5] that the magnitude of HIV viremia, as defined by broad categories of presenting HIV RNA level, is associated with the rate of CD4 cell loss and extend this observation to patient populations comprising both men and women. Despite this association, however, only a small proportion of the interindividual variability in the rate of CD4 cell decline can be explained by plasma HIV RNA level, even after accounting for the effect of measurement error. Likewise, the intrinsic irregularity of clinical data is unlikely to account for the findings, since the results were similar when the analysis was replicated in a data set derived from an interval cohort study.

Exploratory analyses by sex, risk factor, and presenting HIV RNA stratum did not reveal any characteristic resulting in a greater predictive ability of presenting HIV RNA level on rate of CD4 cell loss, although these subgroup analyses may have resulted in lower coefficients of determination partly due

Figure 2. Histograms of CD4 Cell Count Change by Categories of Presenting Plasma HIV RNA Level Among All Patients in the Study Cohort
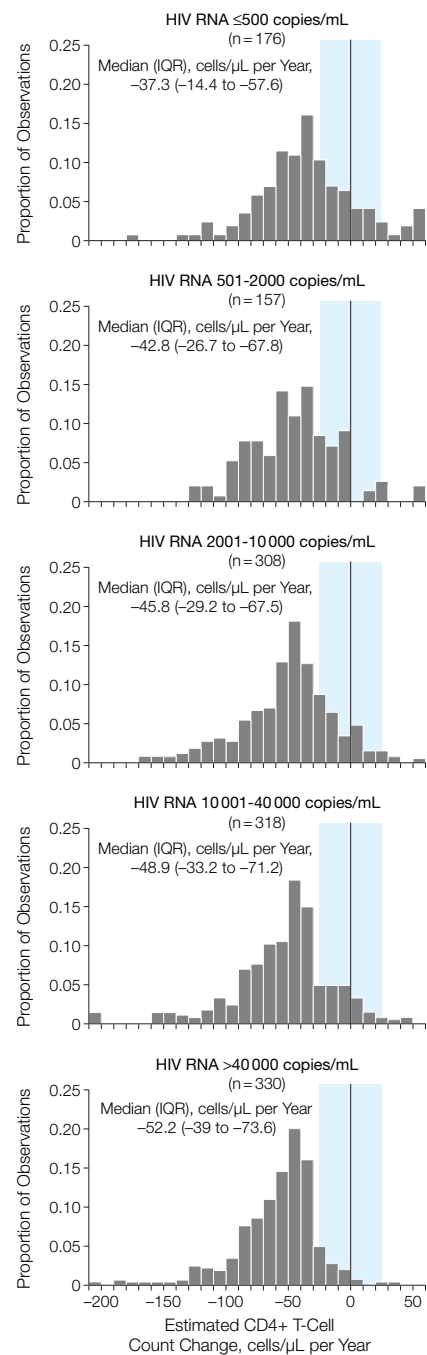

Data are model-based individual estimates of yearly CD4 cell decline obtained from random-effects modeling. The blue tinted region represents the −25 to 25 cells/μL per year range, an arbitrary reference to emphasize the slight increase in CD4 cell decline with increasing plasma HIV RNA levels. Note the extensive overlap across these frequency distributions. HIV indicates human immunodeficiency virus; IQR, interquartile range.

to the reduced variability in presenting plasma HIV RNA level within each subgroup. Using the median of multiple HIV RNA measurements instead of a single presenting value increased the coefficient of determination slightly, but only by using all the available HIV RNA measurements over a several-year period, an approach that would have no translation into clinical practice.

This study did not address the predictive value of HIV RNA level at the beginning of the rapidly progressive phase of HIV infection, which typically heralds a subsequent decline in CD4 cell count.[29] Whereas it is possible to speculate that the coefficient of determination may have been higher had the HIV RNA level used in these analyses been the one preceding the advanced phase of HIV disease, the clini-

cal application would be limited, as it would require advance knowledge of the individual patient's "inflection point," the time at which HIV RNA begins to increase steeply, to select the plasma HIV RNA level on which to base a prediction of CD4 cell decline rate. Nonetheless, we attempted to capture these features of plasma HIV RNA trajectory by estimating the slope of HIV RNA over time among participants with

Figure 3. Use of Multiple Measurements of HIV RNA Improves the Predictive Ability of HIV RNA Level on CD4 Cell Decline Rate Only Minimally
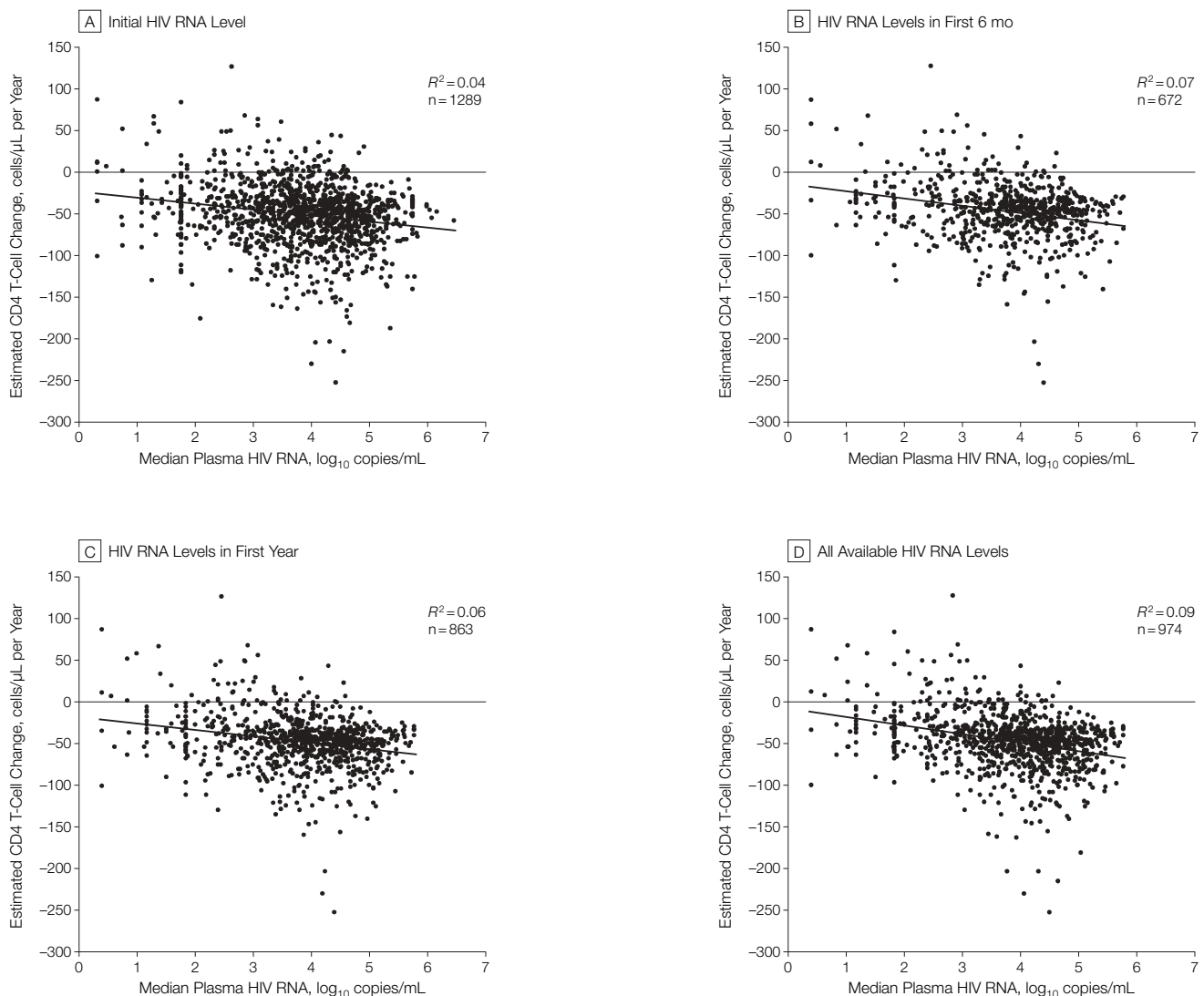
Scatterplots of estimated CD4 cell count change per year against either presenting HIV RNA level (1 measurement per patient, A) or median HIV RNA level over the first 6 months (B), the first year (C), or the whole observation period (D) among those with more than 1 available measurement. The fit line and R^2 are derived from linear regression. Although the coefficient of determination increases slightly, even using the maximum number of available measurements improves the predictive ability of plasma HIV RNA level only minimally. HIV indicates human immunodeficiency virus.

multiple measurements available and found that even this approach resulted in a very small coefficient of determination; admittedly, this could reflect dilution of the subset of individuals whose inflection point occurred during the follow-up period amid the majority in whom this did not occur.

These findings represent a major departure from the notion that plasma HIV RNA level is a reliable predictor of rate of CD4 cell loss in HIV infection and challenge the concept that the magnitude of viral replication (at least as reflected by plasma levels) is the main determinant of the speed of CD4 cell loss at the individual level. The clinical implications are that in the majority of cases, an individual patient's plasma HIV RNA level at the time of presentation for clinical care cannot predict, to a significant extent, the rate of CD4 cell decline that he or she will experience over the subsequent years and is therefore of limited clinical value in shaping the decision to initiate antiretroviral therapy. This is despite the fact that a group of individuals with an approximately similar level of plasma viremia will, on average, tend to lose CD4 cells at a faster rate than another group with a lower level of viremia, a previously reported finding[5] that stands uncontested by our results.

Data from randomized clinical trials[30-32] and cohort studies[33] leave little doubt that HIV-infected individuals with symptomatic disease or very low CD4 cell counts benefit from immediate antiretroviral therapy, but there exists limited evidence to guide the clinician in deciding the timing of treatment initiation in asymptomatic persons with moderately high CD4 cell counts. In this setting, current guidelines[10,11,34] suggest consideration of the HIV RNA level as an element in the decision, based on the observation of an increased risk of disease progression among persons with high (>100 000 copies/mL) plasma HIV RNA levels.[5,15,33] All the major guidelines, however, consider this approach optional, highlighting the uncertainty surrounding this issue. Our findings contribute information to clarify this critical area of HIV clinical practice by

showing that in the individual treatment-naive patient, presenting plasma HIV RNA levels only explain a small portion of the variability in CD4 cell decline rate. Moreover, this observation holds true even among those patients with high initial plasma HIV RNA levels and among the subgroup of individuals with presenting CD4 cell counts in the 200- to 350-cells/μL range, suggesting that the role of plasma HIV RNA level in deciding the timing of antiretroviral treatment among these individuals is also limited.

Much of HIV pathogenesis is thought to occur in the lymphoid tissues, and plasma HIV RNA level may reflect tissue viral burden inconsistently.[35,36] Nevertheless, the proportion of variability in CD4 cell loss rate that can be accounted for by the level of viremia in this analysis is so low that much of this variability must be related to other unmeasured predictors, even if the correlation between tissue and plasma HIV replication is not perfect.

What factors may account for the residual variability in CD4 cell decay rate? HIV infection is associated with heightened T-cell activation and cellular turnover,[18,37] and expression of immune activation markers is associated with both clinical disease progression[38] and rate of CD4 cell depletion.[39] Cellular immune activation is commonly measured through enumeration of the proportion of cells that express markers such as CD38 and HLA-DR by flow cytometry, although this test is not routinely performed in clinical practice. Proposed mechanisms of immune activation–mediated CD4 cell destruction include programmed cell death, bystander cell killing, and accelerated cellular senescence.[40-42] Although the level of immune activation is in turn associated with the level of viremia,[43] this association is far from perfect. Sooty mangabeys[44] and African green monkeys[45] infected with simian immunodeficiency virus (SIV), a naturally occurring lentivirus closely related to HIV, show near-normal T-cell turnover and a lack of disease progression despite sustained high-level viremia. Conversely, transfer of the same SIV iso-

lates into rhesus macaques results in similar levels of viral replication, but leads to a picture strongly reminiscent of human AIDS, with massive immune activation, CD4 cell depletion, and death.[46]

In humans, the predictive value of immune activation level on HIV disease course, independent of plasma HIV RNA levels, can be demonstrated even when measured during early infection[47] or before actual seroconversion.[48] Thus, immune activation may be a major determinant of T-cell turnover and CD4 cell depletion in chronic HIV infection[49] both in human and animal hosts. Our results provide further support for additional studies exploring the relative contribution of immune activation to the pathogenesis of immune deterioration in treatment-naive, HIV-infected persons.

CONCLUSIONS

The results of our study challenge the concept that CD4 cell depletion in chronic HIV infection is mostly attributable to the direct effects of HIV replication. Future efforts to delineate the relative contribution of other mechanisms will be crucial to the understanding of HIV immunopathogenesis and to the ability to attenuate it.

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