Strong Human Endogenous Retrovirus-Specific T Cell Responses Are Associated with Control of HIV-1 in Chronic Infection

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Eight percent of the human genome is composed of human endogenous retroviruses (HERVs), which are thought to be inactive remnants of ancient infections. Previously, we showed that individuals with early HIV-1 infection have stronger anti-HERV T cell responses than uninfected controls. In this study, we investigated whether these responses persist in chronic HIV-1 infection and whether they have a role in the control of HIV-1. Peripheral blood mononuclear cells (PBMCs) from 88 subjects diagnosed with HIV-1 infection for at least 1 year (median duration of diagnosis, 13 years) were tested for responses against HERV peptides in gamma interferon (IFN-γ) enzyme immunospot (ELISPOT) assays. Individuals who control HIV-1 viremia without highly active antiretroviral therapy (HAART) had stronger and broader HERV-specific T cell responses than HAART-suppressed patients, virologic noncontrollers, immunologic progressors, and uninfected controls ($P < 0.05$ for each pairwise comparison). In addition, the magnitude of the anti-HERV T cell response was inversely correlated with HIV-1 viral load ($r^2 = 0.197, P = 0.0002$) and associated with higher CD4+ T cell counts ($r^2 = 0.072, P = 0.027$) in untreated patients. Flow cytometric analyses of an HLA-B51-restricted CD8+ HERV response in one HIV-1-infected individual revealed a less activated and more differentiated phenotype than that stimulated by a homologous HIV-1 peptide. HLA-B51 tetramer dual staining within this individual confirmed two different T cell populations corresponding to these HERV and HIV-1 epitopes, ruling out cross-reactivity. These findings suggest a possible role for anti-HERV immunity in the control of chronic HIV-1 infection and provide support for a larger effort to design an HIV-1 vaccine that targets conserved antigens such as HERV.

Much of the recent effort in identifying immune correlates of protection in HIV-1 has focused on defining the qualities of a successful T cell response in individuals who maintain low or undetectable viral loads in the absence of treatment, i.e., controllers (1, 6, 9, 27). As a result of these investigations, there have been major advances in the understanding of the immunobiology of HIV-1 infection (30). However, the quest for a fully effective vaccine is still ongoing, and the designs of many vaccine candidates have only varied from each other slightly (31). We have studied an area outside conventional HIV-1 vaccine candidates and explored the potential for a human endogenous retrovirus (HERV)-based HIV-1 vaccine.

Transposable elements make up 45% of the human genome (19). Human endogenous retroelements (which rely on an RNA intermediate before integration into the genome) can be classified into the non-long terminal repeat (LTR) class and the LTR class, represented by endogenous retroviruses (ERV). Many ERV entered the primate germ line as infectious retroviruses at several time points during human evolution (4). Out of the six HERV superfamilies, HERV-K (HML-2) is considered to be the youngest and most transcriptionally active (18). A report in 2008 described the visualization of HERV-K-like viral particles in the plasma of lymphoma patients (8). A clinical study demonstrated the potential utilization of another HERV family in a novel cancer immunotherapy product (33). In a group of patients with metastatic renal cell carcinoma (RCC) who experienced tumor regression after allogeneic hematopoietic stem cell transplant, RCC-reactive donor-derived CD8+ T cells directed against a 10-mer HERV-E peptide were identified. This antigen was expressed in RCC cells but not healthy tissues, suggesting that it could be targeted by a new tumor peptide vaccine or by adoptively infused peptide-specific cytotoxic T lymphocytes (CTLs) (33). Host cells have developed mechanisms to prevent lentiviral replication, as well as to restrict the movement of retroelements in order to maintain genomic stability. Esnault et al. (11) and others (24) reported that the host protein APOBEC3
restricts endogenous retroelements. The HIV-1 Vif protein has been shown to disable APOBEC3G (13, 32), which could lead to the activation of HERV. In support of this hypothesis, our group and others have demonstrated that HERV-K transcripts can be detected in the plasma of HIV-1-infected individuals, suggesting that HIV-1 infection can alter the biology of HERV-K by enhancing its gene expression (7, 12).

We also previously reported that adults with early HIV-1 infection have detectable ex vivo CD8+ T cell responses to HERV antigens and that these responses are absent in healthy subjects (12). The magnitude of the anti-HERV response is inversely correlated with HIV-1 plasma viral load, suggesting a potential role for these responses in the control of HIV-1 (12).

The current study examined the immunological consequences of HERV antigen production and presentation in individuals with chronic HIV-1. We hypothesized that subjects who are able to control HIV-1 in the absence of highly active antiretroviral therapy (HAART) have stronger and more frequent HERV-specific T cell responses than those who are unable to control HIV-1 without HAART. We also hypothesized that responses would be higher in individuals controlling the virus as an apparent consequence of the host response (controllers) than in individuals suppressing the virus as a consequence of antiretroviral treatment. In order to elucidate a possible mechanism of viral control in these individuals, we compared the functionality and phenotype of HERV-, HIV-1-, and cytomegalovirus (CMV)-specific responses. The work outlined in this study has the potential to lead to a vaccination strategy utilizing HERV-specific T cells with the capacity to kill HIV-1-infected cells expressing HERV epitopes. It is envisioned that such a novel vaccine would be part of a combination vaccination approach that includes stimulation of immunity to HIV-1 antigens as well as to HERV.

MATERIALS AND METHODS

Study populations. Samples of peripheral blood mononuclear cells (PBMCs) were selected from participants in two different San Francisco-based HIV-1-infected cohorts, Options (15) and SCOPE (17), as well as from an HIV-1-infected cohort at the University of Toronto. Samples from HIV-1-negative controls were obtained from 22 individuals who donated blood to the Stanford blood bank. The study was approved by the local institutional review boards (University of California, San Francisco [UCSF] Committee on Human Research and University of Toronto Institutional Review Board), and individuals gave written informed consent. Studies were performed on cryopreserved PBMCs.

PBMC samples were obtained from the following categories of HIV-1-infected individuals: 30 untreated virologic controllers (which included 10 viremic controllers with HIV-1 viral loads between 50 and 2,000 copies/ml and 20 elite controllers with viral loads of <50 to 75 HIV-1 copies/ml), 20 highly active antiretroviral therapy (HAART)-suppressed patients (<50 to 75 HIV-1 copies/ml), and 18 untreated virologic noncontrollers (>2,000 copies/ml). All had a CD4+ T cell count of >250 cells/mm3. A fourth group of untreated HIV-1-infected patients was also tested. These 20 individuals were defined as immunologic progressors, with an HIV-1 viral load of >2,000 copies/ml and CD4+ T cell count of <250 cells/mm3. All 88 patients had been diagnosed with HIV-1 at least 1 year prior to inclusion in this study. There was no significant difference in the median ages of the five groups (including HIV-1-infected and uninfected subjects) or duration of diagnosis among the HIV-1-infected groups (P > 0.05 for all pairwise comparisons). Table 1 describes baseline subject characteristics.

PBMC preparation and storage. PBMCs were isolated by standard Ficoll-Hypaque density gradient centrifugation on fresh blood samples and immediately cryopreserved in fetal calf serum (HyClone, Logan, UT) containing 10% dimethyl sulfoxide (Sigma Aldrich, St. Louis, MO) in liquid nitrogen. The cryopreserved cells were stored in liquid nitrogen until they were used.

Peptides and tetramers. A commercially manufactured set of 28 9- to 13-mer individual HERV peptide sets with >95% purity (Genscript, Piscataway, NJ) was used to screen individuals for anti-HERV CD8+ T cell responses (see Table S1 in the supplemental material). The HERV peptides were designed using HLA binding prediction software (with a predicted combined binding score of >0.75 according to NetCTL, version 1.2) (20, 29). They included peptides from several HERV families, including HERV-K, HERV-H, and HERV-L. A subset of these peptides was used in our previous publication (12) and is marked with an asterisk in Table S1. A set of overlapping 15-mer HERV-K Gag and Env peptides (JPT Peptide Technologies, Berlin, Germany) was also used to comprehensively map one elite controller’s (subject P1’s) responses. HIV-1 peptides selected from the Gag, Nef, Env, and Pol proteins (Sigma Aldrich and Invitrogen, Carlsbad, CA) (see Table S2) and a CMV pp65 pool of overlapping 15-mer peptides (JPT Peptide Technologies) were also tested. All HERV peptides were used at a concentration of 100 μg/ml for the initial enzyme-linked immunospot (ELISPOT) screening assays and then titrated down to final concentrations of 1 μg/ml to 10 μg/ml for subsequent ELISPOT assays and flow cytometry experiments. HLA-B51 tetramers were synthesized and provided by the NIH Tetramer Core Facility (Atlanta, GA).

ELISPOT assays. The ELISPOT assay has been described previously (22). In brief, 96-well plates (Millipore, Billerica, MA) were coated with human monoclonal anti-interferon gamma (IFN-γ) immunoglobulin (Mabtech, Mariemont, OH). After plates were washed and blocked with 10% fetal calf serum, PBMCs were added at a concentration of 105 cells per well. Duplicate wells were prepared for each experimental condition. Spot totals for duplicate wells were averaged, and all spot numbers were normalized to numbers of IFN-γ spot-forming units (SFU) per million PBMCs (SPM). The spot values from medium control wells were subtracted, after which a positive response to a peptide was defined as >50 SPM and >2 times the medium control value. The total magni-

### Table 1. Characteristics of study subjects

<table>
<thead>
<tr>
<th>Participant category (n)</th>
<th>Median age (yr [IQR])</th>
<th>Gendera</th>
<th>Ethnicity and no. of subjectsb</th>
<th>Median duration of HIV-1 diagnosis (yr [IQR])</th>
<th>Median CD4+ T cell count (cells/mm³ [IQR])</th>
<th>Median HIV-1 viral load (copies/ml [IQR])</th>
</tr>
</thead>
<tbody>
<tr>
<td>Controller (30)</td>
<td>46.5 (40.5–52)</td>
<td>23 M</td>
<td>C, 13; L, 4; A, 1; AA, 10; NA, 2</td>
<td>13 (6–18)</td>
<td>794 (569–1,059)</td>
<td>50 (40–302)</td>
</tr>
<tr>
<td>HAART-suppressed (20)</td>
<td>48 (39.5–51)</td>
<td>17 M</td>
<td>C, 11; L, 5; A, 1; AA, 3</td>
<td>13 (9.8–17)</td>
<td>686 (568–1,069)</td>
<td>75 (50–75)</td>
</tr>
<tr>
<td>Virologic noncontroller (18)</td>
<td>49.5 (38.5–51)</td>
<td>15 M</td>
<td>C, 9; A, 1; AA, 8</td>
<td>14 (8.5–20)</td>
<td>523 (370.5–744.5)</td>
<td>37,568 (17,573–69,763)</td>
</tr>
<tr>
<td>Immunologic progressors (20)</td>
<td>43 (37–47)</td>
<td>17 M</td>
<td>C, 10; L, 5; A, 1; AA, 3, NA</td>
<td>8 (5–16)</td>
<td>200 (144.5–231.5)</td>
<td>150,974 (89,532–230,970)</td>
</tr>
<tr>
<td>HIV-1 negative (22)c</td>
<td>49 (44–54.5)</td>
<td>12 F</td>
<td>C, 8; L, 2; A, 3</td>
<td>13 (6–18)</td>
<td>794 (569–1,059)</td>
<td>50 (40–302)</td>
</tr>
</tbody>
</table>

a F, female; M, male.

b C, Caucasian; L, Latino/Hispanic; A, Asian; AA, African-American; NA, Native American.

c Some information about the gender and ethnicity of the HIV-1-negative group was not available.
RESULTS

HIV-1 controllers have stronger HERV-specific responses than other chronically infected subjects. We measured T cell responses against HERV using a set of manufactured peptides selected from three HERV families (HERV-K, HERV-L, and HERV-H). The total magnitude of the HERV-specific T cell response was measured as the sum of all of the single peptide responses for each individual. The median total magnitude of HERV-specific T cell responses among HIV-1-negative subjects was 122.1 SPM (interquartile range [IQR], 62.5 to 165 SPM). Individuals controlling HIV-1 viremia in the absence of treatment (controllers) had a median total response magnitude of 571.4 SPM (IQR, 240 to 1,395 SPM). This was significantly higher than the total responses of the HIV-1-negative subjects (P < 0.0001), the HAART-suppressed group (median, 270.3 SPM; IQR, 87.6 to 422.5 SPM; P < 0.05), the noncontroller group (median, 181.3 SPM; IQR, 32.5 to 327.5 SPM; P < 0.05), and the progressors (median, 77.5 SPM; IQR, 22.5 to 175; P < 0.0001) (Fig. 1A). HIV-1-infected controllers also had a significantly higher number (breadth) of HERV-specific re-

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sponses than the HIV-1-negative individuals \((P < 0.0001)\), the HIV-1-infected noncontrollers \((P < 0.01)\), progressors \((P < 0.0001)\), and HAART-suppressed patients \((P < 0.05)\) (Fig. 1B).

Untreated HIV-1-infected subjects with preserved CD4\(^+\) T cell counts do not differ in the magnitude of their HIV-1 and CMV responses. In order to characterize the baseline immune function of the HIV-1-infected subjects, responses to an HIV-1 peptide pool (which included Gag, Nef, Env, and Pol sequences) and a CMV pp65 peptide pool were assessed in a subset of individuals for whom adequate PBMC samples were available. There was no significant difference in the CMV pp65 pool response magnitude among the three HIV-1-infected groups with preserved CD4\(^+\) T cell counts \((P > 0.05\) for all pairwise comparisons) (see Fig. S1A in the supplemental material). However, the progressors had a lower median CMV response magnitude than the controllers and HAART-suppressed patients \((P < 0.01\) and \(P < 0.05\), respectively). The HIV-1 controllers, noncontrollers, and progressors did not differ in their responses to the HIV-1 peptide pool \((P > 0.05\) for all pairwise comparisons), but the controllers had stronger HIV-1 pool responses than the HAART-suppressed patients \((P < 0.05)\) (see Fig. S1B). Consistent with previous reports using the IFN-\(\gamma\) ELISPOT assay (2), there was no correlation between the magnitude of the HIV-1 pool response and HIV-1 plasma viremia in untreated subjects \((r^2 = 0.007, P = 0.525)\) (see Fig. S1C). There was also no significant association between the magnitude of the HIV-1 pool responses and the HERV responses in untreated patients \((r^2 = 0.033, P = 0.164)\) (see Fig. S1D).

HERV-specific immunity correlates with control of HIV-1. Twenty-seven out of the 28 HERV peptides tested elicited at least one positive response in the HIV-1-infected cohort, with the majority of the peptides eliciting responses from two or more individuals (Fig. 2). There was no difference between the number of “unique” HERV peptides that stimulated a response and the number of immunogenic HERV peptides that shared four or more amino acids in common with an HIV-1 epitope (“similar”; median number of responses elicited by both unique and similar HERV peptides, 4; \(P = 0.63\)). In the untreated HIV-1-infected individuals (controllers, noncontrollers, and progressors), the HIV-1 viral load was inversely correlated with the magnitude of the HERV T cell response \((r^2 = 0.197, P = 0.002\) by linear regression; Spearman \(r = -0.535, P < 0.0001)\) (Fig. 3A) and the breadth of HERV T cell
response ($r^2 = 0.115, P = 0.005$ by linear regression; Spearman $r = -0.339, P = 0.005$) (Fig. 3B). There was also a positive correlation between HERV response magnitude and CD4+ T cell count in untreated patients ($r^2 = 0.072, P = 0.027$ by linear regression; Spearman $r = 0.269, P = 0.027$) (Fig. 3C). The inverse correlation between HERV response magnitude and HIV-1 viral load persisted even when the progressor group (CD4+ T cell count of $<250$ cells/mm$^3$) was excluded ($r^2 = 0.091, P = 0.037$ by linear regression; Spearman $r = -0.302, P = 0.037$) (data not shown).

Mapping of responses against HERV-K Gag and Env in an HIV-1-infected controller. In order to more comprehensively characterize the T cell response against HERV-K (the youngest and most transcriptionally active HERV family), we tested a set of 336 overlapping 15-mer peptides spanning HERV-K Gag and Env in a controller (P1, for whom large numbers of PBMCs were available) who was infected with HIV-1 in 1990 and maintained an undetectable viral load over the following 2 decades. We detected a positive response to an HERV-K Env peptide, CIDSTFNWQHRILLV, at a 2003 time point (Fig. 4) that persisted and increased in magnitude over the next 5 years.

Characterization of responses against two similar HERV-K and HIV-1 epitopes in a unique subject. We further investigated one particularly immunogenic peptide from the reverse transcriptase region of HERV-K, FAFTIPAI (FI8). More than one-quarter of the HIV-1-infected controllers that were tested recognized this peptide. One individual (P2) had an especially robust ELISPOT response ($580$ SPM) to FI8, which titrated to $1 \mu g/ml$ (Fig. 5A). In addition, a strong response to an HIV-1 peptide with a similar sequence, TAFTIPSI (TI8), was observed. This subject was infected with HIV-1 in 1999 and maintained a CD4+ T cell count above 400 cells/mm$^3$ and an HIV-1 viral load below 6,000 copies/ml for almost a decade but eventually began HAART in 2007 (Fig. 5B). Longitudinal analysis of this individual’s responses to HERV-K FI8, HIV-1 TI8, and an HIV-1 peptide pool showed that all three waned in 2005, just before the patient initiated HAART, whereas the response to a CMV pp65 peptide pool was maintained (Fig. 5C).

In order to determine the HLA restriction of the HERV-K FI8 response in P2 (who was HLA-B51$^+$), we tested the ability of patient-derived PBMCs to recognize single HLA-allele B cell transfectants that had been pulsed with peptide FI8 in an ELISPOT assay. The peptide-pulsed HLA-B51$^+$ B cells were recognized to a greater degree than the unpulsed B cells and HLA-B51$^-$ B cells, confirming that P2’s response to this epitope is restricted by HLA-B51 (Fig. 6).

The HERV-K FAFTIPAI response is more differentiated and less activated than the HIV-1 TAFTIPSI response. To further characterize P2’s responses to the two similar peptides, HERV-K FI8 and HIV-1 TI8, we evaluated the functionality and phenotypes of the CD8+ T cell populations stimulated by these antigens. We also included a CMV pp65 pool in order to examine how these responses differ from one that is maintained long-term and is associated with a controlled chronic viral infection. An intracellular cytokine detection assay was used to analyze the simultaneous production of IFN-γ and TNF-α in antigen-specific cells. The CD8+ population stimulated by HERV-K FI8 had similar proportions of dual-cytokine (IFN-γ and TNF-α)-producing and monofunctional (IFN-γ$^+$) cells, as opposed to the primarily IFN-γ-only-producing pop-
population specific to the HIV-1 TI8 epitope (Fig. 7A; see also Fig. S2A in the supplemental material).

To assess the differentiation stage of CD8+ T cells that produced cytokines against HERV-K FI8, HIV-TI8, and the CMV pool, three subpopulations based on coexpression of T cell maturation markers CD27 and CD28 were quantified. CD8+ T cells were defined as CD27+CD28+ (early stage), CD27+CD28− (intermediate stage), or CD27−CD28+ (late stage) (3, 5). The IFN-γ+ population stimulated by HIV-1 TI8 had a higher proportion of cells at an early to intermediate stage of differentiation (CD27+CD28+ and CD27−CD28+). In contrast, the IFN-γ+ cells stimulated by HERV-K FI8 had a late stage of differentiation (Fig. 7B; see also Fig. S2B in the supplemental material). In addition, the HERV-K FI8-specific CD8+ T cells were less activated (as measured by a CD38 mean fluorescence intensity [MFI] of 1,615) than the HIV-1 TI8-specific cells (CD38 MFI, 2,603) and had a level of activation that was comparable to that of the CMV-specific CD8+ population (CD38 MFI, 1,458) (Fig. 7C; see also Fig. S2C).

Single-tetramer staining of PBMCs from patient P2 and an HIV-1 uninfected control with either HLA-B51-HERV-K FA FTIPAI-APC (Fig. 8A) or B51-HIV-1 TAFTIPSI-PE (Fig. 8B) showed tetramer-positive CD8+ populations in P2 but not in the control subject. Finally, by performing dual staining with both tetramers, we confirmed that HERV-K FI8 and HIV-1 TI8 stimulated separate CD8+ T cell populations in subject P2. We identified two distinct populations that each stained positive for either the HLA B51-HERV-K FAFTIPAI-APC tetramer or the B51-HIV-1 TAFTIPSI-PE tetramer (Fig. 8C).

**DISCUSSION**

Human endogenous retroviruses (HERVs) are thought to be inactive elements of our genome, normally held in check by host cellular restriction proteins such as APOBEC3G (11). We hypothesized that the impairment of these controls in HIV-1 infection could expose the immune system to HERV antigens that act as immutable targets for cytotoxic lysis, resulting in the containment of HIV-1 viremia. Previously, our lab has shown that HERV expression occurs in HIV-1-infected cells and that HERV-specific CD8+ T cell responses are stimulated in primary HIV-1 infection (12). These responses are negatively correlated with HIV-1 viral load (12). Here, we extend these observations to include individuals who have long-term chronic HIV-1 infection, focusing on a rare subset of individuals who are able to suppress HIV-1 indefinitely in the absence of combination therapy. The mechanism of control in these HIV-1 controllers remains to be fully defined. Although there is evidence that the HIV-specific T cell response contributes to virologic containment (1, 10), it is clear from a number of studies that other factors are likely to be involved (10, 26). A recent genome-wide association study in a multiethnic cohort of HIV-1 controllers and progressors showed that differences
in HLA alleles explain 19% of the variance of host control (27). In order to determine what effect, if any, HERV-specific T cell responses have on viral control, we studied a cohort of HIV-1 controllers and compared their responses to those of untreated virologic noncontrollers and immunologic progressors, patients on HAART, and HIV-1-uninfected controls. While it is likely that robust CD4⁺ T cell help contributes to strong CD8⁺ T cell responses, we sought to rule out that it was the primary cause of any observed differential responses to HERV antigens. Therefore, to avoid a confounding effect of generalized immune dysfunction due to a lack of CD4⁺ T cell help, we performed additional analyses that excluded the immunologic progressor group. Our finding that the magnitude of CMV pp65-specific responses was similar in the controller, HAART-suppressed, and noncontroller groups supports the fulfillment of this criterion.

We found that in chronic HIV-1 infection, controllers have HERV responses with higher magnitude and greater breadth...
than patients with viral suppression on HAART, virologic noncontrollers, immunologic progressors, and HIV-1-negative subjects. Interestingly, controllers who lack HLA alleles that are associated with protection from HIV-1 disease progression (HLA-B27 and -B57) constituted a large proportion of the subjects with the strongest HERV responses, suggesting that there may be an alternative mechanism of HIV control (such as HERV-specific cytotoxic T cells) in these controllers. There was no difference in the magnitude of the HIV-1-specific responses between the controllers, noncontrollers, and progressors (as defined by IFN-γ production) and no correlation between this measure and HIV-1 plasma viremia. These data are consistent with other studies reporting that neither the magnitude nor the breadth of the HIV-1-specific CD8+ T cell response is associated with a difference in viral load (2). Of note, there was also no significant correlation between the strength of HERV-specific responses and HIV-1-specific responses in untreated patients, suggesting that these are independent variables. In contrast, we discovered that the HERV-specific T cell response magnitude was inversely correlated with HIV-1 viral load (even when the immunologic progressors were excluded), suggesting a potential role for these responses in the control of HIV-1. There was also a positive correlation between HERV response magnitude and CD4+ T cell count in untreated individuals. The group with viral suppression on HAART responded to fewer HERV epitopes and had weaker responses, lending support to the hypothesis that these responses are a cause rather than a consequence of viral control.

Longitudinal analyses of HERV responses in two subjects revealed interesting differences. One individual (P2) had a strong, titratable response to the HERV-K FAFTIPAI epitope. This person was able to contain HIV-1 replication to a strong, titratable response to the HERV-K FAFTIPAI, revealing interesting differences. One individual (P2) had a stronger HERV response than the controllers, noncontrollers, and progressors (as defined by IFN-γ production) and no correlation between this measure and HIV-1 plasma viremia. These data are consistent with other studies reporting that neither the magnitude nor the breadth of the HIV-1-specific CD8+ T cell response is associated with a difference in viral load (2). Of note, there was also no significant correlation between the strength of HERV-specific responses and HIV-1-specific responses in untreated patients, suggesting that these are independent variables. In contrast, we discovered that the HERV-specific T cell response magnitude was inversely correlated with HIV-1 viral load (even when the immunologic progressors were excluded), suggesting a potential role for these responses in the control of HIV-1. There was also a positive correlation between HERV response magnitude and CD4+ T cell count in untreated individuals. The group with viral suppression on HAART responded to fewer HERV epitopes and had weaker responses, lending support to the hypothesis that these responses are a cause rather than a consequence of viral control.

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required to generate a response in many of our subjects. Low-avidity T cells (which are more prone to tolerance induction and are stimulated only with high antigen load) have been shown to proliferate better than high-avidity T cells and to lack their replicative defects (16). Therefore, HERV-specific CTLs may play a role in the containment of HIV-1 due to a superior ability to proliferate. Of note, we previously showed that despite being directed against self-antigens, HERV-specific CTLs are not impaired in their ability to kill cells pulsed with their cognate peptide (12). Overall, our present study has identified phenotypic and functional traits of the HERV-specific response that may confer an enhanced ability beyond that of most HIV-specific CTLs to effectively target and lyse infected cells. Our finding that the HIV-1 viral load was lower in patients with stronger and broader HERV responses provides empirical evidence for this.

Our results constitute the discovery of a novel correlate of immune protection from disease progression in chronic HIV-1 infection and have implications for both a better understanding of HIV-1 pathogenesis and a potential new approach to HIV-1 vaccines. While the primary advantage of a therapeutic agent derived from HERVs is their conserved nature, features associated with a superior cytotoxic ability offer an additional benefit to the induction of HERV-specific responses. Our findings show that individuals who can control HIV-1 in the absence of HAART have the greatest HERV responses. This HERV-specific immunity is correlated with the containment of HIV-1 replication and preservation of CD4+ T cell count in untreated subjects and provides strong support for the further investigation of an HERV-based HIV-1 vaccine.

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