Pediatric HIV-1-infection: perspectives on vaccination strategies and immune reconstitution during long-term antiretroviral therapy
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Chapter

Epstein-Barr virus infects B and non-B lymphocytes in HIV-1-infected children and adolescents

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Abstract

Context: Epstein-Barr virus (EBV) is a widespread, persistent herpes virus that can transform B cells and is associated with malignant lymphomas. EBV dynamics and specific immunity in HIV-1-infected children are unknown.

Results: In 74% of EBV-seropositive HIV-1-infected patients at start of HAART, we found EBV DNA levels comparable to those in acutely EBV-infected HIV-negative children. EBV load remained elevated in most HIV-1-infected children for months to years of follow-up. Frequencies of IFN-γ-producing EBV-specific CD8+ T cells were comparable to healthy controls and antibodies against EBV nuclear antigens (EBNA) were detected in 73% of the EBV-seropositive children. Detectable EBV DNA was not correlated with HIV RNA levels or CD4+ T-cell increase upon start of HAART. Because of its reminiscence of chronic active EBV (CAEBV), we studied cellular tropism of EBV in these patients. EBV DNA was not only found in CD19+ B cells, but also - at stable levels - in CD4+ and CD8+ T cells.

Conclusion: Although the reason for T-cell tropism of EBV remains unclear, these data may provide an explanation for the differential EBV dynamics in the presence of normal serology, as well as the long-term risk for the development of lymphoproliferative diseases in HIV-1-infected individuals.
Introduction

Epstein-Barr virus (EBV) is a widespread human γ-herpes virus. Young children are mostly asymptomatic or experience non-specific symptoms during primary EBV infection. In adolescents and adults, EBV infection can cause self-limiting infectious mononucleosis [1]. After the initial increase in viral DNA in the blood, EBV establishes latency mainly within the B cell compartment [2-4], with EBV DNA remaining detectable in a fraction of the memory B cells [5].

In the absence of adequate immune control after hematopoietic stem-cell transplantation or after solid-organ transplantation, strongly elevated or increasing EBV loads constitute a risk factor for the development of post-transplant lymphoproliferative disorders [6]. Similarly, EBV DNA is often detected at considerable levels in blood of HIV-1-infected patients and HIV-1-infected individuals are at increased risk of lymphoproliferative disorders [7-10].

However, most of the studies in HIV-1-infected patients are cross-sectional without considering the role of HAART and have been performed in male adults. In these studies the sequence of infection, HIV prior to or following EBV, is unknown. Although it may be questioned whether the effect of HAART in adults is of any influence [11,12], the relevance of a prospective study in a well-characterized patients cohort is obvious, in particular at an age at which acute primary EBV infection often takes place. Although EBV infection is one of the important risk factors in HIV-1-infected children for developing lymphoproliferative disorders [13], studies on the dynamics of EBV in these children are lacking.

It was suggested that elevated EBV loads in HIV-1-infected adult patients was correlated with a loss of immune control [14,15]. Alternatively, EBV DNA may be elevated as a consequence of immune activation induced by acute HIV-1-infection [10]. However, until now, no biologic explanation for the elevated EBV levels in HIV-1-infected patients has ever been given. In analogy with a syndrome known as Chronic Active EBV (CAEBV) in children or adolescents [3], we investigated whether the cellular tropism of EBV is changed in HIV-1-infected children and if so, whether this would correlate with a failing immune system.

Methods

Subjects
All children and adolescents with HIV-1-infection who started HAART between 1997 and 2004, who were followed-up in our institution and were not pregnant, were included in this study. Since HAART was introduced in 1997 as treatment of pediatric HIV infection, 54 children started treatment, 29 females and 25 males. The median age at start HAART was 3.8 years (Inter-Quartile Range (IQR) 1.2-7.0). Baseline characteristics of the patient cohort are shown in Table 1. The patients started HAART,
Table 1. Baseline characteristics

<table>
<thead>
<tr>
<th>Children on HAART</th>
<th>Number (n)</th>
<th>54</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age at start HAART (years, median (IQR))</td>
<td>3.8 (1.2-7.1)</td>
<td></td>
</tr>
<tr>
<td>Sex (m/f)</td>
<td>25/29</td>
<td></td>
</tr>
<tr>
<td>HIV RNA (median log (IQR))</td>
<td>4.84 (4.14-5.78)</td>
<td></td>
</tr>
<tr>
<td>CDC-classification (n)*</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Non-C</td>
<td>30</td>
<td></td>
</tr>
<tr>
<td>C</td>
<td>24</td>
<td></td>
</tr>
<tr>
<td>Weeks since start HAART</td>
<td>210 (89-315)</td>
<td></td>
</tr>
<tr>
<td>EBV VCA IgG at start</td>
<td></td>
<td></td>
</tr>
<tr>
<td>VCA positive</td>
<td>47</td>
<td></td>
</tr>
<tr>
<td>VCA negative</td>
<td>2</td>
<td></td>
</tr>
<tr>
<td>VCA seroconversion</td>
<td>5</td>
<td></td>
</tr>
</tbody>
</table>

*Patients were classified according to the clinical categories as defined by the US Centers for Disease Control and Prevention (CDC) [30], from A to C, were C indicates the most severe cases.

EBV PCR was done on prospectively collected samples as part of clinical follow-up and on frozen PBMCs.

Lymphocyte subset determination
Fresh human PBMCs were isolated from EDTA anti-coagulated whole blood samples by Ficoll-Paque density centrifugation (Pharmacia Biotech AB, Uppsala, Sweden). Numbers of B cells (CD19+), T cell (CD3+) and T cell subsets (CD3+CD4+, CD3+CD8+) were determined by standard FACScan procedures with monoclonal antibodies (Sanquin Research/CLB, Amsterdam, the Netherlands).
EBV DNA was amplified and detected using an end-point quantitative PCR with TBR-labeled probes for wild-type EBV DNA and internal control DNA (IC), in a similar way as earlier described [16]. Primers (Biotinylated-EBV-1: 5'-TGG TGC GCC GGT GTG TTC GTA TA-3', EBV-2: 5'- GGT GGA AAC CAG GGA GGC AAA TC -3', amplimer length: 329 bp) were diluted in TE buffer (10 mM Tris-HCl, 1 mM EDTA, pH 8.0) to 100 ng/μL. The primer binding sites are situated in the EBNA-1 gene. The limit of detection of the assay was 80 EBV copies/mL of whole blood. In cases where whole blood EBV DNA data is missing and serology or EBV PCR on PBMCs was negative, the whole blood EBV DNA load was set at 1 for the analyses.

EBV serology
Specific IgM and IgG against the Epstein Barr virus viral capsid antigen (EBV-VCA) and specific IgG to Epstein-Barr nuclear antigen (EBNA) were determined using the ‘anti-EBV VCA IgG ELISA’ and the ‘anti-EBV EBNA IgG ELISA’ of Biotest respectively (Dreieich, Germany). All tests were performed according to the manufacturers’ instructions. Seropositivity is defined by the presence of a positive specific IgG after the age of 12-18 months.

Plasma HIV-1 RNA determination
Plasma HIV-1 RNA concentration was determined by the use of the Nuclisens HIV-1 RNA QT (Biomérieux, Boxtel, the Netherlands) until 2001 with a lower detection limit of 400 copies/mL. After January 2001, the Versant HIV-1 RNA 3.0 (Bayer, Tarrytown, NY, USA) was used with a lower limit of detection of 50 copies/ml. All tests were performed according to the instructions of the manufacturers. For the present analyses all HIV-1-RNA below 400 copies/mL were considered as undetectable.

Cell sorting
Purified CD4+ T cells, CD8+ T cells and CD19+ B cells were obtained by using multi-parameter magnetic cell sorting (MACS). Briefly, fresh PBMCs were thawed and after careful washing, the cell pellet was re-suspended in MACS-buffer. Cells were labeled by adding 20 μL CD19 MACS Beads (Miltenyi Biotech, distributed by Sanquin Reagents, Amsterdam) per 107 total cells and incubated for 15 minutes at 4°C. CD19+ B cells were separated using a positive selection column (type MS+/RS+) placed in a separator. After washing the column with MACS-buffer, the column was removed from the separator and the B-cells were eluted into a collection tube. The negatively selected cells were further separated by positive selection using CD4 and subsequent CD8 MACS beads. As a consequence B-cell contamination of the T-cell fractions was minimized. FACS analyses of purified fractions showed that the contamination of CD4+ and CD8+ T-cell fractions with CD19+ B cells was less than 0.1%.

EBV DNA per mL of whole blood was calculated as absolute EBV DNA copy number times the compartmental cell count per mL.
Flow cytometry and intracellular IFN-γ staining after antigen-specific stimulation

EBV-specific T-cell responses were measured in PBMC of 18 children with a detectable EBV DNA level after stimulation with an overlapping EBV-peptide pool derived from the immunodominant BZLF-1 antigen. Four-color fluorescence analysis was performed as previously described [17]. First, 2×10⁶ freshly isolated PBMC/ml were stimulated with 2 μg/mL of a 15-mer 11 amino acids overlap BZLF-1 peptide pool (Jerini AG, Berlin, Germany) or an irrelevant peptide pool in the presence of 2 μg/mL co-stimulation with both anti-CD28 (Sanquin reagents) and CD49d (Becton Dickinson (BD)) monoclonal at 37°C for 6 hours, as previously described [18].

Statistical analyses

All statistical analyses were performed using SPSS for Windows version 11.5 (SPSS, Chicago, Illinois, USA). Differences between groups were evaluated using the Fisher’s exact test for categorical data and the Kruskal Wallis test for continuous data. Differences between baseline and follow-up were evaluated using a paired sample t-test. Age correction for CD4⁺ and CD8⁺ T cells and CD19⁺ B cells was done by dividing the counts by the mean of an age-matched healthy control group [19]. Correlation was tested using the Spearman-rank-sum test. A linear regression model was used to correlate continuous data and various independent variables in a univariable model. A stepwise backward method was chosen for the multivariable model. All variables were added to the model when they were significant at the 0.1 level in the univariable analysis. All tests are 2-sided. A p-value <0.05 was considered statistically significant.

Results

Immunology: CD4⁺, CD8⁺ T cells, CD19⁺ B cells

T- and B-lymphocyte populations and EBV DNA load were analyzed over time in all 54 patients from the start with HAART onward. Median time since HAART at time of analyses was 210 weeks. Lymphocyte counts were adjusted to the mean of age-specific reference groups and expressed as ratios. The median CD19⁺ B-cell ratio at baseline was 0.60, and significantly increased during 48 weeks of HAART to 0.80 (p=0.006), and 0.88 during 96 weeks of HAART (p=0.006). An increase from median CD4⁺ T-cell ratio of 0.35 at baseline, to 0.78 at week 48 (p<0.001) and to 0.78 at week 96 (p<0.01) was seen. The CD8⁺ T-cell ratio remained relatively stable over time (1.53, 1.49 and 1.43 at week 0, 48 and 96, respectively).

EBV DNA at baseline

Of the 54 children, 47 children were EBV seropositive at the start with HAART, five seroconverted during treatment and 2 children remained seronegative during the entire follow-up period (Table 1). Anti-EBNA serology was positive in 37 of 47 (73%) VCA-positive patients. The EBV DNA load before initiation of HAART was known in 40
EBV-seropositive children (40/47 (85%)). The load was compared with EBV DNA in healthy controls that were either acutely or latently infected with EBV.

An increased proportion of HIV-1-infected children had detectable EBV DNA compared to otherwise healthy children with latent EBV, viral capsid antigen (VCA) IgG-positive and Epstein-Barr nuclear antigen (EBNA) IgG-positive (35 of 40 patients (88%) vs. 10 of 45 controls (22%); p<0.001).

Median EBV DNA was much higher in HIV-1-infected children compared to latently EBV-infected children (median 3.7 vs 0.0 log copies/mL; p<0.001) (Fig.1). Baseline EBV load in HIV-1-infected children was higher than in children with acute EBV-infection (median 3.7 vs. 3.1 log copies/mL; p=0.02) (Fig.1).

The EBV load in HIV-1-infected children at baseline was not correlated with age (r= -0.76, p=0.7), HIV RNA concentration (r= 0.04, p=0.8), or age-corrected CD4+ T cell ratio (r= 0.2, p=0.2) and age-corrected CD19+ B cell ratio (r= 0.3, p=0.1).

No change in EBV-DNA levels in most of the HAART-treated patients
The effect of HAART on EBV DNA was studied longitudinally in all children. EBV DNA in whole blood remained high, despite the numerical recovery of CD4+ T- and CD19+ B-lymphocytes and proliferative capacity under HAART (Fig.2A; and data not shown). Only 11 EBV-seropositive children had EBV loads below the lower limit of detection (<80 copies/mL) on two consecutive occasions (Fig.2B).

Of these children, 7 were able to suppress EBV after the start with HAART in a median time of 158 weeks (IQR 45-240), whereas in only 4 children EBV DNA was already undetectable at the start of HAART.
Of the 54 children, 7 were EBV-seronegative at start with HAART. They were younger than the EBV-seropositive children (median age 0.8 vs. 4.7 years, p=0.002). Their baseline T- and B-cell ratios and HIV RNA were not different from the EBV-seropositive patients (data not shown). During follow-up, subclinical primary EBV-infection was observed in 5 out of these 7 HAART-treated children. This resulted in persistently elevated EBV DNA loads (Fig. 2C). Their EBV DNA also persisted during follow-up, similar to the HIV-1-infected children who already were EBV-positive prior to HAART, even though HIV-1 RNA was continuously below the lower limit of detection due to HAART in 2 of these patients.

EBV DNA remained detectable during follow-up in 41 HIV-1-infected children. None of the variables tested was found to be associated with detectable EBV DNA or not (Table 2). Also the change in EBV load over time and change in CD19+ B or CD4+ T cells were not associated (p=0.17 and p=0.75).
Table 2. EBV DNA in whole blood; factors potentially associated with suppression of EBV below the lower limit of detection.

<table>
<thead>
<tr>
<th></th>
<th>EBV suppression *</th>
<th>Persistent EBV *</th>
<th>Total #</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Number</strong></td>
<td>11</td>
<td>41</td>
<td>52</td>
<td></td>
</tr>
<tr>
<td>Age at start HAART (years, median (IQR))</td>
<td>4.8 (1.8-15.5)</td>
<td>4.0 (1.1-6.4)</td>
<td></td>
<td>0.32</td>
</tr>
<tr>
<td>Sex (m/f)</td>
<td>4/7</td>
<td>21/20</td>
<td>25/27</td>
<td>0.50</td>
</tr>
<tr>
<td>Mode of HIV-1 infection (vertical vs sexual)</td>
<td>9/2</td>
<td>38/3</td>
<td>47/5</td>
<td>0.29</td>
</tr>
<tr>
<td>CDC (C / non-C)</td>
<td>3/8</td>
<td>20/21</td>
<td>23/29</td>
<td>0.31</td>
</tr>
<tr>
<td>HIV virologic failure vs. response at week 48</td>
<td>1/10</td>
<td>6/35</td>
<td>7/45</td>
<td>1.00</td>
</tr>
<tr>
<td>EBV VCA serology at start (pos vs neg)</td>
<td>11/0</td>
<td>36/5</td>
<td>47/5</td>
<td>0.57</td>
</tr>
<tr>
<td>EBNA IgG at start (pos vs neg)</td>
<td>9/2</td>
<td>28/12</td>
<td>37/14</td>
<td>0.71</td>
</tr>
<tr>
<td>EBNA IgG after 48 weeks (pos vs neg)</td>
<td>7/2</td>
<td>24/10</td>
<td>31/12</td>
<td>1.00</td>
</tr>
</tbody>
</table>

* Cut-off at 80 copies/mL. # Excluded in these analyses are 2 children that remained EBV seronegative during the entire follow-up period.

**EBV-specific CD8+ T-cell responses**

As EBV-specific CD8+ T cells are known to play a major role in the control of EBV infection, we studied whether the elevated EBV load in HIV-1-infected children could be explained by diminished frequencies of these cells. Therefore EBV-specific CD8+ T-cell responses, determined by measurements of IFN-γ production after stimulation with the immediate-early lytic protein BZLF-1 were compared between 18 HIV-1-positive children and 11 HIV-negative children. The EBV-specific response was below the sensitivity of the assay in 6 (35%) HIV-1-infected children. Median IFN-γ producing CD8+ T cells was 0.14% (IQR 0.03-0.53%). In healthy pediatric controls the EBV-specific response was below the sensitivity of the assay in 5 (38%) children, with a median of 0.10% (IQR 0.0-0.20%) of CD8+ T cells producing IFN-γ upon stimulation with BZLF-1-peptide pool (Fig.3). In HIV-1-infected children, numbers were not correlated with the EBV load determined simultaneously (r=0.34; p=0.24).

![Figure 3. EBV specific cellular responses. % IFN-γ producing CD8+ T cells after stimulation with BZLF-1 peptide pool, comparing HIV-1-infected children with pediatric controls. Horizontal bars represent median values.](image-url)
EBV DNA present in CD19\(^{+}\), CD4\(^{+}\) and CD8\(^{+}\) T cells

Neither immune function nor serology was associated with persistent high EBV concentrations. Hence, we considered the possibility that EBV was present in other cell types than CD19\(^{+}\) B cells, similar to conditions such as chronic active EBV (CAEBV) and acute EBV-associated hemophagocytic lymphohistiocytosis (EBV-HLH) [3,20]. In order of attendance, 16 patients with an elevated EBV load were chosen for further investigation of EBV DNA in CD4\(^{+}\) T, CD8\(^{+}\) T and CD19\(^{+}\) B cells. In these cell fractions, EBV DNA was found in CD19\(^{+}\) B cells (median 3.01 log copies/106 cells, IQR 2.8-3.7), but also in CD4\(^{+}\) (median 2.3 log copies/106 cells, IQR 1.1-2.7) and CD8\(^{+}\) T cells (median 1.8 log copies/106 cells, IQR 1.3-2.4) (Fig.4A). Two of the HIV-1-infected patients became infected by EBV many weeks after the start of HAART (>30 weeks), having complete and prolonged suppression of HIV-1.

In otherwise healthy children with acute EBV infection, EBV DNA was predominantly detected in the CD19\(^{+}\) B-cell compartment (Fig.4A). In an additional two children with EBNA-IgG in the absence of VCA-IgM, and with whole blood EBV loads of 600 and 800 copies/mL, respectively, we could obtain enough cells for further cell fractionation. EBV DNA was only found in their CD19\(^{+}\) B-cell compartment, similar to most of the controls suffering from acute EBV infection. One of these two became negative for EBV PCR on whole blood within the next 4 months of follow-up (data not shown).

Seven out of 16 HIV-1-infected patients with serial samples were reanalyzed 6 months after the first sample analysis. Similar distribution of EBV in the various lymphocyte-types was found in most of these 7 patients, apart from one patient with a decrease in EBV copies, as observed in particular in the CD4\(^{+}\) T cells (Fig.4B). This patient also showed a decreasing EBV load in the whole blood PCR test explaining the results obtained.

**Figure 4A.** EBV DNA in cellular compartments. EBV DNA in peripheral blood mononuclear cells measured in separated cell fractions of 17 HIV-1-infected patients and 6 otherwise healthy patients with acute EBV infection.

**Figure 4B.** EBV DNA in cellular compartments over time. EBV DNA in PBMC in 6 patients measured on 2 occasions 6 months apart. Due to technical causes not all cellular compartments were available for EBV PCR at both time points. Numbers of data missing per time-point and fraction ranges between 0 and 2.
Discussion

In this prospective study we demonstrate that the EBV dynamics and cellular tropism in HIV-1-infected children and adolescents differ dramatically from HIV-negative controls and were independent of the effects of HAART. First, patients showed relatively normal anti-VCA and EBNA serology or EBV-specific CD8+ T-cell responses, as may be expected after re-convalescence of acute EBV infection, while their EBV load remained elevated. Second, primary EBV infection after HAART-related immune reconstitution still resulted in an abnormal pattern of EBV infection and clearance with similar unusual dynamics as observed in patients who were EBV-positive prior to HAART.

EBV DNA was detected in CD4+ and CD8+ T cells, and CD19+ B cells, whereas otherwise healthy children and adolescents suffering from acute EBV infection showed EBV DNA only in their CD19+ B-cell population. Due to the sequential purification steps, the contamination of the CD4+ and CD8+ T cell compartments was less than 0.1%. We calculated that a median contamination of 17% of B cells would be needed to account for the EBV load detected in the sequentially purified T cell fractions, based on the assumption that the copies of EBV were equally found in all B cells. This makes the possibility of B cell contamination as an explanation for our findings highly unlikely.

Even though we tried to relate the abnormal EBV dynamics to various HIV-related disease and immune parameters, or to failing EBV-specific immunity, the exact mechanism behind the aberrant T-cell tropism and failure in normal EBV suppression remains unclear. Also after immune reconstitution and increased CD4+ T cell counts due to HAART, EBV loads remained elevated at individual levels. It has been described that after HIV-seroconversion EBV DNA increases thereby installing a new viral set point [10]. In our study population EBV load also seems to be elevated at individually defined set points. HAART does not seem to be able to change this altered balance between the virus and the host. This is reminiscent of the situation described in one study on adult homosexuals treated with HAART [12,21].

In a minority of patients (7 of 35) with detectable EBV DNA at the start of HAART, this EBV load became undetectable during follow-up - irrespective of the response of HIV after 48 weeks on HAART. We could not identify any indicator that favors this suppression of EBV as is observed in the non-immunocompromized controls. We did not find a difference between vertical transmissions and sexually acquired HIV-1 infection. Also HAART treatment prior to primary EBV infection showed surprisingly little impact on subsequent EBV dynamics. However, the small number of children in both groups makes it impossible to completely rule out any association tested.

The finding that in HIV-1-infected individuals EBV infects other blood cell types besides the B lymphocytes is reminiscent of chronic active EBV (CAEBV) and acute EBV-associated hemophagocytic lymphohistiocytosis (EBV-HLH) [3]. In the acute phase of EBV-HLH, EBV is detected in the activated CD8+ T-cell population, whereas in CAEBV populations of CD4+ T cells and NK cells are primarily infected instead of the B cells [3]. However, these EBV-related diseases are characterized by the presence
of serious clinical symptoms and a high mortality rate [20], in contrast to chronic EBV infection in HIV-1-infected patients where both B and T cells are infected. Whether the balance of EBV tropism between B and T cell compartment defines the clinical symptoms remains unclear. Two Caucasian patients with CAEBV in our department (one transplanted with an HLA-identical sib after treatment for Hodgkin’s disease and one died from a cerebral non-Hodgkin lymphoma) EBV was detected in both B and T cells [Dolman, Van Lier, Kuipers, unpublished].

In immunocompetent individuals EBV establishes latent infection in B-cells [5]. Infection of these B-cells occurs through the complement receptor 2 (CR2, CD21). We now show that in HIV-1-infected children EBV also infects T cells, as also observed in CAEBV and HLH. Infection of T cells may occur through the same CD21 receptor that may be expressed under conditions of T-cell activation [22]. On the other hand, coreceptors or alternative ligands for EBV entry into activated T cells cannot be excluded, since in vitro data suggested that CD21+ T cells can be infected by EBV as well [23]. In this respect, the ectopic CD40 expression on T cells from CAEBV patients may support an aberrant EBV tropism [24]. However, we were not able to find any CD21 or CD40 staining on peripheral blood T cells from treated HIV-infected children (n=19) or pediatric controls (n=15) (data not shown). Whether T-cell activation in the secondary lymphoid organs or the expression of CD21 on double-negative T lymphocytes in the thymus, may render T cells susceptible to EBV infection [25], cannot be excluded.

In otherwise healthy individuals the EBV load during primary EBV infection drops rapidly with 2 log10 in the first 3 weeks following presentation with acute mononucleosis [26]. In the CD8+ T-cell response against EBV, the predominance of lytic over latent protein-specific CD8+ T cells is well documented, in particular during the (sub) acute infection. Both become detectable after a few weeks and remain stable over long periods of time [26-29]. EBV-specific CD8+ T-cell responses to BZLF1 in our population were detected at similar or slightly increased frequencies as determined in healthy EBV-seropositive children. These results at least suggest no impairment of EBV specific CD8+ T-cell responses. It may be at the level of CD4+ T-cell reactivity that the EBV-specific immunity is failing, different from the normal B-cell help and serologic responses against EBV.

For the first time to our knowledge, a difference is being revealed in the cellular tropism and EBV dynamics of EBV in HIV-1-infected individuals in this study. It is unknown whether these children with aberrant T-cell tropism of EBV are prone to develop EBV-related lymphomas. The prognostic value of high EBV loads and the infection of non-B lymphocytes by EBV are to be studied in larger cohorts. Our results demonstrate longitudinally that most HIV-1-infected children are not able to prevent the dissemination of EBV over the immune system during acute infection and cannot suppress EBV appropriately — irrespective of the success of HAART.
Acknowledgment

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References


