Parvovirus B19 in HIV+ adult patients with different CD4+ lymphocyte counts

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Abstract

Purpose. Human parvovirus B19 (B19V) can cause anemia in immunocompromised patients. We aimed to investigate the presence of B19V in HIV+ adults with different CD4+ T cell counts, to recognise the frequency of B19V in these different conditions and its possible association with anemia.

Methodology. We studied B19V specific IgM, IgG and DNA in 98 HIV+ patients and in 52 healthy individuals. HIV load, CD4+ counts and haemoglobin level were also determined in the patients.

Results. No individual in the control group had detectable IgM, 41/52 (78.8 %) had IgG and 5/52 (9.6 %) had B19V DNA. Among HIV+ patients, we found 5/98 (5.1 %) IgM+, 66/98 (67.3 %) IgG+ and 15/98 (15.3 %) had B19V DNA (no significant differences between the two groups compared). Considering the CD4+ cell range in HIV patients, 37 had <200 CD4+ cells ml⁻¹, 31 had 200 – 500, and 30 had >500. Anti-B19V IgG prevalence in patients with >500 CD4+ cells ml⁻¹ was significantly higher than in the rest (P=0.004) and compared to the control (P=0.046). B19V DNA concentration was always <10³ IU ml⁻¹, including 5 healthy individuals and 15 HIV+ patients. There was no significant association between B19V IgM or DNA and anemia nor between B19V DNA and HIV load.

Conclusions. The results indicate that B19V is not a high-risk factor for anemia in adult HIV+ patients under HAART treatment. Further studies will contribute to elucidate the mechanisms and significance of B19V DNA prevalence/persistence in adults, independently of the CD4+ cell status.

INTRODUCTION

Human parvovirus B19 (B19V) is the agent of the fifth disease, a pathogen of endemoepidemic circulation worldwide common in children and teenagers [1–4]. It is also associated to arthritis or arthralgia in adults, which have been related to the presence of the virus in local tissue. Given B19V tropism for the progenitor cells of the erythroid lineage, the virus may cause severe anemia under certain hematological and immunological conditions of the host and interfere with the normal course of pregnancy [5, 6]. B19V replication triggers the death of hematopoietic precursors, but usually there is no clinical impact in previously healthy, immunocompetent hosts. However, when the immune response is deficient or absent, the ongoing viral replication can cause anemia [7]. Thus, in lack of an adequate immune response that controls the primary infection in immunosuppressed individuals, they develop a persistent infection associated with chronic anemia [8, 9].

B19V induced-anemia in HIV+ individuals usually resolves satisfactorily after the initiation of highly active antiretroviral therapy (HAART), as limiting the retrovirus replication allows the restoration of the immune function and B19V control [10]. This demonstrates the role of specific immune response in the balance of B19V infection (acute/persistent). On the other hand, there have been complicated cases requiring blood transfusion in patients receiving antiretroviral therapy properly [11, 12], which could suggest a mechanism that facilitates viral persistence independent of immune failure. In fact, B19V DNA has been observed in healthy individuals during prolonged periods after acute infection, but the mechanism by which the clearance of the virus is slow, allowing its persistence, is not known [13–15].
In this context, our objective was to investigate the presence of B19V in HIV+ adult patients with different counts of CD4+ T cells and in healthy individuals, to estimate the frequency of the parvovirus in these different conditions and its possible association with anemia.

METHODS

Design

Descriptive, transversal study. We carried out a retrospective analysis of data from prospectively collected blood samples.

Study population

HIV+ patients and healthy adult individuals were studied. HIV+ patients were recruited during attendance for a follow-up control in the downtown area of Cordoba, Argentina, during 2012–2014. Healthy individuals were enrolled as participants of the reference group in situations of routine control, in the same period and area of the city. One serum sample was studied from each participant. An independent Ethics Committee (UNC-06200846379) reviewed and approved the protocol. Participants of both groups signed informed consent.

Serology tests

Anti-B19V IgM and IgG were detected by means of ELISA test RIDASCREEN (R-Biopharm) as detailed by the manufacturer. Samples with equivocal results were re-tested and samples with a second equivocal result were discarded.

Qualitative B19V DNA detection

Nucleic acids were extracted from 200 µl aliquots of serum using a method based on lysis with guanidine thiocyanate buffer and precipitation with silica, as described previously [4]. PCR was performed with primers designed to amplify a region of NS1 (nucleotides 2035–2058 and 2255–2276 of the genome with GenBank accession number NC_000883.2), using 0.2 µM of each primer, 0.8 mM desoxinucleotide mix, 2.5 mM MgCl2, 0.02 U µl−1 Taq polymerase (Invitrogen) and 5 % DMSO.

B19V DNA quantitation

B19V DNA+ samples were tested by semi-quantitative PCR and real time-PCR techniques to estimate B19V concentration. For these assays, DNA was extracted from a stored aliquot of serum, using QIAamp DNA Mini Kit (Qiagen). Both protocols used the same set of primers, which target a region of NS1 (nucleotides 2082–2099 and 2254–2274 of the genome with GenBank accession number AF162273) and were described previously by Koppelman et al. [16]. The results are expressed as international units (IU), according to WHO recommendations during the creation of the first B19V DNA standard [17] and subsequent revisions [18, 19]. The limit of detection of the real-time PCR was 500 IU ml−1 of plasma.

Analysis of data

To examine the possible association between B19V and immunosuppression, HIV+ patients were classified into three subgroups according to the number of CD4+ T cells mm−3 (<200, 200–500 and >500), which were determined with a Beckman Coulter Epics XL Flow Cytometer. The presence of anemia was identified according to WHO standards, which define anemia when the haemoglobin level is less than 13.0 g 100 ml−1 in adult men and less than 12.0 g ml−1 in women (not pregnant). A value of haemoglobin below 8.0 g ml−1 represents severe anemia [20]. The levels of haemoglobin were determined with CELL-DYN RUBY Hematology Analyzer (Abbott Diagnostics). HIV viral loads were determined by RT-PCR COBAS TaqMan HIV-1 test (ROCHE). For patients with undetectable HIV viral loads, an arbitrary value of 49 copies ml−1 was assigned in order to calculate mean values of each CD4 count group (the limit of detection of the quantification technique was 50 copies ml−1). In the comparisons performed, statistical significance was determined using ANOVA and Fisher exact test (α=0.05).

RESULTS

Fifty-two healthy adults with no immunological compromise and 98 HIV+ patients constituted the reference and the study group, respectively. In the first one, 26/52 (50.0 %) were males, and the age range was 18–57 years old (mean 35.7±10.1). The characteristics of the HIV+ group are shown in Table 1.

Among these patients, the average viral load of HIV was higher in the subgroup with lower CD4+ count. In general, concentration of haemoglobin did not display a marked tendency following the variation of HIV load but was lower the lesser CD4+ cell counts (Fig. 1); according to the definition given, 11 HIV+ patients had anemia and all of them had low CD4+ counts (Table 1). All HIV+ patients had initiated HAART therapy by the moment of obtaining the blood sample for this study.

No individual in the control group had anti-B19V IgM, while 5/52 (9.6 %) participants had B19V DNA and 41/52 (78.8 %) were immune (IgG+). In the HIV+ group, IgM anti-B19V was detected in 5/98 (5.1 %) patients (P=0.164 compared to the control group), and the prevalence of DNA and IgG were 15/98 (15.3 %, P=0.451) and 67/98 (68.4 %, P=0.186), respectively (Fig. 2a). Two of the IgM+ patients also had detectable B19V DNA. Considering the patients classified according to CD4+ cell count, there were no significant differences in the distribution of IgM or viral DNA in the 3 subgroups; however, patients with CD4+ cell counts >500 mm−3 had significantly higher prevalence of IgG (Fig. 2b, Table 2), which was also significantly different from the control group (P=0.046).

B19V DNA concentration was ≤104 IU ml−1 in all the identified positive samples (5 controls and 15 HIV+ patients). Three patients in the cluster of 15 B19V DNA-positive patients (20 %) were diagnosed with anemia; all three had CD4+ cell counts less than 200 mm−2 and one had IgM anti-B19V (Table 3). On the other hand, 8/75 (10.7 %) HIV+ patients without B19V DNA detection were anemic.
As a result, the frequency of anemia in the HIV+/B19V DNA+ versus the HIV+/B19V DNA- patients was not significantly different ($P=0.366$). In addition, three of the four patients identified with B19V DNA but without immunological markers of infection (IgM/IgG) were not anemic (patients 1, 6, 9, and 11, Table 3). Finally, an association between the presence of B19V DNA and the condition of detectable or undetectable HIV viral load was not evident ($P=0.133$).

**DISCUSSION**

Overall, the parameters of B19V infection were not different in the group of HIV+ patients compared to the control group. The exception was the prevalence of anti-B19V IgG in the subgroup of HIV+ patients with CD4+ cell count above 500 mm$^{-3}$, which was significantly higher than in the rest (Fig. 2, Table 2).

The decrease in the haemoglobin level as the number of CD4+ cells diminishes (Table 1, Fig. 1) can be attributed to both the treatment and the individual impaired immune and hematological condition of these patients. Zidovudine or AZT is the most commonly antiretroviral drug used in our study population and it is known to generate anemia [21]. It should also be noted that one of the complications characteristic of advanced stages of infection may involve weight loss of 10% or more of the body mass; this and opportunistic infections can generate an anemic state that is a hallmark of the battle of the organism against external agents [22, 23]. Therefore, we focused our study considering these aspects in search of cases of anemia. In the entire

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**Table 1.** Demographical and clinical features of the group of HIV+ patients

<table>
<thead>
<tr>
<th>CD4+ cell mm$^{-3}$</th>
<th>&lt;200</th>
<th>200–500</th>
<th>&gt;500</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>N</td>
<td>37</td>
<td>31</td>
<td>30</td>
<td>98</td>
</tr>
<tr>
<td>Age (years)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean±SD</td>
<td>43.8±14.5</td>
<td>50±15.9</td>
<td>43.2±10.9</td>
<td>46±14.1</td>
</tr>
<tr>
<td>Sex</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Women</td>
<td>5/37 (13.5%)</td>
<td>14/31 (45.2%)</td>
<td>7/30 (23.3%)</td>
<td>26/98 (26.5%)</td>
</tr>
<tr>
<td>Men</td>
<td>32/37 (86.5%)</td>
<td>17/31 (54.8%)</td>
<td>23/30 (76.7%)</td>
<td>72/98 (73.5%)</td>
</tr>
<tr>
<td>Blood parameters</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Range</td>
<td>5–198</td>
<td>204–472</td>
<td>505–1207</td>
<td></td>
</tr>
<tr>
<td>Mean±SD</td>
<td>100±60</td>
<td>345±39</td>
<td>737±175</td>
<td></td>
</tr>
<tr>
<td>HIV load (genome copies ml$^{-1}$)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Detectable</td>
<td>23/37 (62.2%)</td>
<td>10/31 (32.3%)</td>
<td>8/30 (26.6%)</td>
<td>42/98 (43%)</td>
</tr>
<tr>
<td>Undetectable</td>
<td>12/37 (32.4%)</td>
<td>19/31 (61.3%)</td>
<td>20/30 (66.7%)</td>
<td>50/98 (51%)</td>
</tr>
<tr>
<td>No data*</td>
<td>2/37 (5.4%)</td>
<td>2/31 (6.4%)</td>
<td>2/30 (6.7%)</td>
<td>6/98 (6%)</td>
</tr>
<tr>
<td>Range</td>
<td>82–3.3×106</td>
<td>349–2.42×105</td>
<td>43–7.3×105</td>
<td></td>
</tr>
<tr>
<td>Mean±SD</td>
<td>1.9×105±5, 8×105</td>
<td>1.3×104±4</td>
<td>1.3×104</td>
<td></td>
</tr>
<tr>
<td>Anemia</td>
<td>9/37 (24.3%)</td>
<td>2/31 (6.5%)</td>
<td>0/30 (0%)</td>
<td>11/98 (11.2%)</td>
</tr>
</tbody>
</table>

*In some patients, no quantitative PCR was done (by medical criterial) at the time of serum sample extraction for this study.

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**Fig. 1.** Hemoglobin levels and HIV genome copies ml$^{-1}$ (a) or CD4+ cell mm$^{-3}$ (b).
group of HIV+ patients, the presence of B19V had no significant association with anemia, regardless of the stage of HIV infection. Cases of AIDS patients with persistent B19V replication and severe anemia have been reported [24] and a study of AIDS patients found higher prevalence of B19V DNA compared to healthy individuals [25]. Other authors have not been able to replicate these findings [26], though the expected rate of primary infection cases in which B19V is the cause of anemia in adult patients with AIDS is relatively low [27]. All the more so considering the maximum incidence of B19V in childhood, the seroprevalence steadily increasing with age [1–4] and the existence of a single serotype of the virus which determines that the infection typically occurs only once in life [28].

There are few published surveys on B19V in adult HIV+ patients. Most publications pointing at cases of anemia due to B19V are case reports that either were published when HAART was less effective than today, or were conducted in countries where HAART is not very common. Thus, these publications may create the impression that B19V is a major risk factor for severe anemia in HIV+ patients. One well-controlled study in HIV+ children published in 2004 by LaMonte et al. [29] concluded that B19 infections could persist in children without the development of symptomatic anemia. We studied a population of adults and arrived to the conclusion that B19V infection is generally not a risk factor for adult HIV+ patients under HAART, thus supporting previous data on children and contributing significantly to the current knowledge of B19 infection.

Concerning B19V-DNA detection, all positive samples had low level B19V DNAemia and HIV-infected patients 1, 6, 9 and 11, had B19V DNA without specific IgM and/or IgG (Table 3). In this regard, patients 1 and 6 had less than 200 CD4+ T lymphocytes mm$^{-3}$. Specifically, patient #1 had 35 cells mm$^{-3}$ and patient #6 had 44 cells mm$^{-3}$. With these low cell counts, the possibility that in these two cases the immunodeficiency is responsible for the absence of detection of B19V immunoglobulins M and G is feasible. On the other hand, patients #9 and #11 had CD4+ T cell counts between 200 and 500 mm$^{-3}$. In them, an immune response should be detectable and thus in these cases the combination of immunological and virological parameters is interpreted as potential acute infections (in a phase before the presentation of clinical manifestations and the appearance of specific immunoglobulins in circulation). A similar constellation of diagnostic markers has been observed in healthy individuals also, as published before [30–32].

The concentration of B19V genome found in our positive samples was always \(\leq 10^3\) IU ml$^{-1}$ (Table 3), which demonstrates that B19V is unable to replicate at a large scale despite the condition of immunological compromise of these patients (particularly individuals with less than 200 CD4+ cells mm$^{-3}$ and high HIV load).

The frequency of anti-B19V IgM in the subgroup of HIV+ patients with less than 200 CD4+ cells mm$^{-3}$ was 10.8 % (Fig. 2b, Table 2). This matches data reported by Raguin et al. [33] but is lower than the 40 % reported by Calvet et al. [34], who studied immunosuppressed patients with heart or lung transplant. In our series, these patients may not represent cases of primary infection by B19V due to several reasons. One is that B19V primary infection main incidence occurs before 15 years old, while our study group included adults exclusively (average age above 40 years old,

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**Table 2. Markers of B19V infection in HIV+ patients**

<table>
<thead>
<tr>
<th>CD4+ cell mm$^{-3}$</th>
<th>&lt;200</th>
<th>200–500</th>
<th>&lt;500</th>
<th>(P)</th>
</tr>
</thead>
<tbody>
<tr>
<td>IgM</td>
<td>4/37 (10.8 %)</td>
<td>0/31 (0 %)</td>
<td>1/30 (3.3 %)</td>
<td>0.155</td>
</tr>
<tr>
<td>DNA</td>
<td>6/37 (16.2 %)</td>
<td>5/31 (16.1 %)</td>
<td>4/30 (13.3 %)</td>
<td>0.937</td>
</tr>
<tr>
<td>IgG</td>
<td>20/37 (54.1 %)</td>
<td>21/31 (67.7 %)</td>
<td>27/30 (90.0 %)</td>
<td>0.004</td>
</tr>
</tbody>
</table>

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**Fig. 2.** Frequency of detection of immunological and virological markers of B19V infection (a) in the control group and HIV+ patients; (b) in subgroups of HIV+ patients with different CD4+ cells mm$^{-3}$. 

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Table 3. Characteristics of HIV+ patients with B19V DNA detection

<table>
<thead>
<tr>
<th>Patient</th>
<th>Age</th>
<th>Sex</th>
<th>B19V load</th>
<th>HIV load copies ml⁻¹</th>
<th>CD4⁺ cell mm⁻³</th>
<th>B19V-IgM</th>
<th>B19V-IgG</th>
<th>Anemia</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>35</td>
<td>M</td>
<td>&lt;10³ IU ml⁻¹</td>
<td>102 800</td>
<td>&lt;200</td>
<td>–</td>
<td>–</td>
<td>+</td>
</tr>
<tr>
<td>2</td>
<td>42</td>
<td>M</td>
<td>&lt;10³ IU ml⁻¹</td>
<td>318 500</td>
<td>&lt;200</td>
<td>+</td>
<td>+</td>
<td>–</td>
</tr>
<tr>
<td>3</td>
<td>37</td>
<td>M</td>
<td>&lt;10³ IU ml⁻¹</td>
<td>772 800</td>
<td>&lt;200</td>
<td>+</td>
<td>+</td>
<td>–</td>
</tr>
<tr>
<td>4</td>
<td>51</td>
<td>M</td>
<td>&lt;10³ IU ml⁻¹</td>
<td>77 800</td>
<td>&lt;200</td>
<td>–</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>5</td>
<td>58</td>
<td>M</td>
<td>&lt;10³ IU ml⁻¹</td>
<td>1580</td>
<td>&lt;200</td>
<td>–</td>
<td>+</td>
<td>–</td>
</tr>
<tr>
<td>6</td>
<td>36</td>
<td>M</td>
<td>&lt;10³IU ml⁻¹</td>
<td>29 600</td>
<td>&lt;200</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>7</td>
<td>47</td>
<td>F</td>
<td>&lt;10³ IU ml⁻¹</td>
<td>–</td>
<td>200–500</td>
<td>–</td>
<td>+</td>
<td>–</td>
</tr>
<tr>
<td>8</td>
<td>53</td>
<td>F</td>
<td>&lt;10³ IU ml⁻¹</td>
<td>349</td>
<td>200–500</td>
<td>–</td>
<td>+</td>
<td>–</td>
</tr>
<tr>
<td>9</td>
<td>73</td>
<td>M</td>
<td>&lt;10³ IU ml⁻¹</td>
<td>15989</td>
<td>200–500</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>10</td>
<td>45</td>
<td>F</td>
<td>&lt;10³ IU ml⁻¹</td>
<td>20 100</td>
<td>200–500</td>
<td>–</td>
<td>+</td>
<td>–</td>
</tr>
<tr>
<td>11</td>
<td>68</td>
<td>M</td>
<td>&lt;10³ IU ml⁻¹</td>
<td>–</td>
<td>200–500</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>12</td>
<td>41</td>
<td>F</td>
<td>&lt;10³ IU ml⁻¹</td>
<td>–</td>
<td>&gt;500</td>
<td>–</td>
<td>+</td>
<td>–</td>
</tr>
<tr>
<td>13</td>
<td>46</td>
<td>M</td>
<td>&lt;10³ IU ml⁻¹</td>
<td>–</td>
<td>&gt;500</td>
<td>–</td>
<td>+</td>
<td>–</td>
</tr>
<tr>
<td>14</td>
<td>28</td>
<td>M</td>
<td>&lt;10³ IU ml⁻¹</td>
<td>–</td>
<td>&gt;500</td>
<td>–</td>
<td>+</td>
<td>–</td>
</tr>
<tr>
<td>15</td>
<td>25</td>
<td>M</td>
<td>&lt;10³ IU ml⁻¹</td>
<td>272 535</td>
<td>&gt;500</td>
<td>–</td>
<td>+</td>
<td>–</td>
</tr>
</tbody>
</table>

Table 1). Only two of the IgM+ patients had concomitant B19V DNA, in both cases at low concentration and with detectable specific IgG (Table 3). In addition, it has been demonstrated that the detection of certain immunological markers can be linked to the polyclonal activation that occurs in the progression of HIV infection [35–37]. A proportion of IgM detection could even be endorsed to the difficulty that still circumscribes the interpretation of the results of anti-B19V IgM tests. It has been pointed before that factors such as the lack of standardisation of the procedure and the presence of circulating antigen-antibody complexes in cases of high viral load during acute infection, may alter the outcome of IgM assay [37, 38]. A constant generation of antigen-antibody complexes is expected in HIV+ patients with detectable HIV viral load [39].

The frequency of B19V DNA in our group of HIV+ patients did not show an association with high or low viral load of the retrovirus, and rather was equivalent to the control group of this study (Fig. 2a), which included adults with an average age of 35 years old. The prevalence of DNA B19V in our control group is higher than described for asymptomatiac individuals from Chile, France, Belgium and the US [40]. However, these reports are not necessarily comparable to our study, since they were carried out using samples from blood banks. The lower prevalence in blood banks is likely because voluntary donors understand the essence of the action of donation and behave accordingly, not only attending spontaneously to donate blood but also concerning health care. Furthermore, the observed prevalence can be influenced by the sampling period. When it comprises an outbreak and the time immediately after, it effects the higher frequency detected compared to inter-epidemic periods or times immediately preceding an outbreak. Our sampling took place in 2012–2014, coinciding with a predicted epidemic period [4] which was actually corroborated by reports from different regions of the world [41, 42].

The meaning of the detection of B19V DNA in serum at low concentrations is not clear yet. It is known that cardiac tissue, among others, is a site where the virus can potentially reside for a certain amount of time [43] although given B19V tropism for the erythroid lineage, assigning such presence to this cause is not easy [44]. Due to B19V DNA detection in absence of specific antibody markers, the status of acute or persistent B19V infection in the subpopulation of DNA-positive patients may remain uncertain (especially in transversal studies like this, without follow-up samples analysed). However, because of the low level DNAemia chronically persistent B19V infection is probable. In contrast to acute B19V infection or to persistent infection associated to high levels of DNAemia, values below 10⁷ IU ml⁻¹ have only rarely been associated to anemia [43, 45]. It has been described that individuals who cannot mount an immune response or have hematological disorders can develop anemia. In immunosuppressed patients, the lack of an adequate immune response that eliminates the virus leads to a persistent infection and the continuous replication of the virus is associated with chronic anemia [46]. In the presence of hematological diseases involving an increased rate of erythrocyte turnover, massive infection of nucleated precursor cells occurs and high-titer replication may result in bone marrow suppression, triggering a life-threatening drop of haemoglobin values [47]. In this context, subjects of our study population in whom B19V DNA was detected seem to fit in the general framework of the balance between B19V infection and clinical manifestations.

Concerning the prevalence of anti-B19V IgG in HIV+ patients, in the whole group the frequency was 68.4% (Fig. 2a) and it increased with the number of CD4⁺ lymphocytes (Fig. 2b, Table 2). These results contrast those published by Azadmanesh et al. [48] and are partially in agreement with the findings by Abdollahi et al. [49]. Azadmanesh et al. [48] reported a prevalence of only 11.1% and the frequency
was higher in patients with lower numbers of CD4+ cells, while Abdollahi et al. [49] reported a prevalence of 81.1 %, which was higher in patients with less than 200 CD4+ cells mm⁻³. It is worthy to note that the average age of the study populations alluded in these two Iranian studies was comparable and both were conducted in the same year and geographical region using similar methodology and reagents. This evidences the complexity around performing and interpreting results of serology tests for B19V, to which we referred above. The significantly lower frequency of IgG in the group of patients with less than 200 CD4+ cell mm⁻³ compared to the group with CD4 cell count above 500 mm⁻³ observed in our study could have an explanation in the decreased specific humoral immunity that is known to occur in the later stages of HIV infection. This fact does not contradict the higher levels of total immunoglobulins typical of late stages of HIV infection, since it is a consequence of polyclonal activation. When CD4+ T lymphocyte count decreases, memory B cells also diminish; then, as the infection progresses and CD4+ cell count falls below 200, the individual begins to lose the specific humoral immune response [50–52]. The average count of CD4+ lymphocytes in the most immunocompromised subgroup of our study population is 100 cell mm⁻³ (Table 1), therefore, it can be inferred that a substantial proportion of patients cluster in this situation. Accordingly, our findings in this regard are consistent with those of Bucher Praz et al. [53], who indeed reported the presence of B19V DNA in the absence of specific IgG in a patient with HIV and 75 CD4+ cells mm⁻³. Watanabe et al. [54] described similar results and even more, anti-B19V IgG turned detectable after initiating HAART therapy. The subgroup of 200–500 CD4+ cell mm⁻³ presumably represent a combination of patients who started treatment before AIDS stage and have not had a significant decrease in their memory B cells, on the one hand, and, on the other hand, patients who reached AIDS stage, responded well to treatment and their cell counts are recovering. Finally, the group of patients with CD4+ cell count above 500 mm⁻³, with a higher prevalence of anti-B19V IgG compared to the control group, would have the expected prevalence among immunocompetent individuals plus the contribution of the polyclonal stimulation already described.

In conclusion, our results indicate that B19V prevalence in HIV+ adult patients is not significantly different from HIV-seronegative adults and it is not presumed to be an important cause of anemia among HIV+ patients receiving HAART. Further studies will contribute to elucidate the mechanisms and significance of B19V DNA prevalence/persistence (albeit at low concentration) in adults, independently of their CD4+ cell status. Efforts should focus in standardising serological tests for B19V, since at present similar study designs may deliver widely varying results.

Conflicts of interest
The authors declare that there are no conflicts of interest.

Ethical statement
An independent Ethics Committee (UNC-06200846379) approved the study. The authors assert that all procedures contributing to this work comply with the ethical standards of the relevant national and institutional committees on human experimentation and with the Helsinki Declaration of 1975, as revised in 2008.

References

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