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High prevalence of parvovirus B19 infection in patients with chronic kidney disease under hemodialysis: A multicenter study

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ABSTRACT

Objectives: Parvovirus B19 (B19V) infection is commonly acute and self-limited, but in chronic kidney disease (CKD) patients under dialysis treatment, this infection could increase susceptibility to acute and chronic anemia. The aim of this study was to evaluate the frequency and risk of B19V infection among Brazilian CKD patients under dialysis.
Methods: A study was conducted among 221 CKD patients and a control group of 142 blood donors. B19V infection was evaluated in serum samples by real-time PCR, and ELISA (anti-B19V IgM and IgG).
Results: B19V DNA was detected in 65% (145/221) of CKD patients, which was significantly higher ($p < 0.001$) than in the blood donors (6.3%). Simultaneous detection of B19V IgG and viremia was shown in 40.3% of CKD patients, which was indicative of persistent B19V infection. CKD patients showed an increased risk of developing B19V infection (OR = 28.1, CI = 13.5–58.5, $p = 0.001$).
Conclusions: Despite an absence of clinical signs of B19V infection, these data highlight the importance of B19V infection in this high-risk population, since a persistent B19V infection could become clinically significant after renal transplant. Moreover, the persistent viremia should be considered as a potential risk, mainly because of the contamination of dialysis equipment.

Introduction

Parvovirus B19 (B19V), a member of *Parvoviridae* family and *Erythroparvovirus* genus, is a small, non-enveloped, icosahedral, ssDNA virus that infects only humans (Cotmore et al., 2019).

B19V is mainly transmitted in close contact, by the inhalation of viral particles in aerosol (i.e. respiratory spread transmission). Transmission may also occur through blood-donor transfusion and transplantation, mostly during the viremia that precedes the seroconversion period, as well as vertically, from mother to fetus (Gallinella, 2018; Ozeki et al., 2006).

Common B19V-related diseases are associated with age, and the hematological and immunological status of the host. In immunocompetent individuals, B19V infection is commonly acute, benign, and self-limited by the appearance of specific IgM and IgG antibodies, leading to a rapid and complete clearance of viremia (Adamson-Small et al., 2014). It usually causes a common childhood exanthematic disease known as erythema infectiosum (EI), arthropathy in adults, and aplastic crisis in patients with hemolytic diseases, with an extremely low mortality rate. In immunocompromised patients, B19V infection is not readily cleared and its long persistence leads to chronic anemia, which can be severe due to the notable tropism of B19V to human bone marrow and erythroid precursor cells for replication (Qiu et al., 2017).

However, B19V infection may persist in some individuals, regardless of their immunocompromised level and hematological status. This persistent infection could affect erythroid and non-erythroid tissue, such as bone marrow, myocardium, liver, skin, and kidney. The clinical significance of this delayed clearance and low-level viremia is often unknown and debatable (Adamson-Small et al., 2014).

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Chronic kidney disease (CKD) is a condition arising from various diseases, including diabetes mellitus and hypertension, which leads to a gradual loss in the rate of glomerular filtration. In patients with CKD, the treatments of choice are, in general, dialysis and kidney transplantation. Infectious diseases are the second major cause of death in CKD patients (Naqvi and Collins, 2006), and patients undergoing dialysis therapy may have increased susceptibility to many viral infections, including B19V infection, that may result in an increase in acute or chronic anemia (Webster et al., 2017).

Anemia is a common complication of CKD due to: reduced production of erythropoietin by the kidney; significantly decreased erythrocyte survival and iron deficiency; chronic uremic condition; and mineral bone disease caused by distrupted vitamin D, calcium, and phosphate metabolism (Guiserix et al., 1996; Duranay et al., 1998). Persistent B19V infection in these dialyzed patients after renal transplantation and immunosuppressive therapy can manifest as transient aplastic crisis or chronic anemia (Adamson-Small et al., 2014; Eid et al., 2013; Söderlund-Venermo et al., 2002; Norja et al., 2006; Waldman and Kopp, 2007). Nonetheless, there is a lack of information regarding B19V infection among Brazilian patients under dialysis. The aim of this study was to determine the prevalence of B19V viremia and to evaluate the risk of B19V infection among dialyzed CKD patients.

Methods

Study population and ethical aspects

This was a cross-sectional study that investigated parvovirus B19 infection in archived serum samples, collected during 2013–2015, from a total of 221 CKD patients who were under dialysis treatment in three different Brazilian dialysis units: unit 1 (32 patients), located in the Central region of Rio de Janeiro (Southeast Brazil), collected during January to February, 2013; unit 2 (61 patients), located in the North region of Rio de Janeiro, collected during June to July, 2014; unit 3 (128 patients), located in Ceará State (Northeast Brazil), collected during July to August, 2015. Serum samples with insufficient volume to carry out molecular and serological tests were excluded. In addition, serum samples from blood donors (n = 142), collected in Rio de Janeiro during 2015, were included as a control group.

An epidemiological questionnaire was provided to collect demographic, clinical, and hematological data from these patients, such as age, gender, *per capita* income, race, marital status, educational level, dialysis duration, history of hepatitis, blood transfusion, and anemia.

Anemia was defined according to the World Health Organization (WHO) criteria, as a hemoglobin (Hb) concentration below 13.0 g/dL for adult males and postmenopausal women, and below 12.0 g/dL for premenopausal women; severe anemia was defined as an Hb concentration below 7 g/dL (WHO, 2011).

All sera samples had been previously tested for hepatitis B surface antigen (HBsAg), hepatitis B core antibody (anti-HBc), hepatitis B surface antibody (anti-HBs), and hepatitis C antibody (anti-HCV), detected by enzyme-linked immunosorbent assay (ELISA) (Euroimmun, Germany), as previously reported (Ribeiro Barbosa et al., 2017).

Molecular and serological tests for B19V

B19V infection was investigated in the serum samples collected on the day of dialysis procedure, after signed consent by the patient, using serological and molecular assays.

For molecular detection and quantification of B19V, DNA was extracted from sera samples using a QIAamp DNA Blood Mini Kit (Qiagen, Hilden, Germany). Real-time PCR was carried out (Alves et al., 2019) using the TaqMan system (Applied Biosystems 7500 Real-Time PCR System, Applied Biosystems, USA). For absolute quantification, a synthetic standard curve of the B19V NS1 region (IDT, USA) was designed (nt 1905–1987, GenBank: NC_000883.2). Primers for the NS1 region (nt 1905F and 1987R) and a single labelled 5′ FAM probe (nt 1925–1948, GenBank: NC_000883.2) were used.

All serum samples from CKD patients were investigated for the presence of anti-B19V IgM and IgG antibodies by ELISA (VirionSerion, Germany), according to the manufacturer's instructions.

For B19V genotyping, a nested PCR (Abe et al., 2007) using the primer pairs VP1/VP1R (nt 3502–3883, GenBank: NC_000883.2) and VP2/VP2R (nt 3541–3867, GenBank: NC_000883.2) were performed to amplify the 326bp fragment of the VP1/VP2 capsid gene. The nested-PCR products were purified using the QIAquick gel extraction (Qiagen, Germany) and then sequenced directly using the BigDye terminator v. 1.1 cycle sequencing kit (Applied Biosystems, USA) and an ABI Prism® 3730 DNA analyzer (Applied Biosystems, USA). Both strands of each amplicon were sequenced.

For phylogenetic analysis, sequences were aligned using the BioEdit Sequence Alignment Editor v7.2.5. A neighbor-joining tree was constructed based on Kimura-2 parameter distances available in MEGA v.10.0 software (Kumar et al., 2018). The B19V genotype was determined by including reference sequences for genotypes IA, IB, II, IIIA, and IIIB, available in Genbank.

Statistical analysis

Statistical analysis was conducted using R studio (version 1.2.1335, Boston, USA). Continuous variables were expressed as means and were compared using the Mann–Whitney U test Categorical variables were expressed as number (%) and were identified according to: age (<52 and ≥52 years), gender (male or female), anemia, mean time on dialysis, HBV/HCV infection, blood transfusion, and kidney transplant. The chi–square test or Fisher exact test was used for measurement of categorical variables. The odds ratio (OR) for B19V DNA detection in CKD patients was analyzed using univariate logistic regression. All p-values were two-sided, with those <0.05 considered as statistically significant.

Results

Characterization of the studied population

A total of 221 CKD patients under dialysis were enrolled in the study. The mean age was 51 ± 14.1 years and 61.5% (136/221) were male (Table 1). In dialysis units 1, 2, and 3 were enrolled 32 patients (mean age 49 ± 11.8 years; 50% male), 61 patients (mean age 54 ± 15.2 years; 54% male), and 128 patients (mean age 50 ± 13.8 years; 68% male), respectively. The mean age of the blood donors' group was 35 ± 12.6 years, with 55% male.

The mean duration of dialysis was 70.2 ± 75.5 months and the dialysis interval was three times a week (Table 1). According to the dialysis units, the mean durations of dialysis were 72.8 ± 73.4 months in unit 1; 36.4 ± 40.4 months in unit 2; and 85.9 ± 83.8 months in unit 3. The difference between units 2 and 3 was statistically significant ($p < 0.001$; Figure 1).

Previous blood transfusions and kidney transplantations were reported by 59.7% (132/221) and 9.9% (22/221) of the patients, respectively. Among the kidney transplant recipients, two were from unit 1, two from unit 2, and the other 18 were from unit 3. Anemia and HBV/HCV infection were present in 32% (71/221) and 45.7% (101/221) of the individuals, respectively (Table 1).

Table 1
Demographic, clinical, and laboratory findings among CKD patients attending three different Brazilian dialysis units during 2013–2015.

Parameter	Total*n* (%)	B19V+*n* (%)	B19V–*n* (%)	*p*-value[a]	OR (95% CI)
Gender					
Male	136 (61.5)	94 (69.1)	42 (30.9)	0.24	1.22
Female	79 (35.8)	51 (64.5)	28 (35.5)		(0.68–2.21)
Unknown	6 (2.7)	0 (0)	6 (100)		
Age group (years)					
<52	105 (47.5)	74 (70.5)	31 (29.5)	0.17	1.31
≥52	110 (49.7)	71 (64.5)	39 (35.5)		(0.73–2.33)
Unknown	6 (2.8)	0 (0)	6 (100)		
Per capita income					
$110–$180	62 (28.1)	42 (67.7)	20 (32.3)	0.38	0.76
$181–$360	57 (25.8)	36 (63.2)	21 (36.8)		(0.28–2.01)
$361–$540	28 (12.7)	20 (71.4)	8 (28.6)		
$541–$720	14 (6.2)	12 (85.7)	2 (14.3)		
≥$721	30 (13.6)	22 (73.3)	8 (26.7)		
Unknown	30 (13.6)	13 (43.3)	17 (56.7)		
Race/ethnicity					
White	55 (24.9)	37 (67.3)	18 (32.7)	0.47	0.98
Black	146 (66.1)	99 (67.8)	47 (32.2)		(0.50–1.89)
Asian	5 (2.3)	5 (100)	0 (0)		
Unknown	15 (6.7)	4 (26.6)	11 (73.4)		
Marital status					
Unmarried	61 (27.6)	41 (67.2)	20 (32.8)	0.32	0.80
Married[b]	110 (49.8)	79 (71.8)	31 (28.2)		(0.41–1.59)
Divorced	26 (11.8)	17 (65.4)	9 (34.6)		
Widowed	14 (6.3)	8 (57.1)	6 (42.9)		
Unknown	10 (4.5)	0 (0)	10 (100)		
Educational level					
Non-literate	15 (6.8)	11 (73.3)	4 (26.7)	0.12	0.41
Primary	69 (31.2)	43 (62.3)	26 (37.7)		(0.13–1.37)
Secondary	40 (18.1)	26 (65.0)	14 (35.0)		
Higher secondary	54 (24.4)	39 (72.2)	15 (27.8)		
Graduate	20 (9.1)	16 (80.0)	4 (20.0)		
Postgraduate	11 (5.0)	9 (81.8)	2 (18.2)		
Unknown	12 (5.4)	1 (8.3)	11 (91.7)		
Time of dialysis (months)					
<48	103 (46.6)	67 (65.0)	36 (35.0)	0.26	0.71
≥48	108 (48.9)	78 (72.2)	30 (27.8)		(0.39–1.28)
Unknown	10 (4.5)	0 (0)	10 (100)		
Hemoglobin level (g/dL)	11.53	11.60	11.87	0.85	1.23 (0.54–2.94)
Anemia					
Yes	71 (32.1)	59 (83.1)	12 (16.9)	0.83	1.11
No	49 (22.2)	40 (81.6)	9 (18.4)		(0.42–2.86)
Unknown	101 (45.7)	46 (45.5)	55 (54.5)		
HBV/HCV infection					
Presence	101 (45.7)	63 (62.4)	38 (37.6)	0.28	0.73
Absence	120 (54.3)	83 (69.2)	37 (30.8)		(0.42–1.29)
HBV/HCV infection and anemia					
Anemia	37 (36.6)	30 (81.1)	7 (18.9)	0.003	4.02
No anemia	64 (63.4)	33 (51.6)	31 (48.4)		(1.54–10.49)
Blood transfusion					
Presence	132 (59.7)	88 (66.6)	44 (33.4)	0.29	0.71
Absence	76 (34.4)	56 (73.7)	20 (26.3)		(0.38–1.34)
Unknown	13 (5.9)	1 (7.7)	12 (92.3)		
Blood transfusion and anemia					
Anemia	47 (35.6)	40 (85.1)	7 (14.9)	0.24	2.00
No anemia	27 (20.5)	20 (74.1)	7 (25.9)		(0.61–6.49)
Unknown	58 (43.9)	28 (48.3)	30 (51.7)		
Kidney transplant					
Presence	22 (9.9)	19 (86.4)	3 (13.6)	0.06	3.19
Absence	188 (85.1)	125 (56.6)	63 (43.4)		(0.91–11.19)
Unknown	11 (5.0)	1 (9.1)	10 (90.9)		

Table 1 (*Continued*)

Parameter	Total*n* (%)	B19V+*n* (%)	B19V–*n* (%)	*p*-value[a]	OR (95% CI)
Kidney transplant and anemia					
Anemia	8 (36.4)	8 (100)	0 (0)	0.28	5.66
No anemia	9 (41.0)	7 (77.8)	2 (22.2)		(0.23–137.8)
Unknown	5 (22.6)	4 (80.0)	1 (20.0)		

n: number; B19V: parvovirus B19; OR: odds-ratio; 95% CI: 95% confidence interval; HBV: hepatitis B virus; HCV: hepatitis C virus.
[a] *p*-value relating to the Pearson χ^2 test.
[b] Stable unions included as married.

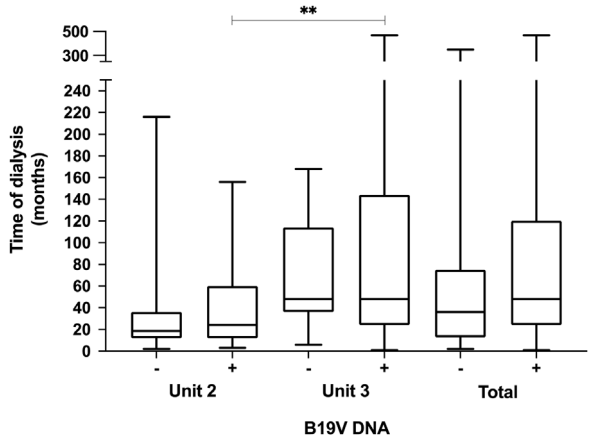

Figure 1. Time of dialysis among CKD patients between the units studied. Bars represent the standard deviation. Selections with **($p < 0.001$) represent significant differences between groups. Results analyzed with non-parametric Mann–Whitney test.

Molecular and serological B19V markers

B19V DNA was often detected in CKD patients (65.6%). Of these, one (3.1%) was reported in unit 1, 37 (60.6%) in unit 2, and 107 (83.6%) in unit 3. In all dialysis units, the mean viral load detected was 10^4 IU/mL (Table 2).

Anti-B19V IgM and IgG were investigated among the CKD patients (n = 221). Anti-B19V IgG prevalence was 79.2% (175/221). According to the dialysis units, anti-B19V IgG prevalence was 78.1% (25/32) in unit 1, 81.9% (50/61) in unit 2, and 78.1% (100/128) in unit 3 (Table 2). Anti-B19V IgM was detected in 15.4% (34/221) of patients: 8.2% (5/61) in unit 2 and 22.6% (29/128) in unit 3 (Table 2). No CKD patient from unit 1 had anti-B19V IgM.

As shown in Table 3, 28.5% (n = 63) of the patients presented acute infection (B19V DNA+ and/or IgM+). However, only in 12.2% (n = 27) was B19V DNA simultaneously detected with anti-B19V IgM. Anti-B19V IgG was found in conjunction with B19V DNA in 40.2% (n = 89) of the patients, suggesting persistent infection among them. According to the dialysis units, unit 3 showed the highest number of acute (n = 52) and persistent (n = 62) infections, while unit 1 observed more patients with prior B19V exposure (n = 24) and patients susceptible to B19V infection (n = 7).

Nine blood donors (6.3%) tested positive for B19V DNA. Most of them were male (55.5%), with an average age of 42 ± 2.6 years. B19V DNA detection was significantly higher ($p < 0.001$) in the CKD patients under dialysis (65%) than in the blood donors (6.3%). A significant association was observed between CKD patients under hemodialysis and B19V infection when compared with blood donors (OR = 28.1, CI = 13.5–58.5, p = 0.001).

Nucleotide sequences were obtained successfully from only two B19V isolates from dialysis unit 3. Phylogenetic analysis revealed that these two B19V isolates were classified as genotype 1A (Figure 2). The nucleotide sequences obtained during this study were deposited in the GenBank database under accession numbers MT534175 and MT534176.

Clinical manifestations and biochemical analysis among CKD patients

According to Table 1, 71 of the 221 CKD patients presented with anemia (32.1%), with a mean hemoglobin level of 11.2 ± 1.7 g/dL. Among the anemic patients, 83.1% (59/99) had B19V DNA. Severe anemia (Hb < 7.0 g/dL) was registered in three patients (1.4%), and all of these had B19V DNA and anti-B19V IgG.

Among the CKD patients, 101 (45.7%) presented with infection with hepatitis B virus (HBV) and/or hepatitis C virus (HCV). Most of these HBV/HCV-infected patients (62.4%) also presented with B19V infection. Coinfection of HBV/HCV with B19V increased the occurrence of anemia (OR = 4.02, CI = 1.54–10.49, p = 0.003) (Table 1).

Blood transfusion and kidney transplant were reported by 59.7% (132/221) and 9.9% (22/221) of patients, respectively. B19V DNA was detected in all kidney transplant patients with anemia (n = 8).

There was no significant relationship between the presence of B19V DNA and anemia, mean duration of dialysis, blood transfusion, renal transplant, age, or gender ($p > 0.05$; Table 1).

Discussion

This was the first study to evaluate whether CKD patients undergoing dialysis are at high risk of B19V infection in Brazil. A high prevalence of B19V DNA was found among CKD patients (65%), particularly in unit 3. This was probably due to the large proportion of long-term dialysis patients compared with other units, which was significantly greater than in unit 2 ($p > 0.001$). A significant correlation between length of time on dialysis and

Table 2
Molecular and serological B19V markers among CKD patients attending three different Brazilian dialysis units during 2013–2015.

Study group	B19V DNA *n* (%)	Viral load (IU/mL)	Anti-B19V IgM *n* (%)	Anti-B19V IgG*n* (%)
Unit 1 (n = 32)	1 (3.1)	4.0×10^4	0 (0)	25 (78.1)
Unit 2 (n = 61)	37 (60.6)	3.8×10^4	5 (8.2)	50 (81.9)
Unit 3 (n = 128)	107 (83.6)	4.1×10^4	29 (22.6)	100 (78.1)
Total (n = 221)	145 (65.6)	4.0×10^4	34 (15.4)	175 (79.2)

B19V: parvovirus B19; qPCR: real-time quantitative PCR; IU/mL: international units per mL; PCR: polymerase chain reaction; IgM: immunoglobulin M; IgG: immunoglobulin G.

Table 3
B19V biomarkers among CKD patients attending three different Brazilian dialysis units during 2013–2015.

B19V biomarkers			Unit 1 *n* (%)	Unit 2*n* (%)	Unit 3*n* (%)	Total*n* (%)
DNA+	IgM+	IgG+	0 (0)	5 (8.2)	20 (15.6)	25 (11.3)
DNA+	IgM+	IgG−	0 (0)	0 (0)	2 (1.6)	2 (0.9)
DNA−	IgM+	IgG+	0 (0)	0 (0)	7 (5.5)	7 (3.2)
DNA+	IgM−	IgG−	0 (0)	6 (9.8)	23 (17.9)	29 (13.1)
DNA+	IgM−	IgG+	1 (3.1)	26 (42.6)	62 (48.5)	89 (40.3)
DNA−	IgM−	IgG+	24 (75.0)	19 (31.2)	11 (8.6)	54 (24.4)
DNA−	IgM−	IgG−	7 (21.9)	5 (8.2)	3 (2.3)	15 (6.8)
Total			32 (100)	61 (100)	128 (100)	221 (100)

B19V: parvovirus B19; IgM: immunoglobulin M; IgG: immunoglobulin G.

detection of B19V DNA has been reported in dialyzed patients from Iraq (Kadhom and Hussein, 2018).

Another possible explanation for the lower B19V DNA prevalence in unit 1 (1%), compared with units 2 and 3, may be the period over which samples were collected. Samples from unit 1 were collected during January to February 2013, while those from units 2 and 3 were collected during 2014 and 2015, respectively. Previous studies conducted in Brazil have demonstrated that B19V has exhibited a cyclical pattern of occurrence every 4–5 years, characterized by years of high levels of infection, followed by a period of low incidence. During epidemic periods, the wide range of clinical conditions associated with B19V infection may be diagnosed. Furthermore, the seasonal pattern of the infection in the northeast and southeast regions of the country showed a peak occurrence of B19V cases in the second half of the year–late winter and spring–with sporadic cases during the first half of the year (Cubel et al., 1996; Cubel et al., 1997; Cubel Garcia et al., 2017; Oliveira et al., 2008; Oliveira et al., 2002).

Two outbreaks of exanthematous illness and a case of fetal loss due to B19V infection were reported in Brazil during 2014–2015 (Di Paola et al., 2019; Figueiredo et al., 2019; Oliveira et al., 2019), coinciding with the period of sample collection at units 2 and 3 of this study. These findings could explain the high prevalence of B19V viremia found among these CKD patients. There is no epidemiological register of B19V outbreaks in the states of Rio de Janeiro and Ceará at the time of the study, because notification of B19V infection is not compulsory for epidemiological surveillance. However, the finding of B19V DNA in asymptomatic blood donors during 2015 in Rio de Janeiro corroborates the finding that 2015 was an epidemic year.

Few studies have been carried out worldwide on the detection of B19V DNA in patients under dialysis; most surveys have been focused on the prevalence of B19V in kidney transplant recipients (Rosado-Canto et al., 2019; Hai An et al., 2019; Baek et al., 2017).

Previous studies conducted in Iran found B19V DNA in 10% (5/50) of CKD patients (Sharif et al., 2016) and in 13.1% (13/99) of those infected with HIV (Azadmanesh et al., 2015). In a single Brazilian

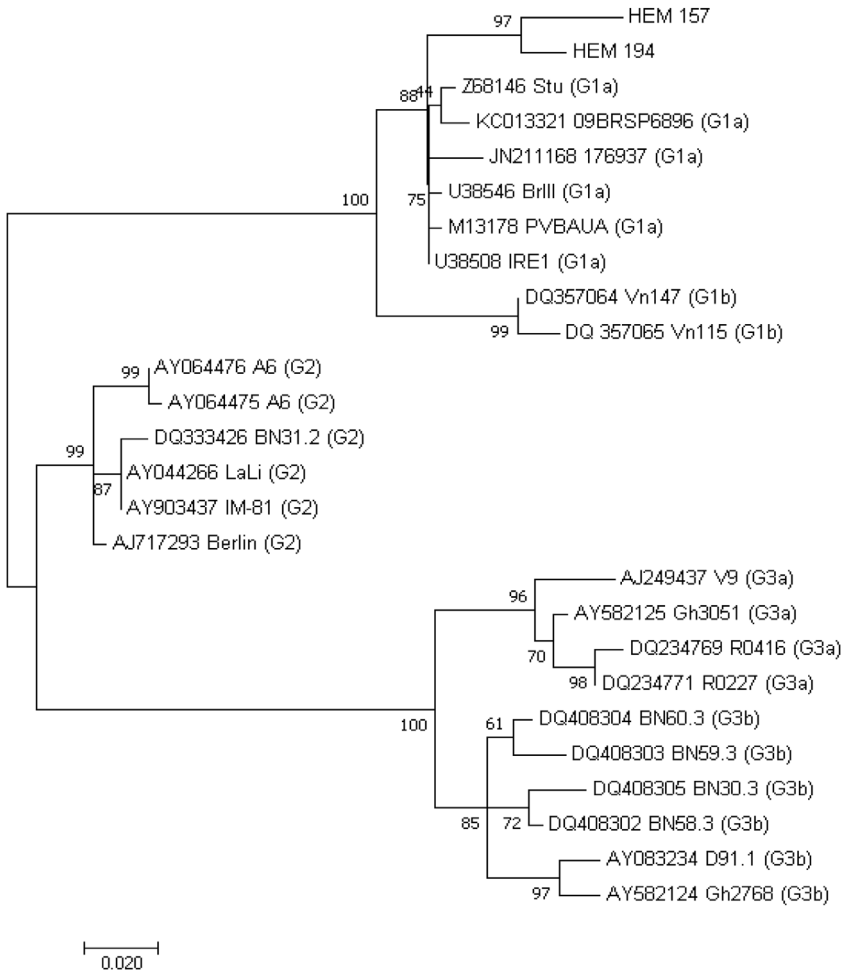

Figure 2. Evolutionary history of B19V genotypes inferred by using the Maximum Likelihood method and Tamura-Nei + I model. All analysis were conducted in Mega X. The samples of this study were named by HEM prefix.

study, carried out in Minas Gerais state during 2012, B19V DNA was found in only 10 of 120 CKD patients (8.1%) (Alves et al., 2014). The higher prevalence of viremic patients in our study could be explained by the difference in sensitivity of the assays used to detect B19V DNA–quantitative PCR over conventional nested PCR. Another point to consider is that the earlier study was probably conducted in a period that did not include an epidemic year for B19V (Furtado et al., 2016).

Phylogenetic analysis revealed that two samples from unit 3 were clustered to genotype 1 and classified as genotype 1A. Genotype 1 is the most common in Brazil, with a frequency of 87.9% according to previous studies (Cubel Garcia et al., 2017; Di Paola et al., 2019; Figueiredo et al., 2019; Freitas et al., 2008; Pereira et al., 2014).

The mean viral load detected in CKD patients from all dialysis units was 10^4 IU/mL. As reported in previous studies, this mean viral load could be linked to a risk of B19V transmission through blood components (Molenaar-de Backer et al., 2016a; Molenaar-de Backer et al., 2016b; use Cfmpfh, 2012).

Among CKD patients who tested positive for B19V DNA (n = 145) there were cases of acute infection, with concomitant detection of anti-B19V IgM in 27 patients, and no anti-B19V antibodies detected in 29 patients. This finding highlights the importance of combining serological and molecular assays for the accurate diagnosis of acute B19V infection. On the other hand, most patients with detectable B19V DNA were also positive for anti-B19 IgG (61.6%), suggesting persistent-B19V infection. This was probably due to a qualitative defect of antibodies in neutralizing the virus, since CKD patients usually have some degree of immunocompromized status due to their chronic uremic condition (Vaziri et al., 2012).

Persistent-B19V infection is an important finding in the pathogenesis of this virus, because many people may harbor the virus asymptomatically, but could pose an infection risk for certain groups, such as immunocompromized patients. Persistent infection is characterized by the presence of low viral load ($\leq 10^4$ IU/mL) associated with the presence of anti-B19V IgG antibodies. Studies correlating B19V DNA, RNA, and proteins detected with some diseases–such as hepatitis, glomerulonephritis, and myocarditis–are limited and often contradictory (Adamson-Small et al., 2014; Söderlund-Venermo et al., 2002; Norja et al., 2006). However, the clinical significance of B19V infection in patients undergoing dialysis should be considered a potential risk in terms of the contamination of dialysis equipment and the subsequent threat to dialysis patients during transplantation procedures.

Two hypotheses are commonly used to explain persistent B19V infection: the persistence of the virus in different tissues, with prolonged release of virions, or at least B19V DNA; and the lack of neutralization of virions by the anti-B19V IgG antibody. The latter hypothesis was overturned by studies that demonstrated high levels of avidity and neutralization in antibodies directed against epitopes of the viral VP1 protein (Hourfar et al., 2011; Juhl et al., 2014). In addition, some data have demonstrated that prolonged persistence of B19V DNA would be the norm rather than the exception (Dodd, 2011).

The impact and role of parvovirus B19 infection in patients with chronic kidney disease are not known. Nevertheless, there are several reasons to think that parvovirus may be an important pathogen in these populations, since anemia is a common consequence of B19V infection and is also a predictable consequence of chronic renal insufficiency (Kazmi et al., 2001). Although the prevalence of severe anemia is high at the onset of dialysis, and the early presence of anemia in end-stage renal disease (ESRD) is associated with a greater rate of subsequent hospitalization and mortality (Ma et al., 1999), many studies have not found a significant association between anemia and B19V DNA detection in dialysis patients (Guiserix et al., 1996; Sharif et al., 2016; Alves et al., 2014).

In our study, although anemia was found in 32% of CKD patients, we did not find a significant relationship between the presence of B19V viremia and anemia. However, anemia was significantly associated with B19V viremia in patients coinfected with HBV or HCV (OR = 4.02, CI = 1.54–10.49, p = 0.003), suggesting that coinfection of HBV/HCV with B19V might increase the risk of anemia. Recently, our group reported that coinfection of B19V and HBV increases the severity of disease (Alves et al., 2020). In addition, our experimental study confirmed erythroid hypoplasia in bone marrow from B19V and HAV coinfected cynomolgus monkeys (*Macacca fascicularis*) (Leon et al., 2016). Therefore, the frequency of persistent B19V viremia with chronic anemia needs to be systematically evaluated in a larger cohort of anemic dialysis patients.

Our study found no correlation between time on dialysis, blood transfusion, age, or gender and B19V DNA detection. Although B19V DNA detection was higher in kidney-transplanted than in non-transplanted patients, this difference was not statistically significant. A possible explanation for higher detection of B19V DNA among kidney transplanted patients is that, after kidney transplantation, patients are required to take immunosuppressant medications to minimize the chance of organ rejection; this must continue for their entire lifetime. These medications can have significant side-effects, including increased risk of acquiring infectious diseases, such as parvovirus B19.

B19V DNA prevalence was significantly higher ($p < 0.001$) in patients under dialysis (65%) when compared with blood donors. Univariate logistic regression analyses demonstrated that being a CKD patient under dialysis was a risk factor for B19V infection. CKD patients undergoing dialysis therapy may have increased susceptibility to B19V infection for a range of reasons: their characteristic immune deficiency condition; routine exposure to dialysis procedures, which are invasive and therefore a potential risk for parenterally transmitted infections; and also the frequency of blood transfusion, which considering the mean viral load of 10^4 IU/mL observed in this study, could present a potential risk of transfusion-transmitted B19V (Waldman and Kopp, 2007; Waldman et al., 2008). Other studies on B19V and hemodialytic patients do not make the comparison with blood donors.

These data reinforce the importance of investigating B19V infection in this high-risk group, since persistent B19V infection in CKD patients could become clinically significant after renal transplant and after the introduction of immunosuppressant therapy. Moreover, persistent viremia should be considered a potential risk of spread of infection in enclosed dialysis units.

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Conflicts of interest

The authors have no relevant affiliations or financial involvements with any organization or entity with a financial interest in, or in financial conflict with, the subject matter or materials discussed in the manuscript, aside from those disclosed.

Ethical approval

The study protocol was approved by the ethics committee of the Oswaldo Cruz Institute (Protocol # 3.627.364).

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