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Visceral Leishmaniasis in Renal Transplant Recipients: Study of 30 Cases

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Abstract

Introduction: Visceral leishmaniasis is a disease caused by the protozoan *Leishmania sp*. and is transmitted by *Lutzomyia longipalpis* (sand fly). In renal transplant recipients, Visceral Leishmaniasis causes severe damage to the liver, spleen, and hematopoietic system as well as poor outcomes for patients and transplanted kidneys. This study describes the largest series of cases of this disease in renal transplant recipients, providing important information about the diagnostic routines and therapeutic strategies in this patient population.

Methods: A retrospective, descriptive study was performed to analyze the distribution and evaluate the extent of the epidemiologic, clinical, diagnostic, and therapeutic aspects of 30 renal transplant recipients from endemic regions who presented with Visceral Leishmaniasis in the post-transplantation period.

Results: In this study, Visceral Leishmaniasis was more frequent in men (80%); the mean age of presentation was 40±10.5 years. The majority (66.7%) of patients worked in urban areas. Most of the patients (90%) cohabitated with domestic animals and were from low-income households. In 73.3% of cases, diagnosis was made by direct isolation of *Leishmania* forms. The drug chosen for treatment was liposomal amphotericin, resulting in a high degree of disease remission (80%).

Conclusion: This study describes the largest series of Visceral Leishmaniasis in renal transplant recipients and expands clinical-epidemiological knowledge for transplantation teams to perform adequate disease management for this specific patient population.

Keywords: Visceral Leishmaniasis, renal transplant, *Leishmania*

Introduction

Visceral Leishmaniasis (VL) is an opportunistic disease caused by a protozoan of the genus *Leishmania sp* [1, 2]. The LV has endemic characteristics to all continents and is considered by specialists it to be a neglected disease. There are more than 5 million cases annually and the disease has a high incidence among the low socioeconomic, immunosuppressed, and malnourished population [2,3]. In the Brazil, the incidence of VL is 2 cases per 100,000 inhabitants and in the Northeast of Brazil, where the states of Piauí are located and Maranhão this incidence raises to the Northeast to 4 cases per 100.000 inhabitants. Over the past decade, the average annual cases in the country were 3,156 cases; with 66% of all cases occur in the Northeast and 48% in the states of Piauí and Ceará. The VL was more common in children (54.4%) patients and in the masculine sex (60%) [4].

It is commonly known as a disease itself area dry climate with annual rainfall less than 800 mm, and physiographic environment composed of valleys and mountains, where the so-called "gullies" and "saw-feet." However, with urbanization of VL, especially in the peripheries of large urban centers, there known areas of land in different regions and coasts in the northeast [5].

In recent years, kidney transplantation as the best renal replacement therapy [6], has gone from a procedure performed only in medical-academic centers to a therapy performed in poorer regions as well; this is associated with the standardization of surgical techniques and the ease of access to immunosuppressors [7,8]. However, the relevance of problems due to acute rejection and surgical complications has decreased in these regions. Meanwhile, endemic and opportunistic infections such as VL have become the major preoccupation of transplantation teams, because these infections are directly associated with both graft dysfunction and the survival of renal transplant recipients [9].

There are currently no clinical protocols for the diagnosis or treatment of VL-infected renal transplant recipients, and few studies have focused on the epidemiology and risk factors associated with the disease in this specific patient population [10]. This study constitutes the largest series of this patient population (n=30) ever reported in the literature and aims to not only highlight the epidemiologic, clinical, diagnostic, and therapeutic aspects of VL in renal transplant recipients, but also contribute to the establishment of better practices in the clinical management of these patients, with the possibility of improving patient survival and reducing graft rejection.

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Patients and Methods

Study sample and location

Thirty kidney recipients between January 1989 and January 2013 were studied. Patients resided and were followed-up postoperatively in transplant centers in regions where VL is endemic. The study location was the states of Piauí and Ceará, which are located in the Southern Hemisphere in Northeastern Brazil [11].

Study design

This is a descriptive, retrospective study showing the relative distribution of renal transplant recipients with post-transplant VL, focusing on epidemiologic, clinical, diagnostic, and therapeutic aspects.

All confirmed VL cases in that period and region who agreed to participate were included. Only 2 patients did not agree to participate in the study and were therefore excluded. Deceased patients were included or excluded following telephone contact and/or a domestic visit to direct family members or spouses, which included an invitation, explanation of the methodology and importance of the research, and agreement. There were 10 deceased patients at the time of data collection; only 2 of them were excluded, because family members could not be located because of a change of residence. The study was initiated after informed consent of patients and family, and approval by research ethics committee of the Hospital Geral de Fortaleza (HGF) and Casamater hospitals as well as federal universities of Piauí and Ceará. Patients (n=30) had chronic renal disease and were undergoing dialysis or conservative treatment. Kidney transplantation occurred between January 1989 and January 2013, and VL was detected post-transplantation. The patients were transplanted in the kidney transplant units of the Hospital Geral de Fortaleza (HGF), Hospital Universitário Valter Cantídio (HUVC), Hospital Universitário de Barbalha of the Universidade Federal do Ceará, and Hospital Aliança Casamater, which are all located in the states of Ceará and Piauí in Northeastern Brazil. VL patients were treated with liposomal amphotericin 4 mg/kg/day for 10 days, amphotericin B up to the maximum dose of 1 g, or *N*-methylglucamine 30 mg/kg/day for 20 days [12,13]. VL patients with no symptoms, signs, or laboratorial alterations 6 months post-treatment were considered cured [14].

Relapse was defined as the occurrence of clinical manifestations of VL and new laboratorial identification of *Leishmania* in cases previously treated and considered cured up to 6 months post-treatment [15]. Graft dysfunction was defined as an increase in serum creatinine 30% above the baseline values in biochemical analyses performed before VL treatment in the absence of other factors associated with acute kidney injury [16,17]. Reports showed that renal dysfunction can be caused by acute tubular necrosis, acute injury secondary to VL and the use of drugs used in the treatment of disease and interstitial nephritis [18,19]. In this present study was excluded clinical and laboratory from graft dysfunction related to other causes, except renal changes due to VL.

In all cases studied was performed changing the protocol of immunosuppression in acute VL and during treatment with liposomal amphotericin or amphotericin B was reduced by 30% the dose of Prednisone and Mycophenolate mofetil and patients taking cyclosporine made by conversion to Tacrolimus with doses based on weight and 20% reduction in dose [9].

Variables analyzed

The following variables were analyzed. General characteristics included age, sex, ethnicity, breeding or co-habitance with domestic animals, and ornamental and/or fruit plant breeding grown indoors or outdoors. In this study, the cohabitation of patients with domestic animals was defined as patients raising and/or taking care of animals as well as animal presence in the neighborhood of residence or workplace. Other variables included the existence or absence of paved roads, wastelands, regular waste collection, sewage, and electricity in the neighborhood of residence or at the workplace as well as rural or urban area. Epidemiologic characteristics included education level, family income, housing, and awareness of human or canine VL. Clinical profile characteristics included the type of dialysis before transplant, post-transplantation blood transfusions, donor type, first transplantation or re-transplantation, immunosuppression protocol, bacterial and viral infections, graft rejection, common clinical manifestations in VL patients, VL diagnosis methods, drugs used for VL treatment, and response to therapeutics. Laboratory data included hematocrit, platelets, leukocytes, serum albumin, creatinine, and urea. All laboratory tests were performed at the beginning (day 1), middle (day 5), and end (day 10) of VL treatment as well as 90 and 180 days post-VL treatment in cured patients. Response to therapeutics was classified as disease progression to complete remission, death, graft dysfunction, or return to dialysis.

Data collection

After approval from the hospitals' respective ethics committees, data were collected through patient interviews and a structured questionnaire explained and proctored by the researchers. Detailed questions included demographics, routines, and socioeconomic conditions. Interviews were performed in a private room at the ambulatory care unit and lasted 30 minutes on average. If one or more family members were present, they were also allowed to participate in the interview. In cases of deceased patients, interviews were performed with the spouse or a direct family member during domestic visits. Recording procedures for clinical and laboratory variables, clinical evaluation, and VL treatment were reviewed; a new assessment of patients and grafts was performed at 6 months post-treatment.

Statistical analysis

Statistical analyses were performed with the use of descriptive statistics including means, standard deviations, frequencies, and percentages. To determine if the data were distributed normally, tests of equal proportions among all variables [i.e., general, epidemiologic, and clinical aspects as well as response to therapeutic [20] were performed using the χ^2 test.

Friedman ANOVA was used to analyze laboratory data, because the normality of the data could not be validated. When a significant difference was found by Friedman ANOVA, the Wilcoxon test was used as a post hoc test for multiple comparisons in order to test the pairs of variables that differed significantly; in such cases, significance was tested after performing a Bonferroni correction, in which p-values <0.05 are divided by the number of comparisons made. The baseline values were compared with values at each time point, which led to an a priori significance of 0.0125 (0.05/4 comparisons) for all variables except albumin, which had an a priori significance of 0.0166 (0.05/3 comparisons). Differences were considered significant only at $p \leq 0.0125$ (or 0.0166 in the case of albumin) in the Wilcoxon test [21,22]. The level of significance for all tests performed was $p<0.05$.

Results

General characteristics

The general characteristics of the patients are shown in (Table 1). The proportion of male patients with post-transplant VL was

Variable	Mean (SD)* *n* (%)	*P-value***
Age (years)	40.07 (10.5)	-----
Sex		
Male	24 (80.0)	*0.001*
Female	6 (20.0)	
Ethnicity		
White	13(43.3)	
Black	16(53.3)	*0.002*
Brown	1(3.3)	
Kidney transplant		
First transplant	27(90.0)	*0.002*
Re-transplant	3(10.0)	
Donor type		
Alive	17(56.7)	*0.585*
Dead	13(43.3)	
Donor sex (all donors)		
Male	20(66.7)	*0.099*
Female	10(33.3)	
Degree of kinship of live donors		
Second degree	11(36.7)	
Parents	10(33.3)	0.043
Siblings	8(26.7)	
Not related	1(3.3)	
Disease causing CRD§		
Diabetes mellitus	8(26.7)	
Arterial hypertension	3(10.0)	
Unknown	5(16.7)	0.432
Chronic GN¶	5(16.7)	
Other	9(30)	
Dialysis before transplant		
Hemodialysis	27(90.0)	0.001
Peritoneal dialysis	3(10.0)	

CRD§: chronic renal disease; GN¶: glomerular nephritis.*Mean (SD), Mean (Standard Deviation)**χ^2 test. Categorical variables are reported as n (%)

Table 1: General characteristics of 30 renal transplant recipients with post-transplant Visceral Leishmaniasis.

significantly greater than the proportion of female patients (80% vs. 20%, respectively, $p = 0.001$). Age ranged between 22 and 60 years with a mean ± SD of 40.07±10.50 years. There was a statistically significant difference with respect to ethnicity (p=0.002), with a low percentage of brown-skinned patients (3.3%). The majorities (90%) of patients were undergoing hemodialysis and had undergone their first transplantation before VL. A total of 56.7% of transplants were from live donors, the majority of whom were male (66.7%); parents accounted for 33.3% of donors. The diseases leading to chronic renal disease were arterial hypertension in 50% of cases as well as diabetes mellitus in 33.3% (p=0.002).

Epidemiologic characteristics

The epidemiologic characteristics of the patients are shown in (Table 2). There were uniform distributions of areas of residence (p=1.000) and workplace locations (p=0.099). Only 1 patient had advanced education (3.3%) (p= 0.003), the proportion of higher level of college education (3.3%) being responsible for therefore. Only 1 patient (3.3%) had a family income greater than US$ 601 (p=0.002).

There were no statistically significant differences with respect to VL ($p = 0.200$) between patients who lived with animals (63.3%) and those who did not (36.7%). Except for those who lived with pigs (p=0.265), there were significant differences between patients who lived with dogs, cats, chickens, and birds and those who did not (p=0.005, p=0.001, p=0.016, and p=0.005, respectively). The percentage of VL patients who had plants in or around the house was significantly different from the percentage of VL patients who did not (p=0.000 and p= 0.000, respectively). Sanitation and hygienic conditions housing were considered adequate in the majority of cases (90%). The proportions of patients who were aware of VL disease in humans and the VL vector were not significantly different, whereas there were significantly more patients who were aware of Canine Leishmaniasis (CL) (86.7%) compared to those who were not aware of it ($p = 0.000$).

Clinical characteristics

The clinical data of patients with post-transplantation VL are shown (Table 3). The mean number of blood transfusions before transplantation was 0.80±0.66, the mean time between transplantation and VL was 21.3±16.14 months, and the mean number of acute

Variable	*n* (%)	*P-value**
Residency		
Rural area	15(50.0)	*1.000*
Urban area	15(50.0)	
Workplace		
Urban area	20(66.7)	0.099
Rural area	10(33.3)	
Transplantation Center		
Ceará (HGF§, HUWC¶, Barbalha)	21(70.0)	0.043
Piauí (Casamater)	9(30.0)	
Education level		
Basic	14(46.7)	
Intermediate	15(50.0)	*0.003*
Advanced	1(3.3)	
Cohabitation with domestic animals		
Yes	19(63.3)	0.200
No	11(36.7)	
Cohabitation with dogs		
Yes	23(76.7)	0.005
No	7(23.3)	
Cohabitation with cats		
Yes	24(80.0)	0.001
No	6(20.0)	
Plant breeding at home		
Yes	27(90.0)	0.000
No	3(10.0)	
Awareness of human VL#		
Yes	11(37.9)	0.200
No	19(63.3)	
Awareness of VL vector		
Yes	10(33.3)	0.099
No	20(66.7)	
Awareness of CL⌘		
Yes	4(13.3)	0.000
No	26(86.7)	
Family income monthly		
Low (<US$ 200)	15(50.0)	
Average (between US$ 201 and 600)	14(46.7)	0.003
High (>US$ 600)	1(3.3)	

HGF§:Hospital Geral de Fortaleza; HUWC¶: Hospital Universitário Walter Cantídio; LV#:visceral leishmaniasis; LC⌘: canine leishmaniasis; *χ^2 test = Categorial variables are reported as n (%)

Table 2: Epidemiologic characteristics of 30 renal transplant recipients with post-transplantation Visceral Leishmaniasis.

rejections was 0.93±0.74. There was a significant difference between at least 1 blood serotype, with the proportion of serotype B (10%) apparently causing this difference. The majority of patients had Rh factor (73.3%; p=0.001). The majority of renal transplant recipients with VL had less than 3 incompatibility mismatches with the donor (83.3%), whereas only 16.7% had a greater number of mismatches (p = 0.001). There was no significant difference between the percentages of patients who did and did not receive blood transfusions before transplantation (43.3% v$_s$. 56.7%, respectively; p=0.465). Regarding the use of immune-suppressors, prednisone was used in 100% of cases, whereas there were significant differences with respect to Mycophenolate mofetil and Azathioprine use (p=0.001 and p=0.001, respectively). In contrast, there were no significant differences in the percentages of patients using of Tacrolimus or cyclosporine (p=0.465 and p=0.144, respectively). Significantly more patients underwent induction with a monoclonal antibody than no induction (70% vs. 30% respectively; p=0.028). There were no significant difference between the percentages of patients with and without acute rejection (40% vs. 60% respectively; p=0.362). All cases studied were first-time VL patients. Regarding post-transplantation infections, cytomegalovirus infection occurred in 40% of patients (p=0.362). Meanwhile, bacterial infections occurred in 36.6% of patients (p=0.002).

Symptoms and signs

The symptoms and signs detected in renal transplant recipients with post-transplantation VL are shown in (Table 4). The percentages of patients differed significantly among each of the categories of symptoms and signs (p<0.05), except for the existence of cavity fluids and edema; even though 67.7% of patients had these signs, the difference was not statistically significant (p=0.099).

Diagnosis

VL diagnosis was performed in 100% of patients (Table 5). In 73.3% of cases, *Leishmania* was directly isolated, whereas the detection was indirect (i.e., immunological test) in 26.7% of cases (p = 0.016). In addition, there were significant differences in the percentages of patients with respect to diagnostic method: myelogram (p=0.011), antigen rK39 isolation (p=0.000), polymerase chain reaction (p=0.000), and spleen biopsy (p=0.000).

Significantly more patients were treated with liposomal amphotericin (93.3%) than amphotericin B (6.7%; p=0.000). In addition, most patients did not receive *N*-methylglucamine (23.3% vs. 76.7%, respectively; p=0.005). After treatment, 26.7% of patients experienced VL relapse (p=0.016). Furthermore, there were significant differences in the percentages of patients with respect to achievement of cure (p=0.001), death (p=0.000), and achievement of cure with graft dysfunction (p=0.016). No significant differences were observed with respect to cure with graft loss (p=0.099) or cure with return to dialysis (p=0.099).

Laboratory data

Laboratory data were analyzed for all patients (n = 30) at the beginning and at days 5 and 10 of treatment (Table 6). In addition, the same examinations were performed 90 days and 180 days post-VL treatment in all cured cases (n = 24). Friedman ANOVA was performed to evaluate possible differences among the results of the hematological and biochemical parameters at different time points during VL treatment. Except for urea, which remained unchanged during treatment (p=0.511), there were significant differences among

Variable	Mean (SD)*	n (%)	P-value**
Number of blood transfusions before TxR§	0.80 (0.66)	-	-
Average time between TxR and VL¶ (months)	21.3 (16.15)	-	-
Acute rejection episode	0.93 (0.74)	-	-
Blood transfusion before TxR			
Yes		13(43.3)	0.465
No		17(56.7)	
Blood type of recipient			
Serotype O		14(46.7)	0.025
Serotype A		13(43.3)	
Serotype B		3 (10.0)	
Rh∞ factor of recipient			
Positive		22(73.3)	0.011
Negative		8(26.7)	
Incompatible mismatches, recipient–donor			
≤ 3 mismatches		25(83.3)	0.001
> 3 mismatches		5 (16.7)	
Use of prednisone			
Yes		30 (100.0)	-
No		0(0.0)	
Use of azathioprine			
Yes		6 (20.0)	0.001
No		22(80.0)	
Use of cyclosporine			
Yes		11(36.7)	0.200
No		19(63.3)	
Use of tacrolimus			
Yes		17(56.7)	0.565
No		13(43.3)	
Use of MMF♯			
Yes		27(90.0)	0.000
No		3(10.0)	
Induction with monoclonal antibody			
Yes		21(70.0)	0.043
No		9 (30.0)	
Acute rejection post-transplantation			
Yes		12(40.0)	0.362
No		18(60.0)	
Patients with VL before TxR			
Yes		0. (0.0)	-
No		30 (100.0)	
CMV♯ infection post-transplantation			
Yes		12(40.0)	0.362
No		18(60.0)	
Bacterial infection post-transplantation			
Yes		11(36.6)	0.002
No		19(73.4)	
Recipient with positive serology for other viruses			
No		20(66.7)	0.068
Yes		10(27.0)	

TxR§: renal transplantation; VL¶: visceral leishmaniasis; Rh∞: Rhesus factor; MMF♯, Mycophenolate mofetil; CMV♯: cytomegalovirus. **χ^2 test. Mean (SD)*, Mean (SD), Mean (Standard Deviation); Categorial variables are reported as n (%)

Table 3: Clinical characteristics of 30 renal transplant recipients with post-transplantation Visceral Leishmaniasis.

Variable	n (%)	P-value*
Fever		
Yes	21(70.0)	0.043
No	9(30.0)	
Weight loss		
Yes	30(100.0)	-
No	0(0.0)	
Skin lesions		
Yes	25(83.3)	0.000
No	5(16.7)	
Splenomegaly		
Yes	28(93.3)	0.000
No	2(6.7)	
Hepatomegaly		
Yes	21(70.0)	0.043
No	9 (30.00	
Active bleeding of the digestive tract mucosae		
Yes	7(23.3)	0.005
No	23(66.7)	
Jaundice		
Yes	6(20.0)	0.001
No	22(80.0)	
Pale skin and mucosae		
Yes	21(70.0)	0.043
No	9(30.0)	
Weakness and myalgia		
Yes	23(76.7)	0.005
No	7(23.3)	
Diarrhea		
Yes	26(86.7)	0.000
No	4(13.3)	
Fluids in visceral cavities		
Yes	20(66.7)	0.099
No	10(33.3)	

*χ^2 test.
Categorial variables are reported as n (%)

Table 4: Clinical manifestations in 30 renal transplant recipients with post-transplantation Visceral Leishmaniasis.

the other parameters at different time points. Multiple pairwise comparisons between the baseline and subsequent time points by the Wilcoxon test with a Bonferroni correction showed only creatinine increased significantly between baseline and day 5 (p=0.007).

Hematocrit differed significantly between baseline and day 10 (p=0.000), day 90 (p=0.000), and day 180 (p=0.000), with an increase in red blood cells from day 10. Leukocytes and platelets also differed significantly between baseline and days 5, 10, 90, and 180. Even though albumin was significantly different according to the Friedman ANOVA, the pairwise multiple comparisons between albumin levels at baseline and days 5, 90, and 180 did not reveal significant differences; this indicates albumin remained unchanged from the beginning of treatment until the time points studied (p =0.962; p=0.588, and p=0.182, respectively).

Discussion

Fewer than 100 cases of renal transplant recipients with VL are reported in the literature [23, 24]. This study introduces the greatest experience so far published of thirty VL-cases in renal transplant recipients This work, which is based on scientific evidence and the optimization of knowledge regarding epidemiologic, clinical, and diagnosis characteristics, is expected to become a reference for transplantation teams to adequately follow-up renal transplant recipients with VL; it is also expected to affect treatment, reducing patient mortality and improving the preservation of graft function [9]. In this series, 80% of patients were men. Men are more exposed to the

Variable	n (%)	P-value*
VL§ diagnosis		0.016
Direct identification of the parasite	22(73.3)	
Indirect method (immunological)	8 (26.7)	
Myelogram		0.011
Yes	19(63.3)	
No	11(26.7)	
Antigen rk39¶ isolation		0.000
Yes	5 (16.7)	
No	25 (83.3)	
PCR#		0.000
Yes	1(3.3)	
No	29(96.7)	
Spleen Biopsy		0.000
Yes	2(6.7)	
No	28(23.3)	
Use of amphotericin		0.000
Liposomal amphotericin B	28 (93.3)	
Amphotericin B	2(6.7)	
Use of *N*-methylglucamine		0.005
Yes	7 (23.3)	
No	21(66.7)	
VL relapse		0.016
Yes	8 (26.7)	
No	22(73.3)	
Patients with VL remission		0.001
Yes	24(80.0)	
No	6 (20.0)	
Deceased patients		0.000
Yes	5 (16.7)	
No	25(83.3)	
Patients with VL remission and graft loss		0.099
Yes	10(33.3)	
No	20(66.7)	
Patients with VL remission and graft dysfunction		0.016
Yes	22(73.3)	
No	8 (26.7)	
Patients with VL remission and return to dialysis		0.099
Yes	10(33.3)	
No	20(66.7)	

VL§: visceral leishmaniasis; rK39¶ : antigen extracted from *Leishmania* sp.; PCR#: polymerase chain reaction. *χ^2 test.
Categorial variables are reported as n (%)

Table 5: Methods for diagnosis, treatment, and response to therapeutics in 30 renal transplant recipients with post-transplantation Visceral Leishmaniasis.

Variable	Mean (SD)*	P-value**
Creatinine (mg/dL)		
1st evaluation	1.81 (0.4)	
2nd evaluation	2.21(0.46)	0.002
3rd evaluation	1.9 (1.17)	
90 days post-treatment	1.77(0.27)	
180 days post-treatment	1.80(0.33)	
Urea (mg/dL)		
1st evaluation	105.13(16.91)	
2nd evaluation	97.80(15.89)	0.000
3rd evaluation	65.13(27.18)	
90 days post-treatment	63.40(20.40)	
180 days post-treatment	52.07 (9.98)	
Hematocrit (%)		
1st evaluation	28.72(4.18)	
2nd evaluation	29.72(3.65)	0.000
3rd evaluation	34.94(3.50)	
90 days post-treatment	37.73(2.57)	
180 days post-treatment	38.76(1.72)	
Leukocytes (mm³/dL)		
1st evaluation	3.07(1.05)	
2nd evaluation	4.51(3.27)	0.000
3rd evaluation	5.86(2.71)	
90 days post-treatment	6.18(1.41)	
180 days post-treatment	6.40(0.95)	
Platelets (mm³/dL)		
1st evaluation	110.33(88.18)	
2nd evaluation	178.07(183.17)	0.000
3rd evaluation	163.00(72.02)	
90 days post-treatment	177.00(18.13)	
180 days post-treatment	183.07(11.88)	
Serum albumin (mg/dL)†		
1st evaluation	3.32(0.76)	
2nd evaluation	3.74(0.37)	0.009
90 days post-treatment	3.87(0.34)	
180 days post-treatment	3.99(0.27)	

n = 24 remission patients.
1st evaluation: day 1 of treatment with amphotericin or glucamine.
2nd evaluation: day 5 of treatment with amphotericin or day 10 with glucamine.
3rd evaluation: day 10 of treatment with amphotericin or day 20 with glucamine.
† Serum albumin was not measured at intermediate time points.
** Determined by Friedman ANOVA, the Wilcoxon test or ANOVA factor repetition measurements.
Mean and SD* (Mean (Standard Deviation) of leukocytes and platelets were divided by 1.000.
Categorial variables are reported as n (%)

Table 6: Laboratory data of 24 patients with post-transplant Visceral Leishmaniasis in remission and up to 6 months post-transplantation.

mosquito vector of VL (*Lutzomyia longipalpis*) as a result of professional activities, greater body surface area, and the habit of remaining shirtless in the high-temperature environments characteristic of tropical regions [25,26]. Despite the lack of reports of an association between ethnicity and VL, the majority of patients in the present study were black.

Only 1 study addresses the issue of the period between transplantation and VL diagnosis; however, there are no reports of an association between VL and the type or dose of immunosuppressors used [9] Other recent studies show the mosquito vector has migrated from rural to urban areas in search of food [26,27]. The majority of studies in the literature report dogs are the main intermediary host in the life cycle of VL. However, Alves da Silva and collaborators recently reported that cats are also important hosts of *Leishmania*. VL is a neglected disease that is highly prevalent in the world's poorest regions and is positively correlated with deficient hygiene and sanitation [28,29]. However, the present study shows the majority of VL patient had access to treated water, electricity, and regular waste collection, which suggests immunosuppression may be a determinant cause of VL in these patients related to the use of immunosuppressors, hemodialysis, post-transplant infection, malnutrition, and donor incompatibility, especially in cases of deceased donors [9,30,31]. The VL is considered a disease neglected by the rulers, being prevalent in the poorest countries of low socioeconomic status. It is only by systemic disease that leads to low immunity of patients, so it is more prevalent in malnourished patients with chronic diseases, HIV patients and in patients using immunosuppressive medication (kidney transplant). In endemic areas these groups of patients are exposed to similar epidemiological characteristics and studies have shown similar clinical and laboratory frame. So in endemic areas, the LV should always be part of the differential diagnosis, resulting in early diagnosis and treatment, increasing patient survival and graft [2,3].

Here detected by humoral antibodies current, which seem to have little importance as a defense. Leishmania is an obligatory intracellular parasite cells of the mononuclear phagocyte system, and their presence causes a reversible and specific suppression of cell-mediated immunity, which allows the dissemination and uncontrolled multiplication of the parasite. Only a small proportion of infected individuals develop signs and symptoms of the disease [32]. After infection, where the individual does not develop the disease, it is observed that the exams researching immunity cellular or humoral remain reactive for long periods; this requires the presence of antigens, can be concluded that the *Leishmania* or some of their antigens are present in the infected organism for a long time of his life, after the initial infection. This hypothesis is supported by the fact that some individuals who develop immunosuppression may present framework of LV far beyond the usual incubation period [33].

Several studies failed to demonstrate a relationship between blood serotype and VL; in the present study, there were significant differences in the percentages of patients with respect to blood type and Rh factor, although there is no evident clinical explanation for this [34].

Previous studies report difficulties diagnosing VL in renal transplant recipients because of atypical signs and symptoms [35]. In the present study, clinical manifestations were typical even in immunosuppressed patients and showed that the disease occurred an average of 21.6±16.15 months (range: 6–110 months) after transplantation; this contributes to the differential diagnosis of VL in cases of fever, a characteristic manifestation in all regions including non-endemic regions [36]. Clinical picture, diagnosis, treatment and outcome of patients and grafts was recently published in detail by Alves da Silva et al in a case-control study in which assessed such characteristics in transplant patients with and without visceral leishmaniasis [9].

VL diagnosis was performed in 100% of cases, and myelogram was the main method used to identify *Leishmania* forms; in cases with negative results, indirect determination was performed by the identification of rK39 antigen. In the present study, liposomal amphotericin was the drug of choice for VL treatment, resulting in high levels of disease remission, low relapse, and few deaths [37, 38]. The results also show an association between VL and post-transplantation bacterial and cytomegalovirus infections; this can be explained by the reduced immunity of patients, who become more

susceptible to opportunistic infectious organisms such as *Leishmania* [39,40]. Regarding laboratory analyses, the present results indicate that in the process of evolving to a systemic disease, VL seriously damages the kidneys, liver, spleen, and hematopoietic system. However, early diagnosis and adequate treatment led to significant improvement of these alterations.

Conclusion

In conclusion, this study is the largest series of VL in renal transplant recipients, which focuses on several aspects of the disease, including epidemiology, clinical profile, diagnosis, and treatment. The results herein advance knowledge about VL in renal transplant recipients and may increase awareness of this emerging infection among transplantation teams. Standardized routines for the adequate follow-up of VL patients are recommended, taking into consideration the specificity of renal transplant recipient.

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