Renal Leishmaniasis as Unusual Cause of Nephrotic Syndrome in an HIV Patient

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ABSTRACT

Renal involvement is a rare complication in HIV-1–infected patients leading to various pathologies and clinical symptoms. In addition to the classic HIV-1–associated nephropathy with collapsing-type focal segmental glomerulosclerosis and characteristic tubulocystic changes, which is more common in Afro-American than in Caucasian HIV-1 patients, immune complex GNs such as membranous GN and membranoproliferative GN are particularly common renal manifestations. Besides HIV-1 itself, a number of opportunistic infections may cause renal disease in HIV-1–infected patients. In this study, we report an unusual case of HIV-1 infection with a severe renal manifestation of systemic leishmaniasis that developed years after repeated visits to Mediterranean countries. The case presents several remarkable clinical, pathologic, and therapeutic aspects that may be important for daily clinical practice.

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In April 2008, a 45-year-old Caucasian male patient was admitted to our hospital due to dyspnea and recurrent diarrhea during the past weeks. He had been known to be HIV-1 infected for 23 years, and he had been treated with various antiretroviral drug regimens since 1997. Despite therapy, he had experienced progressive immunodeficiency due to poor compliance and drug resistance. At a prior visit to our hospital in 2001, he had presented with a high viremia (500,000 copies/ml) and depleted CD4 cells (3/μl). At the current admission, he was on an antiretroviral combination regimen consisting of darunavir, low-dose ritonavir, tenofovir, and emtricitabine.

Physical examination revealed an elevated blood pressure of 170/110 mmHg, dyspnea, bilateral pleura effusions, gross edema of legs, scrotum, and penis, and reddish, macular and papular skin lesions on the dorsal hands, chest, abdomen, and thighs. Laboratory analyses showed abnormal renal parameters (serum creatinine 1.49 mg/dl [normal range <1.1 mg/dl], creatinine clearance 35 ml/min [normal value 75–125 ml/min], urea 69 mg/dl [normal range 14–43 mg/dl], uric acid 7.5 mg/dl [normal range 3.5–7 mg/dl], and proteinuria of 9.6 g/24 h). Liver enzymes (aspartate aminotransferase 56 U/L [normal <35 U/L], γ-glutamyl transferase 87 U/L [normal <55 U/L]) and lactate dehydrogenase (502 U/L [normal <250 U/L]) were elevated. Serum ferritin was strongly increased to 1770 ng/ml (normal range 34–310 ng/ml), and iron was slightly decreased to 28 μg/dl (normal range 40–160 μg/dl). Peripheral blood counts showed anemia (hemoglobin 8.3 mg/dl [normal range 12–16 mg/dl]), slight leukopenia (3900/μl [normal range 4000–10,000/μl]), and thrombopenia (99,000/μl [normal range 150,000–400,000/μl]). The number of $CD4^+$ T cells was decreased to 174 cells/μl. Quantitative PCR analyses demonstrated low HIV-1 viremia (790 copies/ml), a low cytomegalovirus viremia (6 copies/10^5 cells), and shedding of BK virus in the urine (4400 copies/ml). PCR analyses yielded negative results for hepatitis C virus in serum and for adenovirus and John Cunningham virus in the urine. All other laboratory parameters including the complement factors C3 and C4, antinuclear antibodies, cryoglobulins, anti-dsDNA antibodies, perinuclear/cytoplasmic anti-neutrophil cytoplasm antibodies, anti–glomerular basement membrane antibodies, and haptoglobin were negative or normal. Chest x-ray and computed tomography scan showed hypervolemia with bilateral pleural effusion.

Because of a further rise of serum creatinine (2.25 mg/dl) and the development of a nephritic sediment with 50% dysmorphic erythrocytes, a renal biopsy was performed, and two biopsy cylinders of 0.4 and 0.5 cm, respectively, were obtained (Figure 1). On light microscopy, renal cortical and medullary tissue with 16 glomeruli was seen. These were hypercellular with mesangial and

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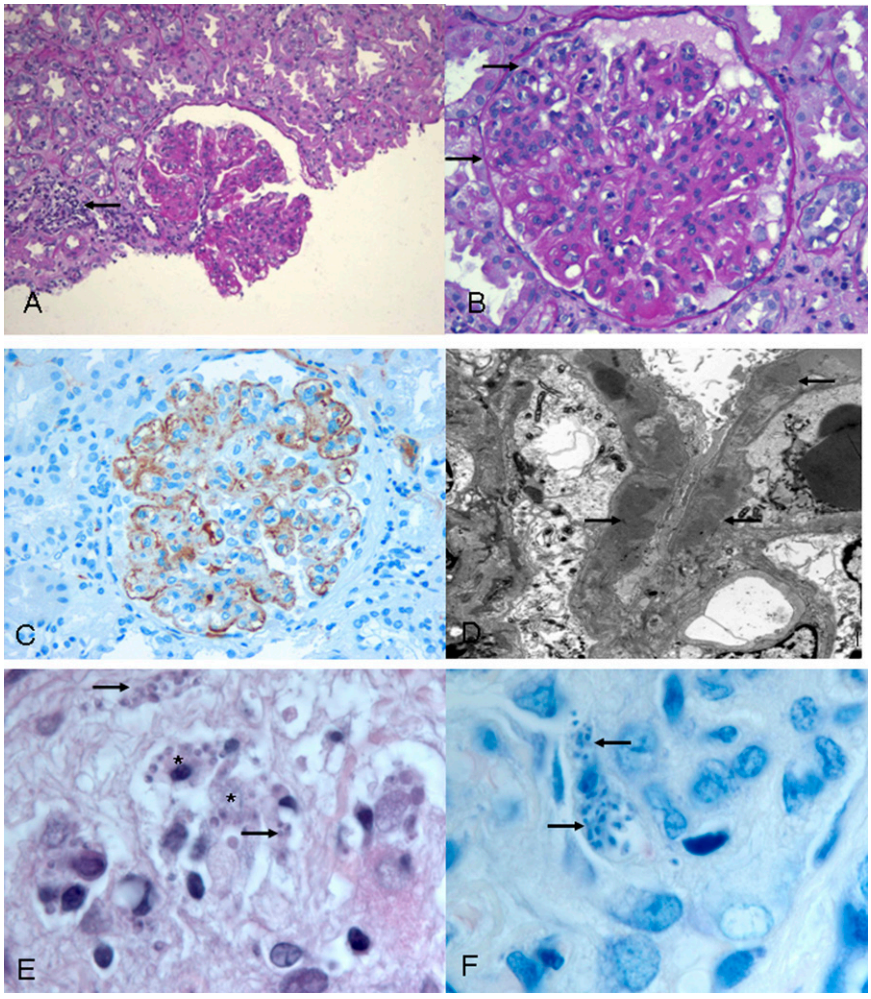

Figure 1. Representative light microscopical, immunohistochemical, and electron microscopical pictures of renal involvement in VL. (A) Renal biopsy, periodic acid–Schiff stain, original magnification ×20. Even at low magnification, lobulation of the capillary tuft with mesangial and endothelial hypercellularity and capillary lumen obliteration is clearly visible. In the adjacent interstitium, a focal inflammatory infiltrate due to *Leishmania* amastigotes is visible (arrow). (B) Renal biopsy, periodic acid–Schiff stain, original magnification ×40. At higher magnification, glomeruli show mesangial and endothelial hypercellularity and thickened and double-contoured GBMs (arrows). (C) Renal biopsy, immunohistochemistry with an antibody against IgG. Granular deposits of IgG can be seen in a mesangiocapillary pattern. Original magnification ×40. (D) Renal biopsy, electron microscopy. Ultrastructural analysis confirmed thickening of the GBM together with subendothelial, mesangial, and intramembranous osmiophilic deposits (arrows). Original magnification ×6000. (E) Renal biopsy, hematoxylin and eosin stain, original magnification ×100. Small round to ovaloid microorganisms with kinetoplasts compatible with *Leishmania* amastigotes can be seen in the peritubular capillaries (arrows) and also in some tubular epithelial cells (asterisks). (F) Renal biopsy, Giemsa stain, original magnification ×100. Giemsa-positive round to ovaloid microorganisms with kinetoplasts compatible with *Leishmania* amastigotes are clearly visible within peritubular capillaries (arrows).

endothelial cell proliferation in a lobular pattern, matrix expansion, and thickened, sometimes double-contoured glomerular basement membranes (GBMs) (Figure 1, A and B). In the glomerular capillaries, only a few leukocytes were seen. In the interstitium tubular atrophy, interstitial fibrosis and very mild, scattered lympho-plasmacellular infiltration were noted. There were no signs of acute tubular damage. Arterial vessels showed marked wall thickening but no vasculitis. Immunohistochemistry with antibodies against IgA, IgG, IgM, C1q, and C3c also revealed a lobular aspect of the glomeruli with marked (+++) and granular deposition of IgG and C3c along the GBM and within the mesangium (Figure 1C), whereas stainings for C1q, IgM, and IgA remained negative. Despite the presence of BK virus DNA in the urine, no positively stained nuclei were detectable in the renal biopsies with a Simian virus 40-specific antibody cross-reacting with BK virus. On electron microscopy (Figure 1D), thickening of the GBM together with subendothelial, mesangial, and also intramembranous osmiophilic deposits (*i.e.*, immune complexes) with mild capillary wall mesangial cell migration was observed. Of note, tubuloreticular inclusions or microorganisms were not seen. Hence, MPGN type I was diagnosed.

Because of his persistent diarrhea, the patient also underwent a coloscopy. The colon biopsies showed mild interstitial inflammation together with dense deposits of round to ovaloid microorganisms in the lamina propria. These were intensely positive in the Giemsa stain with clearly visible kinetoplasts compatible with *Leishmania* amastigotes. The same microorganisms were detected in the skin lesions. Subsequently, *Leishmania infantum* promastigotes (identified by *Leishmania* 18S rRNA gene sequencing) were grown from colon, skin, and peripheral blood samples. When a Giemsa stain of the renal biopsy was performed, *Leishmania* amastigotes were found in the tubulointerstitium (Figure 1E), but also within the glomeruli (Figure 1F). Thus, a final diagnosis of visceral leishmaniasis (VL) with involvement of the kidneys (MPGN), the skin, and the colon was made.

The patient was a German native who had visited southern Italy (yearly from 2000 to 2003) and Tunisia (2004) (*i.e.*, areas endemic for *L. infantum*). Therefore, the VL either resulted from a reactivation and systemic spread of persisting parasites from a first infection or from a slowly progressing *de novo* infection that was acquired during later years.

Intravenous treatment with liposomal amphotericin B (AmB; 3 mg/kg body wt per day) was started, which only led to a limited reduction of the proteinuria to 6 g/dl. After 10 days of treatment, parasites were still grown from another skin biopsy despite their *in vitro* sensitivity to AmB (IC_{50} 1.25 μg/ml). Because of the persisting infection and possible renal side effects, treatment with AmB was stopped after more than 20 infusions and changed to oral miltefosine (150 mg/d). Thereafter, clinical symptoms and renal function improved with a further decline of proteinuria to 3.56 g/L so that the patient could be discharged from the hospital on prolonged treatment with miltefosine. Because of the detectable HIV-1 viremia at admission and known drug resistance as a result of poor compliance, antiretroviral therapy was intensified. A combination therapy consisting of efavirenz, raltegravir, darunavir/ritonavir, tenofovir, and emtricitabine finally suppressed his HIV-1 viral load below 30 copies/ml.

Five months later, serum creatinine had improved but was still elevated with 1.23 mg/dl (normal range <1.1 mg/dl). Urine analysis revealed a persisting proteinuria (dipstick +++) without erythrocyturia. There was an elevation of C-reactive protein to 11.4 mg/L, of γ-glutamyl transferase to 73 U/L (normal range <60 U/L), of uric acid to 8.7 mg/dl (normal range 3.4–7 mg/dl), and of lactate dehydrogenase to 335 U/L (normal range <250 U/L), whereas other chemistry parameters such as aspartate aminotransferase, alanine aminotransferase, and bilirubin were within normal range. Blood counts revealed normal values for platelets (236,000/μl), hemoglobin (13.2 g/dl), and leukocytes (6270/μl) with lymphocytosis of 46%. He showed a detectable HIV-1 viremia of 1200 copies/ml and a CD4 count of 218/μl. The persisting HIV-1 viremia could be explained by poor compliance as serum drug levels were low for efavirenz (168 ng/ml) and below the detection limit for darunavir and ritonavir. Although the number of skin lesions had declined on therapy with miltefosine, several reddish skin lesions persisted. *Leishmania* species PCR was still positive within a biopsy taken from one of these skin lesions. We recommended continuation of miltefosine and modification of his antiretroviral therapy, but the patient failed to return to follow-up visits due to poor compliance.

HIV-1 INFECTION AND RENAL LEISHMANIASIS

As the cause of the renal disease in this patient, HIV-associated nephropathy (HIVAN), classic type FSGS, and various types of GN that are often observed during HIV-1 infection have to be considered[1–5] (Table 1). HIVAN, however, is unlikely to account for the renal manifestations. First, the patient had already been infected for more than two decades and despite drug therapy since 1997 he had previously experienced severe immunosuppression and high HIV-1 viremia for several years due to poor drug compliance and drug resistance. When the renal disease became clinically apparent, his viral load was low, although not fully suppressed (790 copies/ml), and his CD4 count of 174/μl was well above his CD4 nadir of three $CD4^+$ cells/μl 7 years ago. Second, the typical histologic criteria of HIVAN (*i.e.,* collapsing-type FSGS and cystic dilation of tubuli) were not present.[1–3] Classic FSGS and membranous GN could be ruled out by light microscopy and immunohistochemistry, which showed a mesangiocapillary staining pattern for IgG, C1, and C3. In addition, electron microscopy confirmed the subendothelial and intramembranous localization of the immune deposits.

Immune complex GNs are common in HIV-1–infected patients and comprise a wide spectrum (*e.g.,* IgA-GN, membranous GN, MPGN, lupus-like GN,[6–9] and so forth) (Table 1). It is not clear, however, whether immune complex deposition in HIV results from passive trapping of circulating immune complexes or the *in situ* deposition of antibodies binding to HIV-1 antigens. Some renal lesions that are seen during HIV-1 infections, however, are more likely to be related to the presence of systemic co-infections such as hepatitis C.[8] In addition, immunotactoid glomerulopathy, fibrillary GN, and thrombotic microangiopathy have been described to cause renal symptoms in HIV patients[7,8] (Table 1).

VL is a protozoan infection usually transmitted by bites of sandflies. It is prevalent in tropical, subtropical, and in all Mediterranean countries. The clinical presentation of VL varies from an asymptomatic infection to fatal disease. Patients most commonly present with fever, malaise, hepatosplenomegaly, and pancytopenia leading to increased susceptibility to secondary infections and a tendency to bleed. VL is an important opportunistic infection in severely immunosuppressed

Table 1. Spectrum of HIV-associated glomerular disease

1. HIVAN
collapsing type of FSGS and typical tubulointerstitial changes (*i.e.,* microcystic tubular dilation with cast formation and interstitial inflammation)
2. Non-HIVAN glomerular lesions
immune complex GN
MPGN
membranous GN
IgA GN
lupus-like GN
postinfectious GN
minimal change nephropathy
non-collapsing type of FSGS
TMA
immunotactoid glomerulopathy
fibrillary GN
renal amyloidosis
3. Glomerular involvement in systemic infections (*i.e.,* leishmaniasis)

patients including kidney transplant recipients or HIV-1–infected patients, where it either results from the reactivation of latent parasites or from a newly acquired infection.[10–12] HIV-1–infected patients usually have higher parasite loads and present with atypical symptoms (*e.g.*, gastrointestinal), especially with low CD4 cell counts. This group of patients often responds poorly to treatment, and rates of relapse are high.

Infections with *Leishmania* spp. may be accompanied by various renal symptoms including acute renal failure, renal amyloidosis, various forms of GN, and nephritic syndrome.[12–15] Circulating immune complexes containing anti-*Leishmania* antibodies have been detected in humans suffering from VL.[16,17] There is evidence for the deposition of *Leishmania* antigen–containing immune complexes in the kidneys of spontaneously or experimentally infected animals.[18–20] In contrast, autoantibodies (*e.g.*, directed against nuclear proteins) as well as anti-*Leishmania* antibodies cross-reacting with host antigens have been found in humans with VL,[21–23] which could also contribute to the renal inflammation and damage.

MPGN due to infection with *Leishmania* spp. is known from naturally infected dogs, which form a major reservoir of the pathogen in Mediterranean countries.[13,14] Recently, two case reports described nephrotic range proteinuria due to MPGN in VL in HIV-1–infected patients.[14,15]

The number of VL relapses and the average time between them largely depend on the immunologic status of the patient (as defined by the CD4 cell count) and are definitely influenced by the antiretroviral treatment and the use of anti-leishmanial drugs for maintenance therapy. Both the clinical manifestations and the course and prognosis of VL in HIV-1–infected individuals differ markedly from those in non–HIV-infected individuals. VL–HIV-1 coinfection is characterized by significantly lower cure rates, higher drug toxicity, higher relapse rates, and higher mortality rates compared with VL in non–HIV-infected individuals.

CONCLUSION

Diagnosis of VL in AIDS patients can be difficult due to atypical presentations like that in our case. Severe immunosuppression in AIDS patients favors the persistence and systemic spread of the parasite to various organs and predisposes to frequent relapses of VL. Success of treatment depends not only on aggressive therapy for leishmaniasis but also on immune reconstitution by antiretroviral therapy for the underlying HIV-1 infection.

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DISCLOSURES

None.

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