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Visceral leishmaniasis in immunocompromised patients with and without AIDS: a comparison of clinical features and prognosis

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Abstract

Visceral leishmaniasis is basically a disease of healthy infants and adults. However, in the last decade an increasing number of cases of kala azar in immunocompromised patients have been reported with emphasis on atypical manifestations of the disease. During a period of 11 years, 20 immunocompromised patients with AIDS (12 patient), haematological neoplasia (3 patients), corticosteroid therapy (3 patients) or renal transplantation (2 patients) were studied by one or more of the authors. We did not find differences in the presentation of leishmaniasis between patient with or without AIDS and most patients had fever, enlargement of the liver and spleen, blood cytopenias and biochemical abnormalities. Serology was more frequently positive in HIV-negative than in HIV-positive patients (100% versus 63.6%; $P = 0.13$). Bone marrow biopsy was diagnostic in 66% and 87% of patients with and without AIDS, respectively. Failure of anti-leishmanial therapy occurred in 6 of 19 patients treated (31.5%), and 3 patients with AIDS and another 3 without AIDS died during the first episode of leishmaniasis. Of 12 survivors, relapses occurred in five (41.6%). Only patients in whom immunosuppression was ameliorated by means of antiretroviral therapy or by reduction of corticosteroid and other immunosuppressive drugs did not relapse. Treatment of kala azar in immunocompromised host is in satisfactory and new drugs or strategies are urgently needed.

Keywords: Visceral leishmaniasis; Kala azar; Immunocompromised hosts; AIDS

1. Introduction

Visceral leishmaniasis also known as kala azar, is a protozoan disease caused by different species of *Leishmania donovani* complex. The disease is endemic in the tropics of the America, parts of Asia and tropical Africa north of the equator (Berman, 1997). Leishmaniasis is also seen in the Mediterranean basin where *L. infantum* is the major causative agent (Alvar et al., 1997).

People in non-endemic areas, such as North America and most European countries, have acquired the infection after visiting endemic areas. The disease has also been reported in veterans of the Gulf war in the Middle East (Magill et al., 1992).

Leishmaniasis is basically a disease of otherwise healthy children and adults (Badaró et al., 1986a).

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However, a particularly severe form of disseminated leishmaniasis has been described in immunocompromised patients with hematologic neoplasia, recipients of solid-organ transplants and patients with a variety of diseases on corticosteroid therapy (Fernández Guerrero et al., 1987; Berenguer et al., 1998). In addition, the disease has been found in an increasing number of patients with AIDS in Spain, France, Italy and in some parts of Africa and the America (Berenguer et al., 1989; World Health Organization, 1997; Berhe et al., 1995).

We show herein the most relevant clinical and pathologic findings and prognosis of visceral leishmaniasis in immunocompromised patients. A comparison of the manifestations of the disease in patients with and without AIDS is also included.

2. Patients and methods

The clinical, pathologic and microbiologic records of all patients with kala azar seen by the authors in a period of 11 years (1991–2001) were retrospectively assessed. The study was conducted in a 600 bed, tertiary-care, university-affiliated hospital in Madrid, Spain.

Diagnosis of visceral leishmaniasis was confirmed by demonstration of amastigotes in bone marrow smears or biopsy. Anti-leishmanial antibodies were assayed by indirect immunofluorescence using a commercial test (leishmania indirect immunofluorescence antibody test, Mardx Diagnostics, Scotch Plains, NJ). Titers equal or greater than 1:80 were considered positive. During the period of the study, bone marrow biopsies were routinely cultured in Novy-McNeal-Nicolle medium (Benito et al., 1997). Cultures were incubated at room temperature (22–25 °C) and examined every other week for 4 weeks.

Patients were considered immunocompromised when they had a previous diagnosis of hematologic neoplasia treated with chemotherapy and/or radiotherapy, were recipients of a solid-organ transplant or were on high-dose corticosteroid therapy ($\geq$30 mg of prednisolone per day) for prolonged periods (more than 30 days). AIDS was diagnosed according to accepted criteria (Centers for Disease Control and Prevention, 1992).

The following data were obtained routinely by reviewing the clinical charts of the patients: demographic information (age, sex, underlying disease), stage and risk factors for HIV infection, clinical presentation, laboratory tests, methods of diagnosis, treatment, and outcome.

The clinical response was defined as clinical cure if after a complete course of treatment the patient was afebrile and the physical signs and laboratory abnormalities subsided or significantly improved. Clinical failure was defined as persistence of clinical signs and symptoms after completion of anti-leishmanial therapy and when the patient died due to leishmaniasis and its complications. Relapse was defined as a new episode of visceral leishmaniasis occurring after a successful response to treatment of the previous episode.

2.1. Statistical analysis

Quantitative variable analysis was performed with the Student *t* test. The study of qualitative variables was done with the chi-square test with the Yates correction or the Fischer exact test (two-tailed) when necessary.

3. Results

Twenty immunocompromised patients with visceral leishmanisis were studied; 12 (60%) were HIV-infected patients and 8 (40%) had other immunosuppressive conditions such as hematologic neoplasia (3 patients), corticosteroid therapy (3 patients) and renal transplantation (2 patients). Patients with hematologic neoplasia (two with acute leukaemia and one with Hogdkin's lymphoma) had been treated with a variety of cytotoxic drugs plus corticosteroid for a mean of 63 weeks before the onset of leishmaniasis. The recipients of renal transplants were on immunosuppressive treatment with a combination of prednisolone and azathioprine for a mean of 43 weeks. Only patients with hematologic neoplasia and one patient with renal transplantation had a CD4+ T cell count performed. The CD4+ T cell count in these patients ranged from 99 to 310/mm^3 (mean CD4 count was 203/mm^3).

All HIV-infected patients were men, with a median age of 43.7 years (range 22–65 years). Seven

were drug-abusers and five were homosexual men. All these patients had full-blown AIDS with a medium CD4+ lymphocyte of 81.2 ± 46 cells per microliter. The group of patients without AIDS was composed of four men and four women with a median age of 45 years (range 15–60 years). All the patients lived in Madrid and its outskirts, an area with a low annual incidence of visceral leishmaniasis (1.03 cases per 100,000 inhabitants) (Fernández Guerrero et al., 1987).

Clinical findings, diagnostic procedures and outcome of immunocompromised patients with visceral leishmaniasis are shown in Table 1. Fever was a constant findings in both groups and we did not find any significant difference in patients with and without AIDS regarding hepatomegaly (7 of 12 versus 4 of 8); splenomegaly (5 of 12 versus 5 of 8) or enlargement of lymphatic nodes (3 of 12 versus 1 of 8). In addition, pancytopenia was found in 91% (11/12) and 100% (8/8) of patients with and without AIDS, respectively. Serum levels of albumin (2.6 g/dl versus 2.85 g/dl) and γ-globulins (2.96 g/dl versus 3.1 g/dl) were not significantly different in both groups. Minor to moderate elevations of γ-glutamyltranspeptidase, SGOT, SGPT and lactic dehidrogenase were frequently found in both groups.

Table 1
Clinical findings, diagnostic procedures and outcome of a series of 20 immunocompromised patients with visceral leishmaniasis

Variable	HIV-infected patients, 12 (100%)	Non-HIV-infected patients, 8 (100%)	*P*
Male sex	12 (100)	4 (50)	
Fever	12 (100)	8 (100)	
Hepatomegaly	7 (66)	4 (50)	
Splenomegaly	5 (41,6)	5 (62)	0.32
Lymphadenopathy	3 (25)	1 (12,5)	
Leukopenia and anemia	11 (91,6)	7 (87,5)	
Trombocytopenia	11 (91,6)	8 (100)	
Elevations of liver enzymes (SGOT, SGPT and γGTP)	11 (91,6)	6 (75)	
Positive serology[a]	7/11 (64)	6/6 (100)	0.13
Positive bone-marrow smears	10 (83,3)	8 (100)	
Clinical failure	3 (27.2)	3 (37,5)	
Clinical cure	7 (58.3)	5 (62,5)	
Parasitologic cure	4 (57.1)	3 (60)	
Relapse	3 (42.8)	2 (40)	

[a] Indirect immunofluorescence.

Serologic tests were positive in 7 out of 11 (63.6%) HIV-infected patients tested in comparison to 6 out of 6 (100%) immunocompromised patients without AIDS ($P = 0.13$). Titers of antibodies against *Leishmania* ranged from 1:80 to 1:640 in both groups of immunocompromised patients. Diagnosis was achieved by bone marrow smears or bone marrow biopsy in 10/12 (83.3%) HIV-infected patients and in 8/8 (100%) of other immunocompromised hosts. Giemsa-stained smears were positive in 66 and 87% of cases, respectively. Gastric and cutaneous biopsies were diagnostic in two HIV-infected patients. In one patient, diagnosis was achieved by positive bone-marrow cultures.

The patients were treated with meglumine antimoniate 20 mg/Kg for 28 days (13 patients), meglumine antimoniate plus gamma interferon (2 patients) or amphotericin B 0.7–1 mg/Kg for 28 days (four patients). One HIV-infected patient died few hours after diagnosis and did not receive anti-leishmanial treatment and another one died on Day 23 of treatment due to ventricular arrythmia associated with meglumine antimoniate therapy. Overall, 8 (40%) immunocompromised patients died during the first episode of visceral leishmaniasis and 12 (60%) improved or were cured with anti-leishmanial therapy. Clinical failure of anti-leishmanial therapy was observed in 6 of 19 (31.5%) patients treated. Among 12 survivors, leishmanisis relapsed in 5 (41.6%) in a period ranging from 3 to 8 months after discontinuation of treatment.

The comparison between both groups of immunocompromised patients showed the following data: clinical failures were observed in 3 out of 11 (27.2%) HIV-infected patients who did not respond to anti-leishmanial treatment and died during the course of therapy and in 3 out of 8 (37.5%) immunocompromised patients without AIDS who also died during treatment or shortly afterwards. All these patients died of causes related to the underlying disease—AIDS, leukaemia or solid-organ transplantation—and leishmaniasis (bacterial infections including *Pseudomonas aeruginosa* and *Mycobacterium avium* complex infection, *Pneumocystis carinii* pneumonia or haemorrhage).

One or more relapses were seen in three out of seven survivors with AIDS and in two out of five immunocompromised patients without AIDS. Three patients with AIDS who did not relapse showed a

good response to highly active antiretroviral therapy (HAART) with undetectable viral loads and CD4+ lymphocyte counts greater 200/μl. In addition, relapses were not seen in two patients without AIDS who stopped or reduced corticosteroid therapy. These patients have remained free of leishmaniasis one or more years after discontinuation of Glucantime R therapy.

4. Discussion

With increasing travel to endemic areas, diagnosis of leishmaniasis continues to challenge the clinician and pathologist. Development of leishmaniasis in an increasing number of immunocompromised patients complicates the diagnosis and treatment of the infection and shows the new face of the disease that must be known by other providers of medical care such as the parasitologist, and the travel medicine and the infectious disease physicians.

Leishmaniasis causes clinical and histologic manifestations that depend on the host response, and the state of the immune system greatly determines the intensity and efficacy of such response. In immunocompromised patients, leishmaniasis is a severe, life-threatening disease characterized by disseminated visceral involvement, atypical manifestations and suboptimal response to therapy (Fernández Guerrero et al., 1987; Berenguer et al., 1998; Berenguer et al., 1989). Since host defense against *Leishmania* is mediated by T cells and T-cell-derived substances such as interferon gamma directed at infected macrophages, alterations of these mechanisms by disease or drugs may promote the development of visceral leishmaniasis, chronicity and bad response to therapy (Sacks et al., 1987; Kemp et al., 1992; Uyemura et al., 1993). The disease has been found in severely immunocompromised patients with T cell defects such as renal transplant recipients, patients with leukaemia and lymphoma, systemic lupus erythematosus, patients with AIDS and patients on corticosteroid therapy (Fernández Guerrero et al., 1987; Berenguer et al., 1998; Berenguer et al., 1989; World Health Organization, 1997; Berhe et al., 1995; Rosenthal et al., 2000; Portoles et al., 1994; Hussein et al., 2001).

Previous reports have documented some difficulties in the clinical diagnosis of visceral leishmaniasis in immunocompromised patients with emphasis on atypical manifestations and unusual anatomic involvement (Fernández Guerrero et al., 1987; Berenguer et al., 1989; Rosenthal et al., 2000; Górgolas et al., 1997). However, the manifestations of leishmaniasis in immunocompromised patients with AIDS seemed to be similar to those observed in immunocompetent patients without underlying conditions (Montalban et al., 1989; Pintado et al., 2001). In a recent report, Pintado et al. did not find differences in the clinical presentation of visceral leishmaniasis in AIDS, but HIV-infected patients had a lower frequency of splenomegaly than HIV-negative individuals (Pintado et al., 2001). In addition, the sensitivity of serologic studies by means of indirect immunoflorescense for *Leishmania* was significantly lower and the mortality rate higher in HIV-infected than in non-HIV-infected patients (Pintado et al., 2001).

In our review of the experience of a single institution dealing with immunocompromised patients with visceral leishmaniasis several points deserve discussion. Firstly, the clinical presentation and the manifestations of leishmaniasis were similar in both groups of patients. Fever, lymphadenopathy, enlargement of the liver and spleen, cytopenias and the enzymatic profiles were equally found in immunocompromised patients with and without AIDS. However, as shown by others we also noted that HIV-infected patients showed a lesser tendency to develop enlargement of the spleen than HIV-negative individuals (Pintado et al., 2001). This fact has been associated to a lack of proliferative response of macrophagic cells in patients with AIDS (Peters et al., 1990).

Our review also suggested that for diagnosis of leishmaniasis, serology may be less useful in patients with AIDS than in other immunocompromised patients. The reduced sensitivity of the indirect immunofluorescense test in patients with AIDS has been previously noted and the rate of positive serology in various studies have ranged from 22 to 57% (Berman, 1997; Rosenthal et al., 2000; Montalban et al., 1989; Pintado et al., 2001; Peters et al., 1990; López Vélez et al., 1999). The functional impairment of cell-mediated immunity due to HIV infection could result in the absence of an antibody response to *Leishmania* as has been shown for other infections (Lane et al., 1983; Biggar et al., 1987; Haas et al., 1990). On the other hand, serology seemed to be a

sensitive method for the diagnosis of leishmaniasis in other subsets of immunocompromised patients. In this study, all immunocompromised patients without AIDS had positive titers of antibodies to *Leishmania*. Others authors have also found positive serology in a variety of immunocompromised patients with visceral leishmaniasis (Berman, 1997; Fernández Guerrero et al., 1987; Berenguer et al., 1998; Badaró et al., 1986b; Kar, 1995). The lower yield of the bone marrow aspirate in HIV-infected patients in comparison with non-HIV-infected individuals observed in this study, could be explained by the hypoplasia and fibrotic changes of the bone marrow that are frequently found in patients with advanced AIDS (Benito et al., 1997).

The treatment of visceral leishmaniasis in immunocompromised patients is an unresolved question and our study has confirmed the poor results of conventional therapy in these individuals. Clinical failure and mortality was high and 40% of the survivors experienced one or more relapses. This is in frank opposition to results of treatment in immunocompetent hosts who can be cured with a single course of in most instances (Berman, 1997; Gradoni et al., 1995).

A variety of drugs have been used in the treatment of kala azar in immunocompromised patients. Amphotericin B, pentavalent antimony alone and in combination with aminosidine and interferon-gamma, combinations of allopurinol with azole compounds and high-dose liposomal amphotericin B have been used without improvement in prognosis (Peters et al., 1990; Górgolas et al., 1993; Raffi et al., 1995; Davidson and Russo, 1994). A prospective comparison of the efficacy of meglumine antimoniate with amphotericin B in patients with visceral leishmaniasis complicating AIDS did not show any difference in the rates of cure or relapses (Laguna et al., 1999). Miltefosine, a new oral drug recently introduced for the treatment of visceral leishmaniasis, should be assayed in immunocompromised patients (Sundar et al., 2002). This drug, that has been successfully used in Indian patients with kala azar, could be useful for the treatment or prevention of relapses in immunocompromised hosts. Certainly, new drugs for the treatment of visceral leishmaniasis in immunocompromised patients are urgently needed.

At the present time, partial reconstitution of the immune system by HAART in patients with AIDS and discontinuation or drastic reductions of the immunosuppressive drugs in patients with other conditions is of paramount importance to improve prognosis of visceral leishmaniasis in immunocompromised patients. In this series, only the patients who responded to HAART and those in whom corticosteroid therapy could be stopped or reduced did not develop relapses of leishmaniasis and achieved cure after discontinuation of antileishmanial therapy. Unfortunately, the mortality of the first episode of kala azar in immunocompromised patients is still very high and many patients developed severe complications before improvement of the immune functions can be achieved (Alvar et al., 1997; López Vélez et al., 1999).

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