Visceral Leishmaniasis in a patient with common variable immunodeficiency and Evans syndrome: clinical remarks

Patrizia Maio¹, Sebastiano Leone¹, Silvestro Volpe², Giuseppina dell’Aquila¹, Sergio Giglio¹, Mario Magliocca¹, Francesco Saverio Nigro¹, Piera Pacifico¹, Nicola Acone¹

¹Infectious Diseases Department, A.O.R.N. S.G. Moscati, Avellino, Italy; ²Hematology Department, A.O.R.N. S.G. Moscati, Avellino, Italy

Visceral Leishmaniasis (VL) is a potentially life-threatening disease caused by the protozoan Leishmania infantum which infects macrophages through the reticuloendothelial system. It is a vector-borne zoonosis transmitted by the bites of phlebotomine sandflies with canids as a common reservoir host and humans as incidental hosts (Gradoni et al., 2003). Sporadic transmission by blood transfusions or needle sharing among intravenous drug addicts is also described. Infections by Leishmania infantum are widespread in the Mediterranean basin and are endemic in Southern Italy due to the closeness of dogs, vectors and human habitations. In Italy two hundred cases per year and in the Campania region forty up to eighty cases per year are reported (Gradoni et al., 1996). In humans, the course of the illness depends on both the infectant parasite and the host’s immune status, being more severe in immunosuppressed patients such as HIV-infected subjects and transplant recipients or patients treated with corticosteroid/immunosuppressive drugs (Hernández-Pérez et al., 1999; World Health Organization, 1997). A favourable clinical outcome is determined by the TH1 immune response, with γIFN and IL-2 release (Kemp, 2000).

We describe a case of VL in a splenectomized patient with Common Variable Immunodeficiency (CVID) and Evans syndrome (autoimmune hemolytic anemia plus immune thrombocytopenia) (Evans et al., 1951). In 2007 a 46-year-old man was referred to our Infectious Diseases Department because of an irregular fever that had started 10 days before, with muscle weakness and no other relevant clinical symptom. In the past he developed autoimmune cytopenias, firstly immune thrombocytopenia (ITP) which required splenectomy, and then autoimmune hemolytic anemia (AIHA) treated with steroid therapy. Subsequently, he was found to have CVID and started cyclic intravenous immunoglobulin therapy carried on discontinuously. Some months before our clinical examination,
he had been admitted to hospital for pneumonia. At that time, laboratory investigations demonstrated anemia (with positive direct and indirect Coombs tests, decreased haptoglobin, rising unconjugated bilirubin, serum iron and LDH) and thrombocytopenia. The finding of ongoing AIHA plus thrombocytopenia, related to a history of ITP allowed the hematologist to diagnose Evans syndrome, just consisting of AIHA plus ITP. Therapy was carried on with prednisone for 8 weeks with dose tapering. Clinical conditions and blood tests improved but fever reappeared when the therapy stopped.

On admission, he was pale, asthenic, feverish (39°C) and complained of mild headache. On examination only mild hypotension and hepatomegaly were found. A complete blood count showed mild anemia, leucocytosis with lymphocytosis and severe thrombocytopenia. Red blood cell (RBC) count was 3.8x10⁶/µL, hemoglobin 9.1 g/dl, white blood cells (WBC) count was 13.1x10³/µL with 53% lymphocytes and 40% neutrophils, platelet (PLT) count was 4.9x10³/µL. Direct and indirect Coombs tests were positive. Liver biochemical tests showed moderately elevated values (AST 58 IU/L, ALT 105 IU/L, LDH 800 IU/L). Serum proteins, albumin and gammaglobulin value were reduced (5.08 g/dl, 2.9 g/dl, 0.4 g/dl, respectively). All immunoglobulin subclasses were reduced (IgA: 0.20 g/l, IgM: 0.34 g/l, IgG 5.70 g/l). The systemic inflammation indices were abnormal (CRP 65 mg/dl, ESR 42 mm/h, fibrinogen 525 mg/dl) and serum iron was 21 µg/dl. Renal function was normal. Blood culture for bacteria and mycetes, urine culture and stool culture were negative. HIV-RNA, CMV-DNA, EBV-DNA, HCV-RNA, HBV-DNA were absent in his blood. A bone marrow aspirate and biopsy demonstrated Leishmania amastigote, inside and outside the histiocytes. Serum Leishmania antibodies were detectable on IFAT, even though with low titre (1/80). Therapy with Liposomal Amphotericin B (LAB) was started, 4 mg/kg IV q24h on days 1-5, 10, 17, 24, 31, 38. On the tenth day PLT count further decreased to 4.3x10³/L and bleeding began from gums, nose and rectum. PLT transfusions were provided while LAB therapy was continued in accordance with the scheduling and the patient recovered with normal RBC and PLT count restored. CRP returned to normal value within 40 days. A second bone aspirate was performed 10 days after the therapy stopped, demonstrating lack of Leishmania. The patient has remained free of VL in the following ten months.

VL is a cause of "fever of unknown origin" (FUO) in immunocompetent subjects in variable percentages depending on geographic area, patients' age and diagnostic means available. In the Mediterranean basin VL is reported in 4.5% up to 17.1% of FUO (Pasic et al., 2006; Saltoglu et al., 2004). It has been also diagnosed, increasingly, in immunocompromised hosts, like splenectomized, transplanted, HIV-infected, immunosuppressive drug-treated patients and those suffering from hematological malignancies (Bada et al., 1979; Basset et al., 2005; Fernandez-Guerrero et al., 2004; Lozano et al., 1996; Pati et al., 1999; Pavone et al., 2008).

In immunocompetent hosts VL generally causes fever, splenomegaly, hepatomegaly, weight loss, pancytopenia-anemia, leukopenia, thrombocytopenia and hypergammaglobulinemia. Pancytopenia is caused partially by medullary parasitic infiltration and mainly by increased cellular destruction in the enlarged and congested spleen. Hypergammaglobulinemia, mostly IgG, results from polyclonal-B cell activation by Leishmania antigens (Berman, 1997). Serum Leishmania antibodies on IFAT (≥1/40) are found in more than 90% of the patients. Diagnosis is confirmed by Leishmania detection in bone marrow specimens with microscopic examination or, lately, molecular amplification assay (PCR). The protozoan can be also found in liver and spleen tissue or in peripheral blood. In immunocompromised hosts clinical signs and symptoms closely resemble those observed in immunocompetent patients but may be misleading or delayed, allowing a low index of suspicion and a misdiagnosis. Moreover, opportunistic coexistent or superimposed infections from bacteria and viruses, mostly CMV, can modify the clinical picture. To date, transplanted and HIV-infected patients have been the most widely studied among immunocompromised hosts (Antinori et al., 2008; Basset et al., 2005). Fever is the most common symptom. In a recent review concerning VL in immunocompromised hosts, fever was described in 94% of the transplanted and 81% of the HIV-infected patients, in comparison with 99% of immunocompetent patients. Splenomegaly was re-
ported in 75% and 74% respectively, vs. 98% of immunocompetent patients, hepatomegaly in 42% and 77% vs. 86% (Antinori et al., 2008). Among the laboratory findings, leucopenia was the most frequently observed abnormality (93% in transplanted and 90% in HIV-infected patients), followed by anemia (86% and 88%) and thrombocytopenia (85% and 79%). In one liver transplanted patient, Basset reported an isolated AST and ALT increase with no blood cell count abnormality (Basset et al., 2005). Leishmania serology was found positive more often in transplanted patients than in HIV-infected patients (92% vs. 48%) (Antinori et al., 2008). Direct microscopic examination of bone marrow smears was the most frequently used diagnostic procedure in immunocompromised hosts as well as in immunocompetent ones. Its sensitivity was higher in transplanted than in HIV-infected patients (98% vs. 81%), probably due to the bone marrow hypoplasia in advanced HIV disease subjects.

We describe a case of VL in a splenectomized patient with CVID and Evans syndrome, which hampered the diagnosis. CVID is the most prevalent of the primary immunodeficiency diseases and is often associated with infections, mostly of the respiratory tract, malignancies and autoimmune disorders. Evans syndrome is an autoimmune pathology consisting of AIHA plus ITP and has been reported in CVID patients (Chapel et al., 2008; Garcia-Munoz et al., 2008; Wang et al., 2005). Our patient showed anemia and thrombocytopenia associated with leucocytosis and lymphocytosis. The latter features must be related to the previous splenectomy. Anemia with positive Coombs test and thrombocytopenia could have been judged, at the beginning, as an expression of Evans syndrome’s relapse. In fact, the patient was Evans syndrome diagnosed on a previous hospital admission. Moreover, as expected, he showed hypogammaglobulinemia with IgA, IgM and IgG subclass deficiency because of his CVID (Schroeder et al., 2007). A bone marrow biopsy was performed because of suspected lymphoid malignancy. The bone marrow specimen disclosed no abnormality in any cell line but heavy infiltration of bone marrow by Leishmania amastigotes, thereby establishing the diagnosis. Anti-Leishmania serology was also positive (IFAT 1/80). The recovery of immunoglobulin production transiently or permanently following HCV and HIV infection, as well as humoral vaccination response have been reported in patients with CVID (Goldacker et al., 2007; Wright et al., 1987) indicating that this immunodeficiency is associated with potentially reversible defects in immunoregulatory factors and B cell systems. Leishmania antibody response in our patient could confirm this concept.

Through a careful review of the scientific literature, this is the first VL case associated with CVID in adult host, few cases of VL having been reported in patients with different kinds of hypogammaglobulinemia (Martin et al., 1996;
Mendoza et al., 1997; Voutsinas et al., 2001; Wright, 1959). Epidemiological and clinical features, therapy and outcome of two of these patients are reported in Table 1. The outcome in treated patients was good. Not many data on VL clinical course in CVID-affected patients are available. It is known that cell mediated immunity and not humoral immunity is the most important protective mechanism against Leishmania spp. A consistent principle is that healing and resistance to reinfection are associated with an intact T_h1 cell response essentially based on IFN- and IL-2 release as well as activation of macrophages able to kill intracellular amastigotes (Kemp, 2000). In patients with CVID the most common abnormality is defective antibody formation but numerous immune system abnormalities have been reported and both humoral and cell-mediated lymphocytic responses are affected. Moreover, our patient had been treated in the past with steroid therapy for Evans syndrome. Probably, this treatment played an additional favourable leading role in protozoan spread and disease development as corticosteroids inhibit T lymphocytes allowing an increasing susceptibility to infections, especially from intracellular pathogens like Leishmania (Boumpas et al., 1993; Stuck et al., 1989).

Therefore we had a patient who had undergone spleen removal, had CVID and received steroid therapy for autoimmune complications. So, we chose an anti-Leishmania strengthened therapeutic regimen, approved for immunosuppressed patients, consisting of LAB, 4 mg/kg IV q24h on days 1-5, 10, 17, 24, 31, 38 (Meyerhoff, 1999). This therapy proved to be well tolerated and effective. Clinical symptoms passed off within some days. RBC and PLT count normalized and CRP, initially remarkably raised, also reached the normal value within 40 days, confirming it is a reliable marker of the disease’s good course. In fact, CRP is an acute phase protein that proved of good prognostic value in VL (Bern et al., 2007). In a case control study in children with VL at different stages of the disease, Singh showed that during treatment, mean serum CRP levels were significantly higher in late responders than in early responders (p<0.001) (Singh et al., 1999). In another study, Gasim observed that plasma CRP was a serum simple marker useful to identify patients with a high risk of developing post kala-azar dermal leishmaniasis after treatment (Gasim et al., 2000). Unfortunately, there are no data in the literature on the role of PCR in the follow-up of VL in immunocompromised hosts.

Our patient has remained free of Leishmaniasis in the following ten months. We conclude that VL has to be taken into account for diagnosis of FUO in immunocompromised hosts, above all in endemic areas.

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