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Case Report
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Visceral Leishmaniasis in 2 Patients Treated With Lenalidomide and Dexamethasone: A Possible Correlation With Blunted Immune Response

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Immune defects related to multiple myeloma (MM) and immunosuppressive effects of novel antimyeloma agents (NA) are both responsible for an increasing rate of recurrent infections.[1-3] Leishmaniasis is caused by an intramacrophage protozoon of the genus Leishmania; it is transmitted by sandflies, presents in cutaneous, mucocutaneous, and visceral forms and primarily concerns immune-suppressed patients.[4,5] Data regarding the incidence of opportunistic infections, including leishmaniasis in MM patients treated with NA, are limited.[6-9] Herein, we report 2 cases of MM patients who developed visceral leishmaniasis (VL) during therapy with NA and discuss potential relations of VL with immune surveillance disorders.

A 58-year-old female patient of Greek origin was diagnosed in December 2003 with Immunoglobulin G kappa, standard risk MM, stage 3 per International Staging System. She presented with 25% bone marrow clonal plasma cells (BMcPCs), anemia (hemoglobin [Hb]: 7.8 g/dL), normal renal function, and L5 vertebral collapse. She was treated with 2 cycles of vincristine-adriamycin-dexamethasone without response and subsequently, she received four 28-day cycles of thalidomide/dexamethasone, followed by autologous peripheral stem cell transplantation (ASCT) after which she achieved complete response. Four years later, she experienced clinical relapse with anemia, lytic lesions, 30% BMcPCs, and persisting low grade fever. White blood count (WBC) and platelets were normal, blood and sputum cultures, polymerase chain reaction (PCR) for cytomegalovirus (CMV) and Epstein-Barr virus (EBV) and serological tests for aspergillus antigen and toxoplasma, were all negative, therefore, infection was ruled out and fever was attributed to MM. She received three 21-day cycles of bortezomib-dexamethasone; however, treatment was suspended due to peripheral neuropathy and hypotension; lenalidomide (25 mg/d for 21 d) and dexamethasone (20 mg/wk) (LenDex) was subsequently administered leading to partial response (PR) after 6, 28-day cycles. After 12 cycles, she presented with persistent high fever (39°C) and splenomegaly. Laboratory tests revealed pancytopenia (WBC: 1400/μL, neutrophils: 1.100/μL, lymphocytes: 200/μL, Hb: 7.8 g/dL, platelets: 51.000/μL) and elevated C-reactive protein; renal and liver function, echocardiogram and chest imaging were normal. Repeated blood cultures for common pathogens, sputum cultures for mycobacterium tuberculosis, serum test for aspergillus antigen, PCR for CMV and EBV and serological tests for toxoplasma, brucellosis, histoplasma and cryptococcus, were negative. Bone marrow aspiration (BMA) displayed BMcPCs 2% and numerous intracellular amastigotes, indicating VL (Figure 1A), confirmed by subsequent serological and PCR tests. Liposomal amphotericin B (L-AB) was initiated (3 mg/kg/d; days 1-5, 15, and 22)[10] leading to hematological and clinical recovery within a few weeks (WBC: 4100/μL, neutrophils: 2700/μL, lymphocytes: 1300/μL, Hb: 11.3 g/dL, platelets: 137 000/μL). We decided to withhold antimyeloma therapy, considering that she was not keen to restart it and that MM remained in PR. Three months later, she was admitted with intermittent high fever (up to 39.8°C), anemia (Hb: 6.6 g/dL), and thrombocytopenia (platelets: 10 000/μL); BMA was normal with no evidence of MM progression or VL, but serological and PCR tests confirmed Leishmania recurrence. She was retreated with L-AB (3 mg/kg for 10 d), leading to rapid recovery; she remained off myeloma therapy, in PR and 6 months later, she presented with fever and generalized bone pain. After ruling out infections including VL relapse, fever was eventually attributed to disease progression; she refused any antimyeloma therapy and was treated palliatively until death, 1 month later.

A 62-year-old male patient of Greek origin was diagnosed in August 2012 with Immunoglobulin G kappa standard risk MM, stage 2 per International Staging System, presented with anemia (Hb: 6.8 g/dL) and moderate renal dysfunction (estimated glomerular filtration rate: 59 mL/min) due to diabetic nephropathy. He was treated upfront with bortezomib-cyclophosphamide-dexamethasone, for 4 monthly cycles without response and he subsequently received LenDex (lenalidomide: 10 mg/d, dexamethasone: 16 mg/wk for the first cycle and 8 mg thereafter, due to uncontrolled diabetes). After six 28-day cycles he achieved PR. He did not undergo ASCT due to history of coronary heart disease and received 20 mg/wk dexamethasone, as maintenance; 25 months later, he progressed (BMcPCs: 34%, increase of serum M-protein, Hb: 9.2 g/dL). He was retreated with LenDex in the previous dose and he achieved PR.

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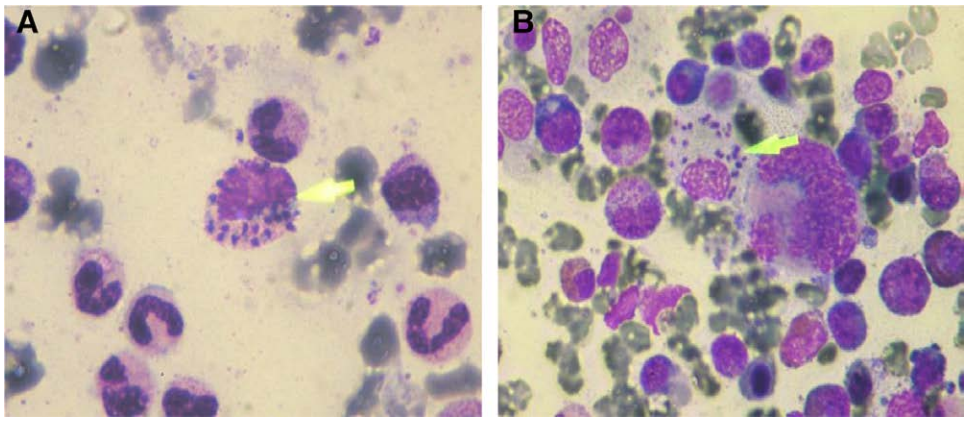

Figure 1. Wright–Giemsa stain of bone marrow smear. Macrophages with intracellular amastigote forms of Leishmania species in the first (A) and the second patient (B), respectively.

Table 1

Alterations of Tregs and Lymphocyte Subsets in Patient No. 2 During VL Infection and Median Levels of the Same Subsets in 18 MM Patients and 20 Healthy Controls.

Variables	VL (n = 1) Diagnosis	VL (n = 1) Recession	VL (n = 1) A Year After Recession	HC[11] (n = 20) Median (95% CI) %	MM Patients Treated With LenDex[11] (n = 18) Baseline Median (95% CI) %	MM Patients Treated With LenDex[11] (n = 18) Response Median (95% CI) %
Tregs%	0.8	0.0075	0.076	0.0015 (0.001-0.0026)	0.2 (0.02-5.7)	0.02 (0.0-0.12)
CD4/μL	385/μL	585/μL	1207/μL	949/μL (784-1315)	589/μL(451-747)	432/μL (333-681)
CD4%	35	39	37	47	34	34
CD8/μL	451/μL	645/μL	1455/μL	605/μL (465-700)	573/μL (371-794)	507/μL (332-730)
CD8%	41	43	44	25	31.8	32.6
CD4/CD8 ratio	0.85	0.84	0.8	1.9 (0.85-2.48)	1.1 (0.33-5.85)	1.01(0.31-4.3)
CD19/μL	14.3	15	42	204 (164-306)	87 (48-118)	23 (15-140)
CD19%	1.3	1	1	9.2	4.3	2.3
NK/μL	187/μL	210/μL	422/μL	210/μL(176-286)	12/μL (216-463)	283/μL (174-420)
NK%	17	14	13	9.6	22	17
NKL/μL	154/μL	345/μL	791/μL	84/μL (47-158)	86/μL (52-202)	62/μL (35-183)
NKL %	14	23	24	4	5.5	4.5

LenDex refers to 18 myeloma patients treated with LenDex in our previous study (adapted from Hadjiaggelidou et al[11]).

CD = classification determinant, CI = confidence interval, HC = healthy controls, LenDex = lenalidomide dexamethasone, MM = multiple myeloma, NK = natural killers, NKL = NK-like T cells, Tregs = T regulatory cells, VL = visceral leishmaniasis.

A year later while on LenDex therapy, he presented with high fever (39.8°C) and no other clinical findings. Laboratory tests revealed pancytopenia (WBC: 3200/μL, neutrophils: 1700/μL, lymphocytes: 1100/μL, platelets: 18 000/μL, Hb: 10.9 g/dL), elevated C-reactive protein (11.8 mg/dL), abnormal liver enzymes, and estimated glomerular filtration rate 48 mL/min. Repeated cultures for common pathogens and *Mycobacterium tuberculosis*, aspergillus antigen test and PCR for CMV and EBV were negative. Chest and heart imaging were normal; BMA revealed the presence of intracellular amastigotes (Figure 1B) whereas BMcPCs were 6%. Subsequent serological and PCR tests confirmed the diagnosis of VL; L-AB was administered in the recommended dose for 10 days; after a month, PCR test was negative, amastigotes were not observed in the bone marrow and blood counts returned to normal (WBC: 4300/μL, neutrophils: 2600/μL, lymphocytes: 1500/μL, Hb: 12.3 g/dL, platelets: 142 000/μL). On clinical recovery, antimyeloma treatment with LenDex was restarted and maintenance with L-AB was continued monthly, as recommended by an infectious disease specialist, without any side-effects or drug interactions. In a recent re-evaluation, the patient remained in sustained PR, without any evidence of VL (negative PCR test for Leishmaniasis, no amastigotes in the BMA). Using multiparametric flow cytometry analysis, we detected regulatory T cells (Tregs; CD4 + CD25highCD127neg/lowFoxP3 + cells quantified from CD4 + T cells) and lymphocyte subpopulations (CD4, CD8, CD19, natural killer [NK], NK-like T cells [NKL]) in peripheral blood at 3 different timepoints, that is, at VL onset, recession and a year after and were compared with previously published data of ours (Table 1).[11]

Patients with MM are susceptible to unusual infections, because of severe immune impairment, including decrease uninvolved immunoglobulins (immunoparesis), reductions in CD4 T and CD19 B populations and abnormalities in T, NK, NKL, and dendritic populations.[3-5] Leishmaniasis is a rare parasitic infection mainly presented as reactivation of a dormant infection, when the individual is residing in a VL-endemic area and, less frequently, as de novo or iatrogenic infection via a contaminated blood product.[3-5] Furthermore, immunosuppressive conditions, such as HIV infection or Hodgkin lymphoma are associated with increased risk of VL reactivation.[4] In this report, VL was considered as a reactivation of previous infection, as both patients were resided in an endemic country and could have been exposed to phlebotomus sand-fly bite. Apart from active

myeloma at VL onset, both had received long-term steroids and one of them underwent ASCT, which have been related with VL.[4,5] Regarding NA impact, 2 case reports described 2 patients treated with LenDex after induction plus ASCT[6,7] who developed VL. Piro et al[8] described a case of VL during second-line treatment with bortezomib-dexamethasone. Ziogas et al[9] published 2 cases of VL treated with LenDex plus daratumumab and elotuzumab, respectively, for relapsed myeloma; none of the reports, has shown a direct association between treatments and VL infection. Regarding our cases, the first patient was severely immunocompromised due to relapsed MM after ASCT[6,7] and long-term use of dexamethasone,[3] which probably led to severe lymphopenia, contributing to parasite dissemination. Of note, immunoparesis presented at diagnosis, was resolved after ASCT, reappeared at the time of MM relapse, and remained throughout VL course. In addition, coadministration of lenalidomide with dexamethasone may alleviate the well-recognized immunostimulatory effects of lenalidomide.[12] The above immunosuppressive factors could be related with ineffective control of the initial episode of leishmaniasis and subsequent recurrence.

Likewise, the second patient developed VL after treatment with LenDex for 1 year; in this case, the dose of dexamethasone was low probably explaining the normal absolute lymphocyte number at VL diagnosis. Immunoparesis was not evident in this patient, either at MM diagnosis or during course of MM; CD4 T and CD19 B cells and CD4/CD8 ratio were lower compared to controls (Table 1); those immune defects may contributed to the reactivation of latent leishmaniasis. Furthermore, normal CD8 levels documented at VL diagnosis probably facilitated disease control, along with the appropriate therapy; CD8 levels further increased after VL recession and 1 year later, confirming observations demonstrating that, in healed VL individuals, the elevated level of CD8 + T cell confer resistance to Leishmania reinfection.[13] Moreover, Tregs were strikingly higher compared with controls and MM patients treated similarly,[11] suggesting a correlation of blunted immune responses with parasite dissemination and suboptimal response to LenDex[11-14]; Tregs' reduction after VL recession indicates that immune balance may effect VL outcome.[13,14] The percentage of NK cells at VL diagnosis was higher compared with controls but similar to that of MM patients treated with LenDex (Table 1)[11]; increased levels of NK cells in peripheral blood of MM patients may indicate a preserved immune function[13]; high levels of NK cells probably contributed to the rapid control of VL after treatment with L-AB. Furthermore, peripheral blood NKL cells were higher compared with both controls and MM patients (Table 1), probably reflecting NKL bone marrow enrichment during VL, as described[15]; NKL cells remained high after VL recession and 1 year after, confirming that this subset of T cells play an important role in immune restoration and probably, durable control of VL.[13,14]

In conclusion, patients with active MM are susceptible in developing VL due to dysregulated immune responses. Furthermore, therapy with NA and particularly prolonged administration of lenalidomide, in combination with dexamethasone could promote parasite dissemination, mainly in endemic areas, therefore VL testing should be included in the investigation of fever and pancytopenia presented in MM patients, treated with such combinations. Patients with MM and VL display alterations in lymphocyte subpopulations which may interact with parasite immune-mediated mechanisms and affect VL outcome. Therefore, monitoring of immune cells may offer information about VL course. Our observations deserve further evaluation.

References

1. Pratt G, Goodyear O, Moss P. Immunodeficiency and immunotherapy in multiple myeloma. *Br J Haematol.* 2007;138:563–579.
2. Blimark C, Holmberg E, Mellqvist UH, et al. Multiple myeloma and infections: a population-based study on 9253 multiple myeloma patients. *Haematologica.* 2015;100:107–113.
3. Nucci M, Anaissie E. Infections in patients with multiple myeloma in the era of high-dose therapy and novel agents. *Clin Infect Dis.* 2009;49:1211–1225.
4. Saporito L, Giammanco GM, De Grazia S, Colomba C. Visceral leishmaniasis: host–parasite interactions and clinical presentation in the immunocompetent and in the immunocompromised host. *Int J Infectious Dis*. 2013;17:572
5. van Griensven J, Carrillo E, López-Vélez R, et al. Leishmaniasis in immunosuppressed individuals. *Clin Microbiol Infect.* 2014;20:286–299.
6. Radujkovic A, Hundemer M, Eisenbach C, et al. Visceral leishmaniasis in a patient with relapsed multiple myeloma receiving high-dose melphalan and autologous stem cell transplant. *Leuk Lymphoma.* 2014;55:2967–2969.
7. Alahmadi M, Chin-Yee IH, Gupta K, Rizkalla K, Hsia CC. Leishmaniasis involving the bone marrow of a patient with multiple myeloma. *Ann Hematol*. 2015;91:1827
8. Piro E, Kropp M, Cantaffa R, et al. Visceral leishmaniasis infection in a refractory multiple myeloma patient treated with bortezomib. *Ann Hematol.* 2012;91:1827–1828.
9. Ziogas DC, Terpos E, Gavriatopoulou M, et al. Coexistence of leishmaniasis and multiple myeloma in the era of monoclonal antibody (anti-CD38 or anti-SLAMF7) containing triplets: one shared story of two exceptional cases. *Leuk Lymphoma.* 2018;59:983–987.
10. Sundar S, Chakravarty J. An update on pharmacotherapy for leishmaniasis. *Expert Opin Pharmacother.* 2015;16:237–252.
11. Hadjiaggelidou C, Mandala E, Terpos E, et al. Evaluation of regulatory T cells (Tregs) alterations in patients with multiple myeloma treated with bortezomib or lenalidomide plus dexamethasone: correlations with treatment outcome. *Ann Hematol.* 2019;98:1457–1466.
12. Gandhi AK, Kang J, Capone L, et al. Dexamethasone synergizes with lenalidomide to inhibit multiple myeloma tumor growth, but reduces lenalidomide-induced immunomodulation of T and NK cell function. *Curr Cancer Drug Targets.* 2010;10:155–167.
13. Jawed JJ, Dutta S, Majumdar S. Functional aspects of T cell diversity in visceral leishmaniasis *Biomed Pharmacother*. 2019;117:109098.
14. Gupta G, Oghumu S, Satoskar AR. Mechanisms of immune invasion in Leismaniasis. *Adv Appl Microbiol*. 2013;82:155.
15. Rai AK, Thakur CP, Seth T, et al. Enrichment of invariant natural killer T cells in the bone marrow of visceral leishmaniasis patients. *Parasite Immunol.* 2011;33:688–691.