Epstein-Barr Virus and Human Herpesvirus 8 Coinfection and Concomitant Extranodal Nasal-Type NK/T Cell Lymphoma and Castleman Disease: Case Report

Claire Larroche,1 Félix Agbalika,2 Hélène Poirel,3 Antoine Martin,4 Agnès Gautheret-Dejean,5 Martine Raphael,3 and Olivier Lortholary1

Departments of 1Internal Medicine and Infectious Disease and 3Biological Hematology and 4Pathology Laboratory, Hôpital Avicenne, Université Paris–Nord, Bobigny, 2Department of Virology, Hôpital Saint-Louis, and 5Department of Virology, Hôpital Pitié-Salpêtrière, Paris, France

We describe a 36-year-old man uninfected with human immunodeficiency virus who had confirmed concurrent infection with Epstein-Barr virus (EBV) and human herpesvirus 8 (HHV-8) and their respective lymphoproliferative manifestations, nasal-type NK/T cell lymphoma and Castleman disease. Antibodies to HHV-8 and EBV DNA were found in plasma and peripheral blood mononuclear cells. An EBV-positive nasal-type NK/T cell lymphoma infiltrated the splenic red pulp, whereas the white pulp contained HHV-8–positive plasmablasts, as found in Castleman disease.

Multicentric Castleman disease (CD), primary effusion lymphoma, and Kaposi sarcoma are 3 diseases associated with human herpesvirus (HHV)-8 infection that occur in HIV-infected and HIV-uninfected patients. HHV-8 and Epstein-Barr virus (EBV) coinfection is well known in immunosuppressed patients, such as HIV-positive persons with primary effusion lymphoma [1], and was recently reported anecdotally in an infant with primary immunodeficiency [2]. Herein we describe a case of EBV and HHV-8 coinfection with the concomitant lymphoproliferative disorders, nasal-type NK/T cell lymphoma and CD, respectively, in an HIV-uninfected man.

Received 2 October 2002; accepted 12 December 2002; electronically published 22 April 2003.


Reprints or correspondence: Dr. Claire Larroche, Dept. of Internal Medicine and Infectious Diseases, Hôpital Avicenne, 125 Rue de Stalingrad, 93009 Bobigny Cedex, France (claire.larroche@avc.ap-hop-paris.fr).

Clinical Infectious Diseases 2003;36:e107–10
© 2003 by the Infectious Diseases Society of America. All rights reserved.
1058-4838/2003/3609-00E2$15.00

BRIEF REPORT

Case report. A 36-year-old man from the Comoro Islands was hospitalized at Avicenne Hospital (Bobigny, France) for fever, chills, and weight loss of 1 month’s duration. Physical examination revealed marked hepatosplenomegaly and jaundice. Blood tests revealed pancytopenia with profound lymphocytopenia (leukocyte count, 2900 cells/mm3, with 250 lymphocytes/mm3 and a CD4:CD8 ratio of 0.76; hemoglobin level, 8 g/dL; and platelet count, 95,000 cells/mm3), inflammatory syndrome (fibrinogen level, 6.2 g/L, and C-reactive protein level, 139.5 mg/L), hepatic dysfunction (aspartate aminotransferase level, 309 IU/L; alanine aminotransferase level, 273 IU/L; and total bilirubin, 28.6 µM), hypertriglyceridemia (triglyceride level, 6.08 mM; range, 0.50–1.67 mM), a high serum ferritin level (27,427 ng/mL; range, 30–280 ng/mL), and hyponatremia (Na concentration, 128 mM). Bone marrow aspirate and liver biopsy revealed pronounced hemophagocytosis.

The patient tested negative for HIV (by serological test and PCR) and for human T cell lymphotropic virus 1 (by serological test). No cytomegalovirus, HHV-6, or HHV-7 replication was detected. We detected EBV reactivation (IgG and IgM antibodies to viral capsid antigen, IgM to early antigen and IgG to nuclear antigen), and plasma and PBMCs were strongly positive for EBV DNA (6.69 log10 copies/mL and 4.40 log10 copies/mL, respectively) and HHV-8 DNA (7.2 log10 copies/mL and 7.08 log10 copies/mL, respectively). Anti-HHV-8 latent and lytic antibodies were also detected. Other causes of hemophagocytic syndrome were excluded. Intravenous immunoglobulin therapy had no effect. The patient’s clinical status worsened. Because of severe spleen involvement, a splenectomy was performed, and intravenous methylprednisolone was prescribed. However, the patient’s condition deteriorated rapidly, and he died of multiple-organ failure.

Methods. For analysis of EBV and HHV-8, isolates were subjected to molecular analysis with quantitative PCR. We analyzed a sample of DNA (1 µg) extracted from PBMC and plasma fractions by means of the QIAamp Blood Kit (Quiagen) and used PCR β-globin amplification of DNA samples as controls. We used the Taqman-based technique (Biosystem) of Kennedy et al. [3] to detect EBV DNA, as described by Kimura et al. [4], and to detect HHV-8 DNA, as described elsewhere [5]. Virus load was estimated from standard curves established from dilutions of DNA from the Namalwa (Burkitt) cell line (for EBV) and diluted plasmid controls (for HHV-8). Calibration copy numbers ranged from 1015 to 106 for 50 µL (r = 0.98–0.99).

Standard histological analyses were performed by 2 pathol-
Figure 1. Photomicrographs showing extranodal nasal-type NK/T cell lymphoma in the splenic red pulp. Immunohistological labeling revealed a sinus containing a dense infiltrate of neoplastic cells that were cytoplasm CD3ε positive (A) and had membranes reactive with CD8 (B) and TIA1 (C). Epstein-Barr virus (EBV) in situ hybridization revealed that most of the neoplastic cells were positive for EBV-encoded small RNA (D).

logists. Immunophenotyping was performed with paraffin-embedded tissue samples taken from the spleen, liver, and bone marrow with use of monoclonal antibodies directed against an HHV-8 latent nuclear antigen encoded by viral open reading frame 73 to detect HHV-8 in tissue samples [6]. EBV was detected by means of in situ hybridization with fluorescein isothiocyanate-labeled, EBV-encoded small RNA (EBER) 1 + 2-specific oligonucleotides (EBER in situ hybridization [ISH]; Dako), according to the manufacturer's instructions [7].

Results and discussion. Histological examination of splenic red pulp specimens identified a dense infiltration of a mixture of medium-sized and large lymphoid cells with irregular nuclei. These cells, all of which were EBER positive, had an immunophenotype consistent with nasal-type NK/T cell
Figure 2. Photomicrographs showing evidence of human herpesvirus (HHV)-8–positive Castleman disease in the splenic white pulp. A, Section showing a hyalinized germinal center surrounded by a broad concentric mantle of small lymphocytes, a finding consistent with the diagnosis of CD. B, At high magnification, small lymphocytes, mature plasma cells, and plasmablasts (arrow) can be seen in the mantle zone. C, Lymphocytes and plasmablasts were labeled by in situ hybridization with the HHV-8 latent nuclear antigen 1–specific monoclonal antibody.

lymphoma: they were CD3e-cytoplasm positive and demonstrated CD8, CD16, CD56, and TIA1 cell-membrane expression (figure 1), but they were negative for cell-surface CD5 and CD3 expression. Rare individual lymphoma cells were detected by EBER ISH of the bone marrow and liver sections. T cell-receptor genes were in germ-line configuration in PBMCs and in the spleen specimen. Hyalinized germinal centers surrounded by a broad concentric mantle of small lymphocytes, mature plasma cells and plasmablasts were seen in splenic white-pulp follicles and in a hilar splenic lymph node specimen,
a finding that is consistent with the diagnosis of CD. ISH studies to detect latent nuclear antigen 1 revealed HHV-8–positive lymphocytes and plasmablasts in the mantle zone (figure 2). A few mature plasma cells were found in the bone marrow and liver, but CD was diagnosed only on the basis of findings from the spleen. No HHV-8–positive cells were seen in the liver and bone marrow specimens. Hemophagocytosis was prominent in the bone marrow, spleen, and liver specimens.

The most striking point in this case is the simultaneous lymphoproliferative manifestations of infection with 2 herpesviruses in an HIV-negative patient: extranodal nasal-type NK/T cell lymphoma due to EBV infection and CD due to HHV-8 infection in the same organ. The coinfection might be a coincidence, because this man originated from an area (the Comoro Islands) where HHV-8 is endemic.

The simultaneous presence of EBV and HHV-8 DNA sequences in lymphoid tissues was recently described in an HIV-negative infant with primary immunodeficiency who did not have lymphoma or CD [2]. Also pertinent is a recent report of concomitant occurrence of EBV-positive Hodgkin lymphoma and HHV-8–positive CD in the same lymph node [8]. In our patient, the EBV-associated nasal-type NK/T cell lymphoma was principally located in the spleen, which is indeed a surprising and extremely rare observation [9]. Of note, it is known that EBV can establish type 2 latency in cell lines derived from patients with this type of lymphoma [10]. However, although the EBV-transforming latent membrane protein 1 is clearly implicated in lymphomagenesis, other transforming events seem to be necessary [11]. HHV-8 contains a battery of cellular gene homologues that encode proteins involved in signal transduction, cell cycle regulation, inhibition of apoptosis, and/or immune modulation [12]. HHV-8 may act indirectly through the release of growth factors targeting stroma cells [13]. Therefore, our hypothesis that HHV-8 induced paracrine stimulation, which has been advanced with respect to Kaposi sarcoma [14], might explain the simultaneous occurrence of CD and NK/T cell lymphoma in the same lymphoid organ.

References