Transient Monoclonal Gammopathy Associated With Cytomegalovirus Infection

By Helen Vodopick, Stuart J. Chaskes, Alan Solomon, and John A. Stewart

A transient monoclonal gammopathy, IgA-lambda light chain, associated with a cytomegalovirus (CMV) infection occurred in a patient with acute leukemia in remission. Evidence of CMV infection was confirmed by indirect hemagglutination titers, initially being 1:4096 and gradually falling to 1:64. During the infection, serum and salivary IgA levels rose to 2450 mg/100 ml and 72 mg/100 ml, respectively. Bence Jones proteinuria, lambda type, (0.1 g/24 hr) occurred during the period of excessive immunoglobulin synthesis. Antibody activity against CMV was detected in serum IgA and IgG. Also unusual were the histologic changes indistinguishable from malignant lymphoma seen in a cervical lymph node. After withholding for 1 wk the immunosuppressive drug (methotrexate) she had been receiving, symptoms and signs of the CMV infection and of the monoclonal gammopathy disappeared without recurrence after maintenance methotrexate was resumed.

UNUSUAL OPPORTUNISTIC organisms, such as fungal, protozoan, and viral agents, often supplant the usual infective bacteria in patients receiving intensive chemotherapy. Once recognized predominantly in premature infants, cytomegalovirus (CMV) has found a suitable host in the patient receiving immunosuppressive drugs for organ transplant or for leukemia. Such patients are thought to be more susceptible because of deficiency of immunoglobulin formation and/or cellular immunity.

Here we describe a patient with acute leukemia in remission, who produced excessive immunoglobulins while she was taking an immunosuppressive drug. A monoclonal IgA gammopathy and Bence Jones proteinuria, related temporally to her CMV infection, disappeared when the infection subsided.

MATERIALS AND METHODS

Serum proteins were separated by electrophoresis in a Beckman Microzone Cell on cellulose acetate membranes, using veronal buffer, pH 8.6 and ionic strength of 0.075. Proteins were stained with Ponceau S, and the percentage of proteins in each fraction was determined with the aid of an Analytrol Scanner and Integrator. Immunoelectrophoresis of serum, saliva (30-fold concentrate), and urine specimens was performed by the method of Scheidegger.1 Monospecific antisera to immunoglobulins G, A, M, and D, and to kappa and lambda Bence Jones proteins were prepared in our laboratory (University of Tennessee Memorial Research Center).

Serum immunoglobulin and C-reactive protein concentrations were measured by the single radial immunodiffusion technique of Mancini et al.2 Briefly, assay plates were prepared using commercially available heavy-chain specific antisera or C-reactive protein antiserum (Hyland Laboratory, Los Angeles, Calif.). Precipitation diameters were read after 48 hr, and the immuno-
globulin or C-reactive protein concentration in mg per 100 ml serum was determined using a
standard curve prepared from precipitation diameters of five concurrently run immunoglobulin
(Hyland Laboratory) or C-reactive protein (Behring Diagnostics, Somerville, N.J.) standards.
Salivary IgA concentrations, estimated by single radial immunodiffusion with a monoclonal
7S IgA standard, were multiplied by 3 in order to correct for the smaller diffusion of the 11S
IgA found in saliva.1 Salivary IgG and IgM concentrations were also measured by the single
radial immunodiffusion technique. Antibody titers to cytomegalovirus were determined by means
of indirect hemagglutination according to the method of Bernstein and Stewart.2

Case Report
A 13-yr-old girl, first seen in December 1969, was found to have acute lymphoblastic leukemia.
After a stormy initial course, a remission of her leukemia was induced with vincristine sulfate
and prednisone. In February 1970 a severe febrile illness, clinically and radiologically consistent
with Pneumocystis carinii, responded to pentamidine isethionate. After her recovery from this ill-
ness, remission of her leukemia was maintained with biweekly oral doses and monthly intrathe-
cal injections of methotrexate. Although no signs of leukemia were found in the marrow or in the
central nervous system, her spleen remained palpable.
In May 1971, she noted painless lymphadenopathy in the left cervical chain. Slight tonsillar
enlargement was also noted. Penicillin was given for Staphylococcus aureus found on culture of
throat secretions. In spite of antibiotic therapy, the tonsils enlarged further, a pseudomembran-
branous tonsillar exudate appeared on them, and the submandibular salivary glands became
visibly enlarged. The chest x-ray remained normal.
Because of no response to an 11-day course of penicillin, administration of tetracycline was
begun. Her temperature rose to 40°C, but she appeared less ill with this fever than one might
expect. Her blood count became abnormal with significant pancytopenia (Fig. 1). The hematocrit
fell from 41% to 28%, the platelet count dropped from 167,500 to 23,200, and the white blood cell
count increased from 3000 to 12,400 but returned to 3900 when the hemoglobin and platelet
count reached their nadir. A differential count during this leukocytosis showed 93% segmented
neutrophils, 4% lymphocytes, and 3% monocytes. Marrow aspirated during the phase of pancyto-
penia showed increased cellularity with granulocytic hyperplasia; the myeloid:erythroid ratio

Fig. 1. Clinical signs and laboratory findings during the patient's illness.
was 6. Megakaryocytes were normal in number, but only a few plasma cells were seen. No morphologic evidence of relapse of lymphoblastic leukemia was found in the marrows obtained during this acute illness. Cold agglutination titer, direct Coombs' test, and heterophil-antibody agglutination titer were negative. Numerous cultures, including blood and marrow, were bacteriologically sterile, while culture of the tonsillar pseudomembrane grew out normal flora. In spite of the administration of several other antibiotics, her clinical picture did not change. However, after methotrexate was withheld for 1 wk, the blood count returned to normal, fever abated, and the tonsillar, salivary, and lymph node enlargement melted away in 3 days. But as the quantitative hematologic values returned to normal, the white cell differential count revealed a reversal of the granulocyte to lymphocyte ratio with pronounced lymphocytosis (absolute lymphocyte count 12,100). The majority of the lymphocytes were notched and had the morphologic features of lymphosarcoma cells. Slight lymphocytosis persisted for 1 mo after her acute illness subsided.

Except for a leukemic relapse, converted to remission by chemotherapy, the patient has continued to be in good health without recurrence of CMV infection or evidence of lymphoreticular disease.

Special Studies

During the course of the patient's acute febrile illness a reversal in the serum albumin and globulin values (1.8 and 3.2 g/100 ml, respectively) occurred. Examination of a serum specimen obtained on June 28 by cellulose acetate electrophoresis disclosed an anomalous protein component migrating in the β-globulin region (Fig. 2). This component, not evident in the electropherogram obtained prior to the febrile illness (June 11), was identified as an IgA (lambda type) protein by immunoelectrophoresis. The protein content of a 24-hr urine specimen was 0.1 g; immunoelectrophoretic analysis disclosed the presence of free lambda light chains, i.e., a lambda Bence Jones protein.

We performed serial measurements of the patient's serum and salivary immunoglobulin concentrations, and measurements of complement fixation (CF) and indirect hemagglutination (IHA) serum antibody to CMV and to herpes virus. As seen in Fig. 3, the serum IgA concentration gradually increased to a
Fig. 3. Temporal comparison of IHA and CF antibody titers to CMV, of salivary IgA concentrations, of serum immunoglobulin concentrations, and of C-reactive protein. The immunoglobulin levels returned to normal much sooner than the CMV antibody titer. Bence Jones protein was found in the urine on July 23. Increased amount of IgG was found in March when her leukemia relapsed.

The IHA titer to CMV, 1:4096 on the initial serum specimen examined (June 11), decreased progressively (Fig. 3), a finding compatible with recovery from an acute CMV infection. As the IHA titer fell, the CF titer to CMV gradually rose during the patient’s convalescence. Antibody titers to herpes virus were consistently less than 1:8.

To determine whether this immunoglobulin represented antibody to CMV, we separated the serum proteins by zone electrophoresis on a Pevikon block. The fact that IHA was detected in fractions eluted from the cathodal and anodal portions of the γ-globulin region of the block was evidence that the antibody activity against CMV was not confined exclusively to the monoclonal immunoglobulin. These two fractions contained IgG and IgG + IgA proteins, respectively.

During the acute illness, a cervical lymph node biopsy was performed; cultures of the node for bacteria and fungi were negative. Histologic examination revealed the normal node architecture to be replaced by masses of mononuclear
cells characterized by abundant pale-staining cytoplasm with a vesicular rounded nucleus containing a prominent eosinophilic nucleolus (Fig. 4). Some of the cells were binucleated and resembled Reed-Sternberg cells. Lymphocytic nodules were absent, and the capsule of the node was infiltrated by lymphocytes. Few plasma cells were present. These morphologic findings were interpreted as those of a poorly differentiated neoplasm, consistent with either reticulum cell sarcoma, leukemia, or a metastatic malignancy. On touch preparation immature lymphoid cells with prominent nucleoli resembled lymphocytes undergoing blastogenic transformation. Examination of the biopsy specimen by electron microscopy revealed many large cells with a vesicular nucleus and a conspicuous intranuclear body thought to be a nucleolus. No viral inclusions were seen.

DISCUSSION

The incidence of CMV infection has risen sharply in children with acute leukemia whose survival has been prolonged by antileukemic drugs and by the administration of blood. Both types of therapies may enhance the chances of developing CMV infection. Transmission of this apparently ubiquitous organism is thought to occur by many routes, a major one being transfusion of whole blood from asymptomatic carriers. The perplexing question arises: How do presumably healthy persons become asymptomatic carriers? In one survey of healthy blood donors, antibodies to CMV were found in 65%, and viremia in 5%. The finding of seroconversion to CMV in patients who receive blood transfusions suggests transmission of the virus by this route. Therefore, if healthy persons can be carriers, even patients who have not been transfused could also harbor the virus which might be activated by changes in their immune status.

Our patient had received 2 U of fresh whole blood 2 yr previously (when the diagnosis of leukemia was first established) but none thereafter. Although the source of her CMV is uncertain, the serologic findings of an initial high titer against CMV and a subsequent decrease in the titer of IHA support the diagnosis of a recent and acute CMV infection. Also, our patient’s clinical course and hematologic changes, especially the numerous circulating atypical lymphocytes, resemble the syndrome, CMV mononucleosis, which mimics infectious mononucleosis without positive heterophil agglutination and Paul-Bunnell tests.
An unusual feature of CMV mononucleosis is lymphadenopathy,\textsuperscript{10} which explains the infrequency of node biopsy in this disease. However, in tissues including lymph nodes of patients with CMV disease, pathognomonic intranuclear inclusion bodies are found. Although not reported in nodes from patients with CMV disease, Reed-Sternberg-like cells, similar to those found in our patient, have been demonstrated in nodes biopsied from patients with infectious mononucleosis,\textsuperscript{11} also a disease of viral etiology. The finding of node changes indistinguishable from a malignant process as reported here makes one wonder what relationship these viruses may have to the induction of malignant neoplasia in man.

Another alteration of special interest is the pronounced changes in serum and salivary immunoglobulin concentrations associated with the CMV infection. McCracken and Shinefield have reported increased IgA and IgM serum levels in nine neonatal infants with congenital CMV infection.\textsuperscript{12} Normal neonatal infants generally have low IgM and no detectable IgA. In another report, transient monoclonal gammopathy was found during CMV infection in two children with acute leukemia, one of whom was in remission.\textsuperscript{13} The monoclonal immunoglobulin was IgG-kappa in one case and IgM-kappa in the other. In our patient, the concentrations of IgM and IgG were within normal limits, although definite fluctuations in the latter were noted, particularly during the relapse of her leukemia 8 mo later (Fig. 3). Most striking was the pronounced increase in concentrations of IgA and the transient occurrence of a monoclonal gammopathy of the IgA lambda type and Bence Jones proteinuria during the acute febrile illness.

Transient monoclonal gammopathies, rarely Bence Jones proteinuria, have been noted during episodes of acute inflammatory disease. The transient or secondary type of monoclonal gammopathy, as the name implies, disappears spontaneously when the underlying cause is resolved or removed. The increasing number of patients reported to have transient monoclonal gammopathies may reflect the more extensive use of serum electrophoretic analysis and development of more sensitive techniques for detecting immunoglobulin alterations. Of the reported cases of transient monoclonal gammopathy, six have been associated with sundry viral infections, five with viral hepatitis,\textsuperscript{13,14} and one with "viral fever."\textsuperscript{12} Of the five with viral hepatitis, four had an IgM gammopathy and one had an IgG monoclonal gammopathy. One patient with acute lymphoblastic leukemia in remission developed methotrexate-associated hepatitis; the monoclonal gammopathy that occurred disappeared as the hepatitis resolved after the drug was discontinued.\textsuperscript{14}

We postulate that the infection with CMV, with its known predilection for secretory organs, provided the antigenic stimulus for the profound effects on immunoglobulin synthesis and also lymph node alterations described in our patient; the temporal association of these features with the infection and the localization of antibody to CMV in the patient's IgA and IgG would suggest an unusual or exaggerated host immune response to this infection in spite of concurrent immunosuppressive therapy. The leukemia presumably played no direct role in this response, but the antileukemic therapy provided the milieu for the florid infection and associated protein abnormalities.
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REFERENCES

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