Single Case Reports

Graft-versus-Host Disease in the Central Nervous System

A Real Entity?

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Graft-*versus*-host disease (GVHD) classically involves the skin, intestines, liver, esophagus, and tongue. Clinically apparent disease involving the heart, lungs, kidneys, and central nervous system (CNS) is frequently secondary to other complicating factors. This report describes a case of an infant with severe combined immune deficiency (SCID) who developed unusual manifestations of GVHD following a bone marrow transplant (BMT). These were complete heart block and respiratory insufficiency in the absence of significant pulmonary disease. He lived 133 days post-transplantation. At autopsy, the brain showed focal lymphohistiocytic aggregates which may represent a hitherto unreported lesion of GVHD. (Key words: CNS; Brain; GVHD; Mononuclear infiltrates) Am J Clin Pathol 1988; 89: 543–546

CLASSICALLY graft-*versus* host disease (GVHD) manifests clinically as dermatitis, diarrhea and elevated liver function tests.[14,16,17] Recently a polymyositis-like syndrome has been recognized.[1,12,20] Myocardial involvement is usually focal and asymptomatic while central nervous system lesions have to our knowledge not been described.[10,16,18] We report unusual clinical and pathologic manifestations of GVHD in an infant with severe combined immune deficiency (SCID) following bone marrow transplant (BMT).

Clinical History

A male infant was born to a 22-year-old G3P2 mother by spontaneous vaginal delivery following an uncomplicated pregnancy (birthweight, 3.7 kg). A previous male sibling had died following bone marrow transplant for SCID. Initial immunologic evaluation showed a profound lymphopenia of 192×10^6 lymphocytes/L of blood (normal $> 1.2 \times 10^9$/L). Only 4% of these were T-cells (normal > 42%) as assessed by the E-rosette technique and reactivity with OKT 3 (CD3), OKT 11 (CD2) monoclonal antibodies. All of the T-cells identified were of maternal origin as assessed by HLA typing. There was no response of the peripheral blood mononuclear cells to stimulation by phytohemagglutination (PHA), *i.e.*, 861 cpm of ^{3}H-thymidine (normal > 100,000). He underwent maternal, haploidentical bone marrow transplant at 3 weeks of age, prior to onset of infectious complications. The infant developed acute GVHD manifesting as diarrhea, dermatitis, a peripheral blood eosinophilia of 740×10^6 cells/L of blood (normal $< 400 \times 10^6$/L), and elevated liver function tests. Concomitantly, the patient's lymphocyte count rose to 2.7×10^9/L and the activities of these lymphocytes with monoclonal antibodies were: OKT 11 (CD 2) 43%, OKT 3 (CD 3) 23%, OKT 4 (CD 4) 8%, OKT 8 (CD 8) 25%, B1 (CD 20) 32%. All T-cells were of maternal origin and all B-cells (B1+) were of host origin as assessed by HLA typing. Stimulation of peripheral blood mononuclear cells by mitogens yielded the following incorporations (cpm) of ^{3}H-thymidine: PHA, 21,000; concanavalin A (con A), 19,000; and pokeweed mitogen (PWM), 44,000. He was treated with intravenous monoclonal antibody and high-dose methylprednisolone. He had several febrile episodes, for each of which multiple viral, fungal and bacterial cultures were obtained. One of these yielded a single positive blood culture (*Staphylococcus epidermidis*). This episode resolved with vancomycin. One month prior to death (day 97 post BMT), he had a cyanotic episode with onset of complete heart block and hypotension. Extensive cardiology work up disclosed mild cardiomegaly with normal ejection fraction and no uptake on gallium scan. He then required a pacemaker and mechanical ventilation with minimal ventilator settings (FIO_2 30%, IMV 15 and PEEP 4) for the remainder of his course. Clinically, he continued to exhibit classic signs and symptoms of GVHD with intermittent periods of remission. He was maintained on low-dose steroids. Specifically he did not receive radiation, cyclophosphamide, cyclosporin A or other chemotherapy prior to or following transplantation. Two skin biopsies and a rectal biopsy showed acute GVHD. A myocardial biopsy and a skin biopsy showed chronic GVHD. On day 132, he became acutely anuric and hyperkalemic and died 26 hours later despite aggressive resuscitative efforts. An autopsy was performed.

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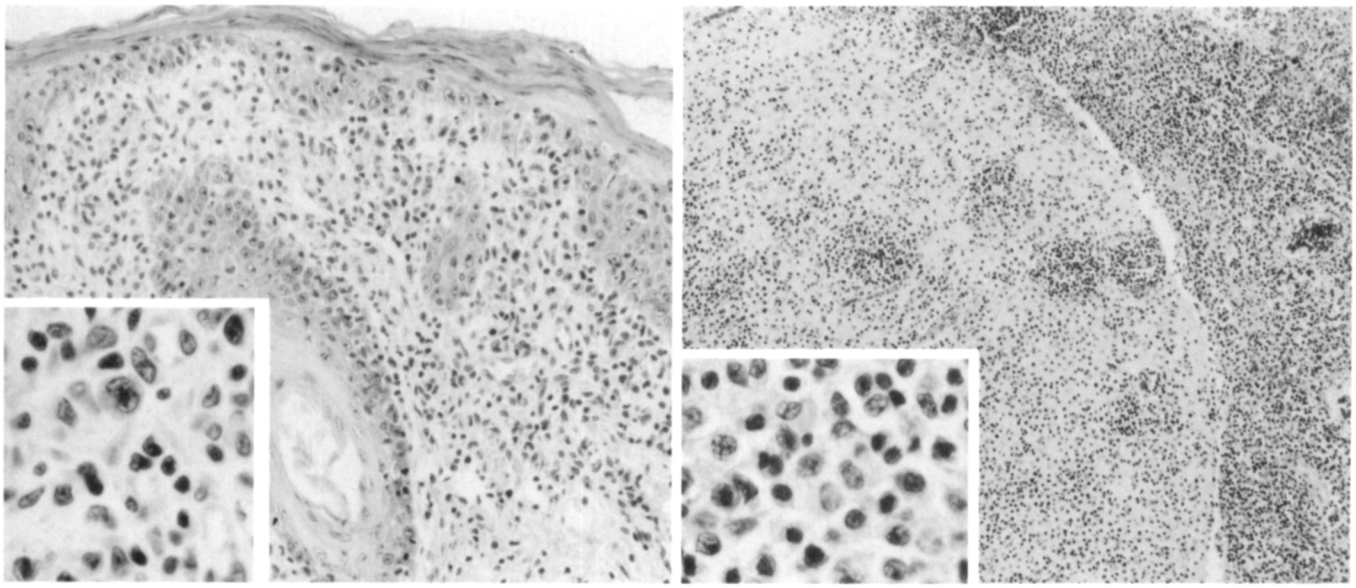

FIG. 1 (*left*). Skin biopsy, one day prior to death, showing band-like infiltrate and epidermal atrophy, classic for chronic GVHD. Hematoxylin and eosin (×63). *Inset:* Higher power view of mononuclear infiltrate showing lymphoid and monocytoid cells. Hematoxylin and eosin (×400).

FIG. 2 (*right*). Section of hippocampus showing extensive leptomeningeal mononuclear infiltrate extending into Virchow-Robin spaces and spilling out into brain parenchyma. Hematoxylin and eosin (×63). *Inset:* Higher power view of mononuclear infiltrate showing lymphoid and monocytoid cells. Hematoxylin and eosin (×400).

Pathological findings

Skin biopsies (days 12 and 56 post-transplant) showed acute GVHD characterized by hyperkeratosis, follicular plugging, hydropic degeneration of the basal layer, dyskaryosis and satellite lymphocytes in the epidermis.

Rectal biopsy (day 38 post-transplant) also showed acute GVHD consisting of focal superficial epithelial cell loss, loss of glands, and reactive epithelium in the remaining glands, some of which were cystically dilated and filled with necrotic cellular debris. An occasional satellite lymphocyte adjacent to a necrotic pyknotic epithelial cell was identified. Granular necrosis was present at the base of many glands.

Liver biopsy (day 106 post-transplant) showed chronic GVHD characterized by paucity of bile ducts and small collections of mononuclear cells in some portal areas. There was also Kupfer cell hyperplasia. Viral cultures were negative.

Myocardial biopsy (day 124 post-transplant) showed interstitial edema, focal mononuclear infiltrate, and individual fiber degeneration.

Skin biopsy (day 132 post-transplant) showed epidermal atrophy, dyskaryosis and satellite lymphocytes, acanthosis, Civatte bodies, melanin pigment incontinence and a dense, band-like mononuclear infiltrate in the papillary dermis, characteristic of chronic GVHD (Fig. 1).

The two biopsies were reacted with monoclonal antibodies Leu 4 + 5b (pan T-cell or CD 3 + CD 2), Leu 2a (T8 or CD 8), Leu 3a plus b (T4 or CD 4), Leu 6 (T6 and Langerhans cells or CD 1), Leu 7 (HNK 1, natural killer cell), Leu M5 (monocyte) and Leu 14 (pan B-cell or CD 22). Both biopsies showed a majority of the infiltrating cells marking as T8 and some natural killer cells, a moderate number of cells marking as monocytes, a few cells marking as T4 cells, and no cells marking as B-cells.

Autopsy

At autopsy, the thymus was markedly atrophic and dysplastic with virtually no lymphoid tissue, abundant adipose tissue and prominent epithelial cells. A single lymphoid nodule was negative for kappa and lambda chains. There were no grossly identifiable lymph nodes or remnants thereof in the sections of mesentery. The spleen showed almost complete absence of white pulp with a rare small periarteriolar lymphocytic aggregate remaining. There were clusters of immunoblasts, eosinophils and focal extramedullary hematopoiesis.

Immunohistochemistry on the rare lymphoid aggregates showed these to be mostly T-cells (Leu 4 + 5b or CD 3 + CD 2) with occasional B-cells (Leu 14 or CD 22). The presence of histiocytes was confirmed by staining for muramidase.

The bone marrow showed marked eosinophilia and numerous early myeloid forms with some maturation. There was marked erythrophagocytosis and dyserythropoiesis.

Chronic GVHD was present in the skin, intestines, and liver. In addition, there was a single 2-mm subcapsular nodule of mononuclear cells in the liver. Stains of this aggregate for kappa and lambda chains, muramidase and alpha-1-antichymotrypsin were negative. Sections of several skeletal muscles, including the diaphragm, showed numerous interstitial mononuclear inflammatory infiltrates and degenerating and regenerating myofibers. These infiltrates were uniformly negative for kappa and lambda chains, but approximately 50% of the cells stained for muramidase and alpha-1-antichymotrypsin. Similar infiltrates (identical morphologically to those in the premortem biopsy) with foci of necrosis and scarring were present in the atrial and ventricular myocardium, atrioventricular node, bundle of His and right and left bundle branches. There was also extensive lymphohistiocytic pericarditis.

The lungs showed mild right upper lobe focal bronchopneumonia and rare organizing thromboemboli within small pulmonary arteries, a few of which had foci of calcification. Viral cultures of liver, lung and heart were negative and no viral inclusions were seen in the sections examined.

The brain was atrophic but normally developed. A 1.0 × 0.5 cm area of yellow discoloration was seen in the right hippocampus. There were focal mononuclear aggregates in the leptomeninges extending into the Virchow-Robin spaces and parenchyma of the right hippocampus (Fig.

2) and focally within the brainstem. Gram, AFB and GMS stains, *in situ* hybridization for CMV and Herpes, and dot blot hybridization for Epstein-Barr virus (EBV) on DNA isolated from brain infiltrates were negative. Approximately 50% of the cells stained positively for muramidase and alpha-1-antichymotrypsin and occasional cells stained positively with antibody to kappa and lambda chains. No frozen tissue was available for monoclonal antibody studies.

Discussion

The major complications of allogeneic bone marrow transplantation for lethal congenital immune deficiencies are GVHD, infection and the more recently reported lymphoproliferative disorders.[3,6,9,15] Infection is frequently the result of significant pathogens acquired early in life. In a series of 12 patients with SCID, Solberg and co-workers[19] found 58 separate infections following BMT. Seven of these patients died of infectious complications: three of gram-negative bacillary septicemia, two of *Pneumocystis carinii* pneumonia, two of *Pseudomonas aeruginosa* pneumonia. Of the latter two patients, one also had parainfluenza virus infection and the other CMV infection.

GVHD is a complex pathologic process initiated by engrafted, immunocompetent donor T-cells reacting against host histocompatibility antigens. Acute GVHD most commonly involves the skin, liver, gastrointestinal tract, lymphoid and hematopoietic systems. The disease may progress to chronic GVHD which exhibits clinical features of autoimmune diseases, especially scleroderma, and may involve multiple target organs.[10,13,14,16,17] Recently, polymyositis has been recognized as a manifestation of chronic GVHD,[1,12,20] and myasthenia gravis has been reported as a complication of long-standing GVHD.[2] Myocardial involvement, when present, is usually focal and asymptomatic.[10,16,18] In a recent study of neurologic complications following BMT, no evidence of GVHD was found in the CNS.[11]

A more recently reported complication of BMT seen in patients treated with anti-T-cell antibody for GVHD, is an EBV associated B-cell proliferation.[9,15] While the most frequent manifestation of EBV infection is infectious mononucleosis, (a benign, self-limited, polyclonal B-cell proliferation), EBV has been associated with abnormal lymphoid proliferations ranging from benign polyclonal B-cell hyperplasias to malignant lymphomas which can be monoclonal, biclonal, or polyclonal especially in immunosuppressed individuals.[5] Two patients who received BMT for acute leukemia developed severe GVHD and a fatal EBV-induced polyclonal lymphoproliferative disorder arising in donor-derived B-cells.[9] The third patient received a BMT for SCID at age 12 years and also developed fatal oligoclonal and monoclonal B-cell proliferations.[15]

Our patient first developed classic manifestations of acute GVHD characterized by dermatitis, enteritis, and hepatitis. Skin and rectal biopsies confirmed acute GVHD. Later skin, liver, and heart biopsies showed chronic GVHD. The histologic features as described above are classical for acute and chronic GVHD, respectively. The patient was treated with monoclonal anti-T-cell antibody followed by steroids which were tapered but maintained throughout his hospital course. He then developed chronic GVHD confirmed by skin and liver biopsies. His condition remained stable until one month prior to death when he developed heart block and an inability to maintain spontaneous respiration despite the absence of significant pulmonary disease. The multifocal lymphohistiocytic infiltrates identified in the myocardium, cardiac conduction system, and diaphragm at autopsy are sufficient to account for these clinical findings. The cellular identity between the premortem skin biopsy with chronic GVHD and the premortem myocardial biopsy supports the concept that the myocardial lesions were also due to chronic GVHD. Negative viral cultures performed on biopsy tissue offer further substantiation. At autopsy, cytologically similar infiltrates were seen focally in the leptomeninges extending into Virchow-Robin spaces and in the brain parenchyma of right hippocampus and brainstem. Infective etiologies, specifically bacterial, fungal and viral (CMV and Herpes), could be excluded based on histochemical stains and *in situ* hybridization studies. Furthermore, no viral inclusions were seen in the multiple sections examined and viral cultures of liver, lung, heart, and kidney were negative.

The morphologic similarities between the CNS infiltrates and those in other organs, as well as the absence of an identifiable infectious agent in the tissues examined, suggest that the CNS infiltrates may be part of the chronic GVHD process. An alternative interpretation of these lesions is that they represent an early stage of an EBV-driven B-cell or other lymphoproliferative process. Limited immunocytochemical studies characterized the cells as lymphocytes and histiocytes with only rare cells staining positively with antibodies to light chains, but B-cells have occasionally been reported to stain with histiocytic markers.[8] DNA isolation from formalin-fixed, paraffin-embedded brain infiltrates, using the method of Goelz and associates,[4] yielded low molecular weight DNA. Dot blot hybridization for EBV using previously described techniques[7] was negative. The suboptimal (low molecular weight) nature of the DNA makes this study inconclusive.

In summary, this patient developed acute GVHD following BMT. This progressed to chronic GVHD which, in addition to the classic organ distribution, involved the myocardium and cardiac conduction system, manifesting as heart block, and the diaphragm, causing significant respiratory embarrasment. In addition, we postu-

late that the CNS infiltrates represent either a hitherto unreported manifestation of GVHD or the very early stage of a lymphoproliferative process rather than an infectious complication of BMT.

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Pseudoglandular Schwannoma

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The glandular schwannoma is a rare variant of schwannoma in which the focally occurring glands are lined by cells resembling intestinal or respiratory type epithelium, or ependyma. The schwannomas harboring these glands are usually malignant and usually arise in the setting of von Recklinghausen's disease. The case presented in this report is that of a 43-year-old man who had no evidence of neurofibromatosis, and developed a benign schwannoma that contained multiple well formed gland-like structures. The cells lining the glands, when examined ultrastructurally and histochemically, proved to be Schwann cells, a unique finding that may provide a clue to the histogenesis of the glandular schwannoma. (Key words: Schwannoma; Glandular schwannoma; Pseudoglandular schwannoma) Am J Clin Pathol 1988; 89: 546–552

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NERVE SHEATH TUMORS may harbor a variety of heterologous elements, including cartilage, bone and striated muscle. This phenomenon is more common in cases of malignant schwannoma, and these elements are usually seen in the form of chondrosarcoma, osteosarcoma and rhabdomyosarcoma.[5] A distinctly unusual occurrence is the formation of glandular structures within schwannomas. This phenomenon was first described in 1892 by Garre'[4] and has since been observed by other investigators[2–4,6–14] who have asserted that these glands resemble ependyma[2,3,7] or respiratory or intestinal type epithelium,[9,12,14] respectively.