Sinus Histiocytosis Massive Lymphadenopathy Syndrome: Histogenesis of the Hepatic Lesion*

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ABSTRACT

Sinus histiocytosis massive lymphadenopathy (SHML) syndrome with hepatic involvement, occurring in a seven-year-old black female, is reported. Morphologic characterization of the hepatic lesion is accomplished utilizing conventional light, fluorescent and electron microscopy, and histochemical techniques and by comparing and contrasting the findings with those in cases of familial erythrophagocytic lymphohistiocytosis (FEL) and virus-associated hemophagocytic syndrome (VAHS). The histiocytic proliferation in the liver in SHML differs by showing: (a) an intralobular distribution with portal sparing; (b) marked steatosis; and (c) lipofuscinosis. The aforesaid intralobular distribution and the accompanying hypertrophy and hyperplasia of Kupffer cells, as well as commonalities of steatosis and lipofuscinosis, and, to a lesser extent, erythrophagocytosis and siderosis, suggest a histogenesis from Kupffer cells. The histochemical finding of fatty acid peroxides and both fluorescent microscopic and histochemical evidence of lipofuscin inclusions, a by-product of lipid peroxidation, in Kupffer cells provide at least a theoretical basis for both erythrophagocytosis and proliferation eventuating in intralobular histiocytosis.

Introduction

Sinus histiocytosis with massive lymphadenopathy (SHML) was first described as a benign clinicopathological entity by Rosai and Dorfman in 1969. The majority of cases (some 80 percent) occur within the first two decades of life and are characterized clinically by lymphadenopathy, fever, leukocytosis, hypergammaglobulinemia, and an accelerated erythrocyte sedimentation rate. Extranodal involvement has been reported in up to 28 percent of cases. Although the liver is a reported site of such involvement, information concerning the morphologic characterization and histogenesis of the hepatic lesion is sparse. In attempting to
expand the latter, the light, fluorescent and electron microscopic and histochemical features of the histiocytic proliferations in the liver of a patient with SHML syndrome are reported; the findings are contrasted with those in cases of familial erythrophagocytic lymphohistiocytosis (FEL)\(^3\) and virus-associated hemophagocytic syndrome (VAHS)\(^1,15\), and a histogenesis from Kupffer cells is proposed.

Case Report

A seven-year-old black female presented with a three week history of recurrent fevers, wheezing, episodes of vomiting and diarrhea, headaches and congestion, and, finally, right upper quadrant (abdominal) pain. Her past medical history included treatment for reactive airway disease. Out-patient laboratory findings obtained two weeks prior, revealed both a normal white blood cell count and hemoglobin concentration and 2+ ketonuria. Therapeutic agents administered during this period included Accurbron\(^5\) (a theophylline preparation) and Alupent\(^6\) (a beta-adrenergic stimulator).

Physical examination at the time of admission to Cook-Fort Worth Children’s Medical Center revealed a somewhat dehydrated child with a temperature of 38.0°C. Pertinent negative findings included absence of lymphadenopathy and organomegaly. Laboratory data revealed: anemia (hemoglobin 7.6 g per dl with expected range for age 11.4 to 15.4 g per dl)\(^4\), thrombocytopenia (125 × 10\(^3\) per mm\(^3\) with expected range for age 250 to 470 × 10\(^3\) per mm\(^3\)\(^2\)), hyperaspartate aminotransferaseemia (142 U per L with expected range for age 20 to 45 U per L) and hyperalanine aminotransferaseemia (57 U per L with expected range for age 2 to 15 U per L)\(^2\) associated with eubilirubinemia (total 1.2 mg per dl with expected range for age up to 1.5 mg per dl)\(^1\). An erythrocyte sedimentation rate performed on the second day of admission with the patient’s hematocrit having been raised to 30.6 percent following a transfusion of packed red blood cells was accelerated (62 mm per hour with expected range for age 0 to 15 mm per hour)\(^3\).

Seronegativity for antibodies to *Mycoplasma pneumoniae*, hepatitis A and B viruses, Epstein-Barr virus and antineutrophil antibodies was documented. Similarly, viral and bacterial cultures collectively of urine, blood, stool, throat and bone marrow failed to demonstrate infectious agents. A bone marrow biopsy revealed plasmacytosis and erythrophagocytosis by histiocytes consistent with a hemophagocytic syndrome. During this first hospital course, she developed cutaneous lesions, and a punch biopsy of the skin revealed a histiocytic proliferation consistent with extranodal involvement by SHML syndrome. Approximately two weeks after discharge, she developed cutaneous lesions, and a punch biopsy of the skin revealed a histiocytic proliferation consistent with SHML syndrome. At the time of the writing of this manuscript (approximately 14 months post discharge), the patient is asymptomatic and in a relatively good state of health.

Materials and Methods

SHML Syndrome

Excisinal lymph node biopsies of the cervical (neck) region and omentum as well as an open liver biopsy provided fresh (unfixed) tissues for the histopathologic studies in this case. Portions of the cervical lymph node were submitted for culture and cryostat sectioning, respectively. Touch imprints were made of the cut-surfaces of both lymph nodes and portions of each were placed in Carson’s (formalin) fixative for routine light and electron microscopy. The wedge-shaped piece of liver showed multiple but pinpoint-sized, yellowish foci in the fresh state. Touch preparations were made and a portion was subjected to cryostat sec-
tioning. The remainder of the liver biopsy was placed in Carson’s fixative.

The aforesaid cryostat sections of fresh tissue were stained with oil red O for lipid; the Winkler-Schultze technique for fatty acid peroxides and periodic acid-Schiff hematoxylin (PASH). Cryostat sections of liver fixed in Carson’s were treated with 1.0 percent osmium tetroxide to demonstrate lipid. Another portion of the Carson’s-fixed liver tissue was post-fixed in 1.0 percent osmium tetroxide and processed for electron microscopy. Thin sections were stained in uranyl acetate and lead citrate and examined in a JEOL transmission electron microscope. Representative deparaffinized sections of lymph node and liver were either stained with hematoxylin-eosin (H and E), Mallory’s procedure for iron, and Fontana-Masson for lipofuscin or mounted in phosphate-buffered saline for detection of possible autofluorescence. The latter was carried out using an epi-illumination fluorescent microscope employing a BG12 + KV418 exciter filter and an OG515 barrier filter. Additional staining techniques performed on deparaffinized sections of liver included: periodic acid-Schiff hematoxylin (PASH) with and without diastase pretreatment; method of Armed Forces Institute of Pathology (AFIP) for lipofuscin; diamino silver hydroxide method for reticulin fibers; aldehyde-thionin method for hepatitis B surface antigen; Brown-Hopps Gram stain for bacteria; Ziehl-Neelsen for acid fast organisms and silver-methenamine for fungi.

**ERYTHROPHAGOCYTIC LYMPHOHISTIOCYTOSES**

Biopsy materials of liver specimens from three cases of familial erythrophagocytic lymphohistiocytosis (FEL) and one case of Epstein-Barr virus-associated hemophagocytic syndrome (VAHS) were available in our files. The histopathologic and histochemical findings in these were compared and contrasted with those in SHML syndrome.

**Results**

As noted previously, excisional biopsies of cervical and omental lymph nodes revealed marked expansion of sinuses by a population of reactive histiocytes containing abundant cytoplasm and associated, on occasion, with intracytoplasmic collections of small lymphocytes. Focal plasmacytosis was also present. The perinodal tissues showed striking multilocular change in the adipocytes.

A proliferation of similar appearing histiocytes was evident in the liver as micronodular foci showing both an intra-lobular distribution and contiguity at their periphery with sinusoidal cells (figures 1 and 2). Other distinguishing histopathologic features of such proliferations included focal hyperemia and associated congestion and dilatation of sinusoidal spaces. Furthermore, in the latter regions, erythrophagocytosis by histiocytes was more readily appreciated. Finally, no microorganisms were detected in these lesions using the aforementioned staining techniques.

Histochemical and fluorescent microscopic studies on histiocytic cells within these foci revealed the following intracytoplasmic findings: occasional Schiff’s positive/diastase-resistant aggregates (figure 3); lipofuscin inclusions as evidenced by the combination of Fontana-Masson and (Kinyoun’s) carbol fuchsin positivity and appropriate autofluorescence (figure 4); and numerous lipid inclusions that were oil red O positive (figure 5) and osmiophilic and frequently exhibited Winkler-Schultze positivity at their cytosolic interface (figure 6). Ultrastructural counterparts of several of these histochemical and fluorescent microscopic findings were also
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FIGURE 1. Low power view of portion of liver lobule (H&E ×200) showing a micronodular focus of relatively pale cells surrounded by trabeculae of hepatocytes.

FIGURE 2. High power view of such micronodular foci in the liver (H&E ×1000) reveals a preponderance of histiocytic cells, some with indented nuclei and abundant foamy to vacuolar cytoplasm.

FIGURE 3. Periodic acid Schiff’s (PAS) positive (red) intracytoplasmic aggregates in several histiocytes within the micronodular foci in the liver (PAS with diastase pretreatment ×1000).

FIGURE 4. Yellowish-orange autofluorescent inclusions consistent with lipofuscin within histiocytic foci in the liver. Note bright green autofluorescent erythrocytes within adjacent, congested vascular spaces (deparaffinized, unstained section mounted in phosphate-buffered saline ×1000).

FIGURE 5. Numerous anisodimensional (red) lipid inclusions occupy the cytoplasm of the intralobular histiocytes in the liver (oil red 0 ×1000).

FIGURE 6. Indophenol blue synthesis consistent with the presence of fatty acid peroxides evident at periphery of lipid droplets in hepatic foci using Winkler-Schultze technique (×1000).

evident in transmission electron micrographs of representative foci (figures 7 and 8). In comparison with other lymphohistiocytic processes and specifically cases of FEL and VAHS, obvious differences were found. Such differences related to the microanatomical location of the histiocytic proliferations and the
degree of steatosis and lipofuscinosis associated with same. These are summarized in table I.

In addition to the aforementioned contiguity of sinusoidal cells with the intralobular, micronodular foci, there was sometimes striking proliferation of Kupffer cells which appeared to be expanding and filling the sinusoids and thereby presumably imposing a physical barrier to the blood flow through same. On rare occasions, plasmacytosis was noted in association with Kupffer cells. Apart from the morphologic similarity of
the proliferating Kupffer cells to the histiocytes in the micronodules, there were other commonalities to include steatosis, lipofuscinosis, and, to a lesser extent, siderosis and erythrophagocytosis. Finally, a positive reaction for fatty acid peroxides using the Winkler-Schultze technique was evident in some sinusoidal, presumably Kupffer cells in association with lipid inclusions.
## Comparative Findings in the Liver in Erythrophagocytic Syndromes

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### Discussion

The clinical and laboratory findings of fever, leukocytosis, polyclonal gammopathy, and an accelerated erythrocyte sedimentation rate when coupled with the development of a lymphadenopathy showing characteristic histopathologic features established the diagnosis of SHML syndrome in this patient. In addition, extranodal involvement by similar appearing histiocytes was evident in biopsies of her skin and liver.

Although SHML syndrome in the liver must be distinguished from other lymphohistiocytic processes such as FEL and VAHS that also show prominent erythrophagocytosis, the histopathological differences, based on the finding in our study, are obvious, and, therefore, make this task relatively easy. Specifically, the histiocytes in SHML differ from those in FEL and VAHS by showing: (1) an intralobular distribution with portal sparing; (2) marked steatosis; and (3) lipofuscinosis.

The histogenesis of the hepatic histiocytic proliferation in SHML appears to be from Kupffer cells. This conclusion is based on the following: (a) the intralobular distribution of the micronodular foci; (b) the contiguity of said foci with sinusoidal (Kupffer) cells; (c) an accompanying hypertrophy and hyperplasia of Kupffer cells; and (d) commonalities of steatosis and lipofuscinosis and, to a lesser extent, of erythrophagocytosis and siderosis.

Although it is realized that the etiopathogenesis of the Kupffer cell proliferation in SHML may involve some inherent hyper-responsiveness on the part of the reticuloendothelial system to normal stimuli, the present authors are intrigued by the possibility that lipid peroxidation may be initiating the process. The collective evidence for lipid peroxidation’s having occurred in the proliferating Kupffer cells and/or micronodular histiocytic foci includes histochemical, autofluorescent and ultrastructural footprints of same. These comprise: the presence of lipofuscin (which is considered to be a by-product of lipid peroxidation, probably 1-amino-3-iminopropene derivatives of malondialdehyde), the finding of Schiff’s positive and granular, osmiophilic aggregates in the cytoplasm of histiocytes by routine histochemistry and transmission electron microscopy, respectively; and the demonstration of fatty acid peroxides in such cells with the Winkler-Schultze technique. The elements nec-
necessary to effect lipid peroxidation are unsaturated fatty acids and molecular oxygen with "iron" serving as a catalyst. In this regard, the presence of siderosis and of steatosis with both granular, osmiophilic densities, and Winkler-Schultze positivity at the cytosolic interface of individual lipid inclusions is noteworthy. Moreover, the activation of "scavenger receptors" in Kupffer cells by products of lipid peroxidation might also stimulate their hypertrophy and proliferation.

The resultant narrowing of sinusoidal spaces would force intimate contact between Kupffer cells and circulating erythrocytes, perhaps leading to peroxidative damage of red cell membranes and erythrophagocytosis. Finally, the relative rarity of extranodal hepatic involvement in SHML syndrome suggests that a combination of circumstances must be operative for this to become manifest in any given individual. In this patient, it is clear that multiple factors promoting lipid peroxidation were present. For example, free fatty acids for lipid peroxidation appear to have been mobilized from adipose tissue stores as evidenced by the previously described multilocular change in adipocytes in biopsy material. Circumstances promoting this over the course of her illness include a poor caloric intake consequent to episodic vomiting and diarrhea and the administration of potential adipo- de (O→) but also the reductive release of iron from expanded ferritin stores in the Kupffer cells (vide supra).

In summary, it is proposed that lipid peroxidation consequent to an interplay of numerous clinical circumstances and therapeutic agents may be responsible for stimulating both erythrophagocytosis and proliferation of Kupffer cells eventuating in intralobular histiocytosis, the hepatic lesion in SHML syndrome.

Acknowledgment

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References