SUMMARY

Though rare, deficiency of glucosylceramidase and/or sphingolipid activator proteins results in storage of glucoceramide in various organs leading to malfunction of the involved organs. Gaucher's disease has three clinical profiles among which type-I is a chronical non neuropathic form observed in adults. We examined in the liver biopsy specimen of a 27 years old, male patient who is clinically considered as a type-I lipidosis both at light and electron microscopic levels. Significant amount of lipid droplets in fat storing cells and presence of large granular cells were determined at light microscopic levels. Lysosomal membranous bodies and myelin figures were detected in these cells at electron microscopic levels. These findings provide clear support to the clinical diagnosis.

Key Words: Gaucher's disease, liver, light microscopy, electron microscopy

INTRODUCTION

In the diagnosis of inherited metabolic diseases, electron microscopy is an important method complementary to clinical, histological and biochemical assays (1). Among these metabolic diseases sphingolipidosis represent a number of clinical syndromes which were studied in detail to obtain data on their genetical basis, biochemical mechanisms and sturctural findings (2,3,4). Storage of sphingolipidosis in a variety of cells types occurs due to defective enzymatic activity for their degredation (4). A number of sphingolipid activator proteins (SAPs) were characterized some of which act as activators of the enzymes or solubilizing agents increasing the efficiecy of the enzymes. Both SAP subgroups which are well determined by biochemical assays are essential for the normal functioning of sphingolipid degradation processes preventing their storage that cause to these metabolitic disorders leading to functional loss of the involved organ, systems (2,4).
These inherited diseases are rather rare thus we examined in the liver biopsy specimen of an individual who was clinically diagnosed as nonneuropathic type-I Gaucher's diseases to obtain further evidence on the ultrastructural changes occurred in the liver and for differential diagnosis to support the clinical findings as electron microscopic findings provide a useful tool for this purpose.

MATERIAL AND METHODS

The needle biopsy specimen obtained by ultrasonography guide for differential diagnosis was examined using routine plastic embedding technique. The patient was 27 years old male clinically reported as Type-I lipidosis. He had significant hepatosplenomegalia which was also supported by ultrasonographic examination. With the exception of mild anemia and hypocalcemia, laboratory findings were normal. No other clinical symptoms especially in neuronal examinations were present with the exception of a very mild paleness.

Fresly taken liver biopsy specimen is fixed in 2% gluteraldehyde solutions for two hours at 4°C. Second fixation was done in 1% osmium tetroxide solution for 90 minutes. Tissues were dehydrated, infiltrated and embedded in araldite CY212. Semithin sections were cut with LBK 11800 pyramidtom and stained with 1% solution of methylene blue-Azur II in 1% borax, examined and photographed using Olympus BH2 light microscope. Ultrathin sections were cut, collected on copper grids, and stained with uranil acetate and lead citrate. The sections were examined and photographed using Zeiss 9S electron microscope.

RESULTS

In the examination of semithin sections at light microscopic level numerous large cells with basophilic granules were seen in the liver (Figures 1,2). Cells with dense basophilic granules were located mostly along sinusoids. There was a relative increase in the lipid droplets of fat storing cells. Many vacuoles of varying sizes within the hepatocyte cytoplasm were also observed (Figures 1,2). When the granules were examined ultrastructurally that they were seen to be osmiophilic (Figures 3,4). Their diameter and outline varied but all of these granules had unique ultrastructural characteristics. They were membrane bound having a multivesicular content with some myelin figures (Figures 5).
Figure 1a,1b: In the semithin sections of liver biopsy specimen liver sinusoids (*), endothelial lining, fat storing cells (arrows), hepatocytes and large cells with basophilic granules (arrow head) that represent Kupffer cells were seen. Large lipid droplet within fat storing cells and vacoules of varying sizes in the hepatocyte cytoplasm were also distinguished. Cells dense basophilic granules were located mostly along sinusoids. Methylene blue-azur II stained, original magnification X 40.

Figure 2: Electron micrograph of a Kupffer cell with numerous osmiophilic granules of varying sizes is seen. Hepatocytes (h) and sinusoidal lumen (l) with tiny granular material were also distinguished. Uranyl Acetate-Lead Citrate counter stained, X 6250.
Figure 3: Higher magnifications of the cell in figure 2. The nucleus (n) and electron-lucent cytoplasm containing few organelles were distinguished. Osmiophilic lysosomal granules with myelin figures of varying sizes and shapes were clearly distinguished. Uranyl Acetate-Lead Citrate counter stained, X 11250.

Figure 4: Details of the cytoplasmic granules of the same cell. The granules were membrane bounded having a multivesicular content some of which exhibiting myelin figures. These granules had an irregular outline and their diameter also varied. Uranyl Acetate-Lead Citrate counter stained, X 21250.

DISCUSSION
Lipid and glycogen storage diseases are rare but because of their severity they were studied in detail. Lipidosis are a group of diseases arising from failure of the degradations of certain macromolecules due to defective enzymatic activity. Defeciency of a number of enzymes and/or their coenzymes were determined to be giving rise to several clinically significant storage diseases (1,4). Gaucher's disease or glucocerebridosis is one of diseases considered in inherited metabolib disorders
have certain clinical features. Three clinical types of the diseases had been described. Type-1 is a chronic non-neuropathic form observed in adults while type-2 and type-3 are observed in juvenile patients among which type-2 having a more rapidly progressing course. Depending upon the type various clinical symptoms of varying degrees can be observed like hepatosplenomegaly, hypersplenism, anemia, thrombocytopenia, jaundice, bone lesions. In juvenile forms motor abnormalities including clonus, hyperkinesia and fasciculations, existence of pathological reflexes like spontaneous Babinski or Moro reflexes may also contribute to the clinical findings. For the differential diagnosis of hereditary storage diseases in addition to above mentioned clinical data, biochemical assays and electron microscopy is essential tools (1,4). In other words, electron microscopic examination is obligatory to differential diagnosis in centres where sophisticated biochemical assays are not available. Thus we examined the needle biopsy specimen of an adult patient for this purpose and presented our findings to further characterize the histopathological and electron microscopic features of the disease to confirm clinical diagnosis.

The most significant findings within the liver of this patient at light microscopic level were the presence of cells with numerous large basophilic granules relative increase in the lipid droplets of fat storing cells and presence of vacuoles of varying sizes within the hepatocyte cytoplasm. Detailed examination of the granules at ultrastructural level revealed the osmiophilic nature and detailed substructure of these granules. Their diameter and outline varied but all of these granules had unique ultrastructural characteristics. They were membrane bound having a multivesicular content with some myelin figures. The structural features of these storage granules were significantly distinct from those observed in Tay-Sachs’ disease (1).

The present findings represent a report of a patient with type-I Gaucher’s disease having rather mild clinical course. Our data will provide a useful guide for the clinical course of the disease on follow up and after comparison with other lipid storage diseases.
ÖZET

KARACİĞERDE TİP I GLUCOCEREBROSİDE LİPIDOSİS’İN ULTRAŞRÜKSEL YAPISI


REFERENCES


