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Wolman's Disease*

A Case Report With Lipid, Chromosome and Electron-microscopic Studies

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Wolman's disease, a rare abnormality of lipid metabolism, was first described by Abramov, Schorr, and Wolman under the title of "Generalized xanthomatosis with calcified adrenals" in 1956[1]. Wolman and colleaques[2], by studying two affected sibs of the original case, reported that the liver, spleen, and adrenals in this disease contained greatly increased amounts of cholesterol and neutral fat. With the suggestion of Crocker et.al.[3] in 1965 this entity was named Wolman's disease. Patrick, Lake and Young[4, 5, 6] have recently demonstrated a total deficiency of acid lipase activity, directed toward the hydrolysis of triglycerides and cholesterol esters. This discovery could help in the explanation of significant biochemical features of this disease.

This rare disease usually becomes clinically evident in the first weeks of life and is characterized by gastrointestinal symptoms, failure to thrive, hepatosplenomegaly, steatorrhea, calcification and enlargement of the adrenals and death before 6 months of age.[1-4, 7] However, some cases

are reported with the biochemical and histological features identical to those of the acute infantile form of the disease, but have a later onset and less severe and more prolonged course.[6, 8]

In this communication we would like to present the 26th case of Wolman's disease,[6, 7, 9, 10] the first in a Turk in whom the diagnosis was well established with morphologic and chemical studies of liver biopsy material.

Case Report

S.K. (HCH 490002), a 45 day-old female infant, was admitted to Hacettepe Children's Medical Center because of a protuberant abdomen and vomiting since birth. Jaundice was noticed on the 4th day after birth and hepatosplenomegaly was verified by the doctor, at 6 days of age. Three days prior to admission to this hospital diarrhea started and the enlargement of the abdomen became more prominent. She was the product of a normal full-term pregnancy with an uneventful delivery. The parents were first cousins and their first child died before reaching three months of age, with very similar complaints, and hepatosplenomegaly confirmed by the doctor (Figure 1).

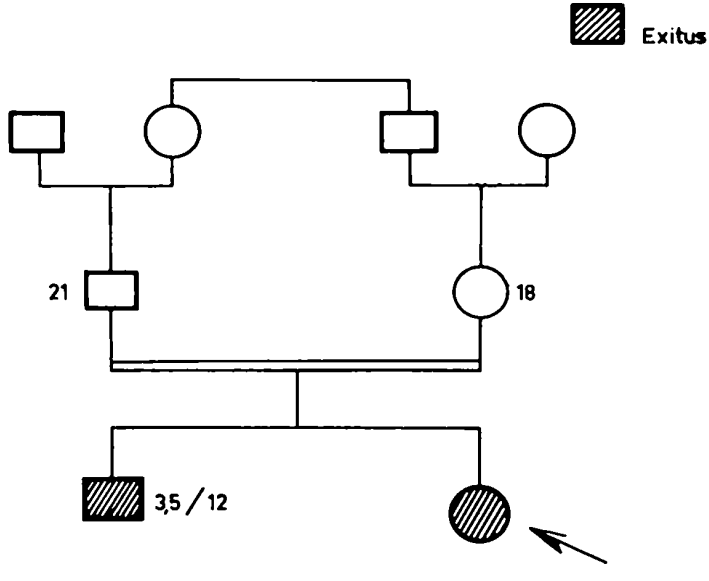

Figure 1
Family pedigree of the patient with Wolman's Disease (arrow indicates the propositus)

Physical examination: Revealed a moderately sick-looking infant with tachypnea (32/minute) and no fever (37°C). Her weight and height were 4400 gm. and 55 cm., respectively. The anterior fontanel was 2x2 cm., the bridge of the nose was depressed, high arched palate was observed and conjunctiva looked icteric. She had a normal size tonsils and a small umbilical hernia. The abdomen was markedly protuberant; liver and spleen were palpable 11 and 13 cm below respective costal margins. The rest of the findings, including neurologic and eye grounds, were non-contributory.

Laboratory findings: The Hb was 10.64 g/100 ml with 18.000 / µl leukocytes; the peripheral smear revealed vacuoles in some lymphocytes with adequate platelets (Figure 2). Liver function tests were grossly

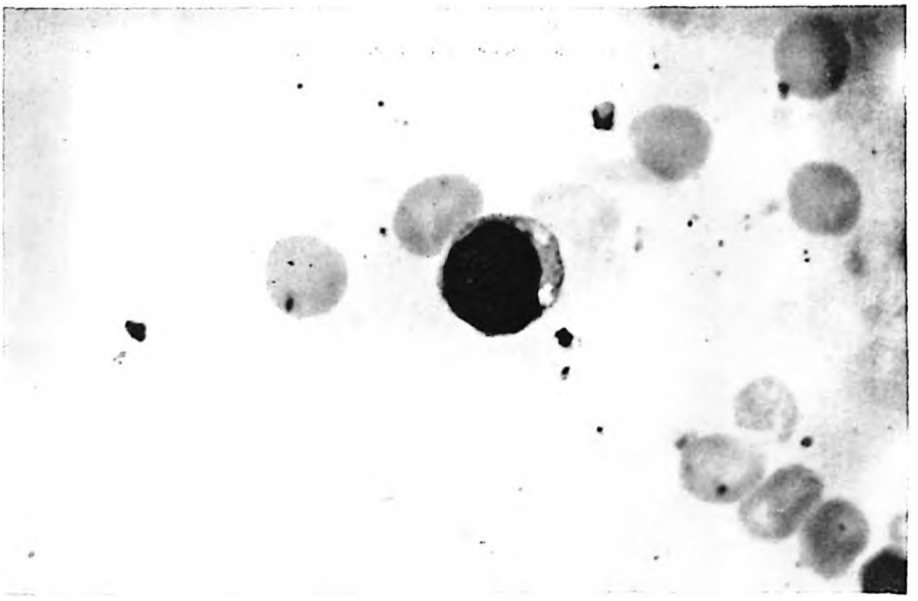

Figure 2
Peripheral smear lymphocytes have occasional vacuoles

abnormal (SGOT 1800 U; SGPT 540 U; total bilirubin 3.6 mg/100 ml, (2.2 mg/100 ml, conjugated): total protein 4.7 g/100 ml, (albumin 2.7 g/100 ml); total lipids 880 mg/100 and cholesterol 226 mg/100 ml, (slightly elevated for her age), prothrombin time 22 sec (normal 12"), PTT, 128 sec (normal up to 120 sec). Bone marrow showed many foam cells (Figure 3). Radiologically, enlarged adrenal glands were visualized with fine, punctate calcifications, (Figure 4). Urinalysis revealed bilirubin and it was negative for reducing sugars. Microscopic examination of the liver

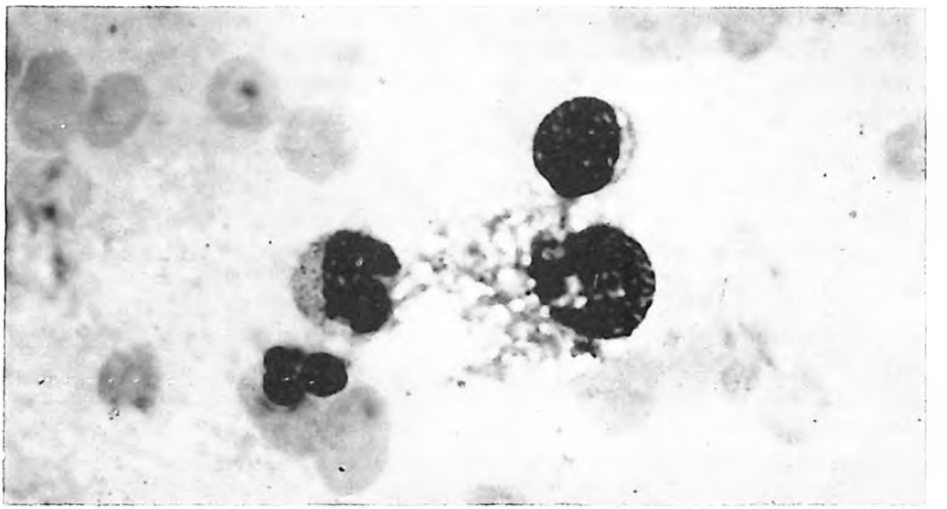

Figure 3
A number of histiocytes with foamy cytoplasm were noted in the bone marrow

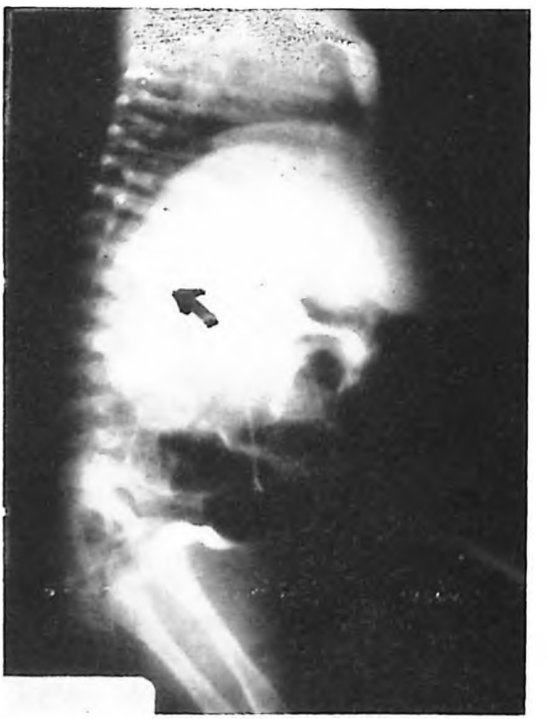

Figure 4
Enlarged adrenal gland is visualized by fine, puncuate calfications over the kidney on IVP films. (Arrow shows the eadrenal gland).

needle biopsy revealed clear vacuoles within the hepatic parenchyma and Kupffer's cells without evidence of fibrosis. By needle aspiration biopsy of the spleen, prominent population of foamy histiocytes were seen. Electronmicroscopic studies of the liver provided suggestive evidence of lysosomal involvement in the storage process. Hepatocytes were packed with lipid droplets and normal lysosomes were not seen. Crystalline inclusions in the macrophages of liver and spleen were interpreted as cholesterol (Figures 5,6,7). The results of biochemical determination of liver specimen revealed marked increase of triglycerides

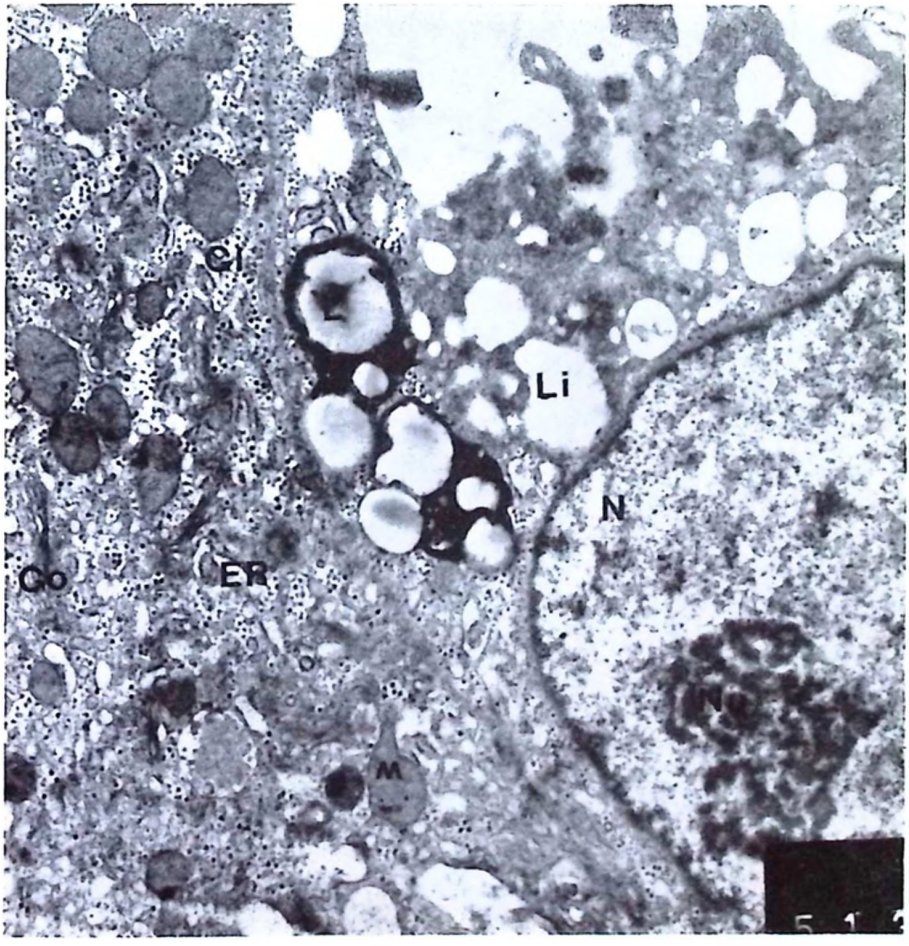

Figure 5

Low power electron micrography of liver parenchymal cell Nu, Nucleolus; N, Nucleus; M, Mitochondrion; Li, Lipid droplets; ER, Endoplasmic reticulum, L., Lysosome;

(720 mg/g), total cholesterol (270 mg/g) and cholesterol esters (209 mg/g) accumulations, determined by the methods of Hsia-Inouye[11] Meites-Faulkner[12] respectively.

Peripheral blood chromosome studies showed normal karyotyping in all 30 metaphases. Jaundice deepened on the 2nd day; on the 4th day of admission hematemesis started and she died in spite of blood, plasma transfusion, and vigorous supportive therapy. Autopsy permission was not granted.

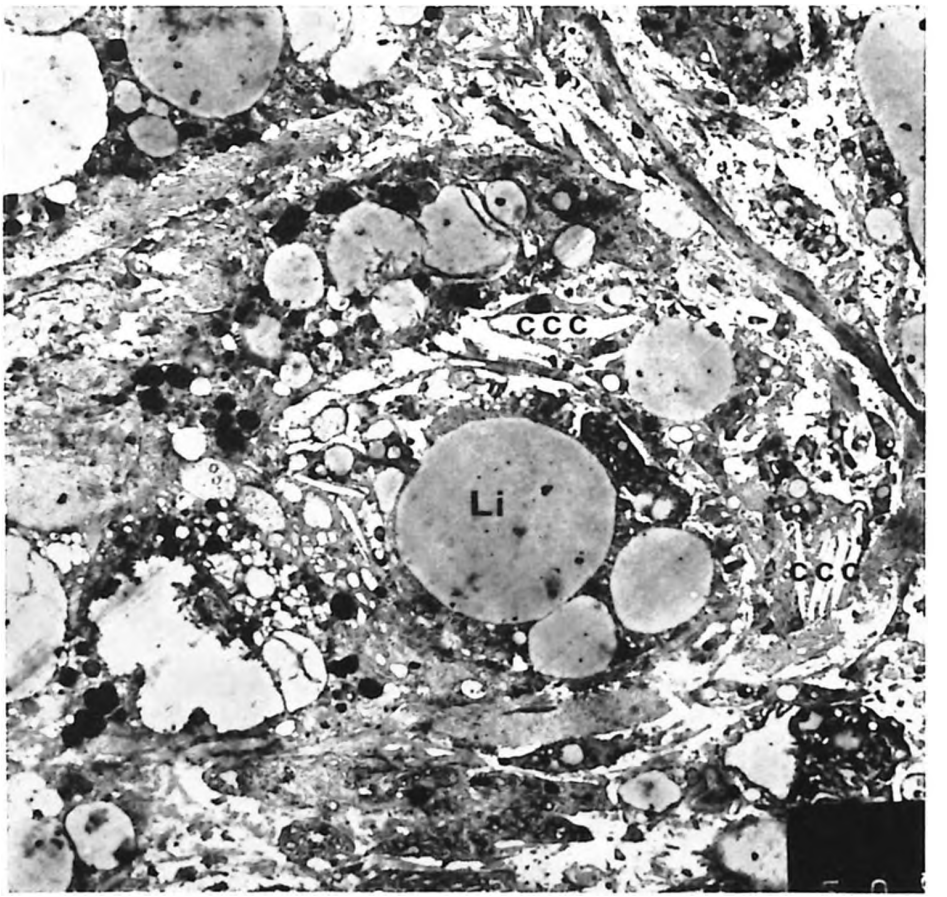

Figure 6

A characterictic view of Kupfer cell in Wolman's Disease. Li, Lipid droplets; CCC, Cholesterol crystal cleft. X 18.000.

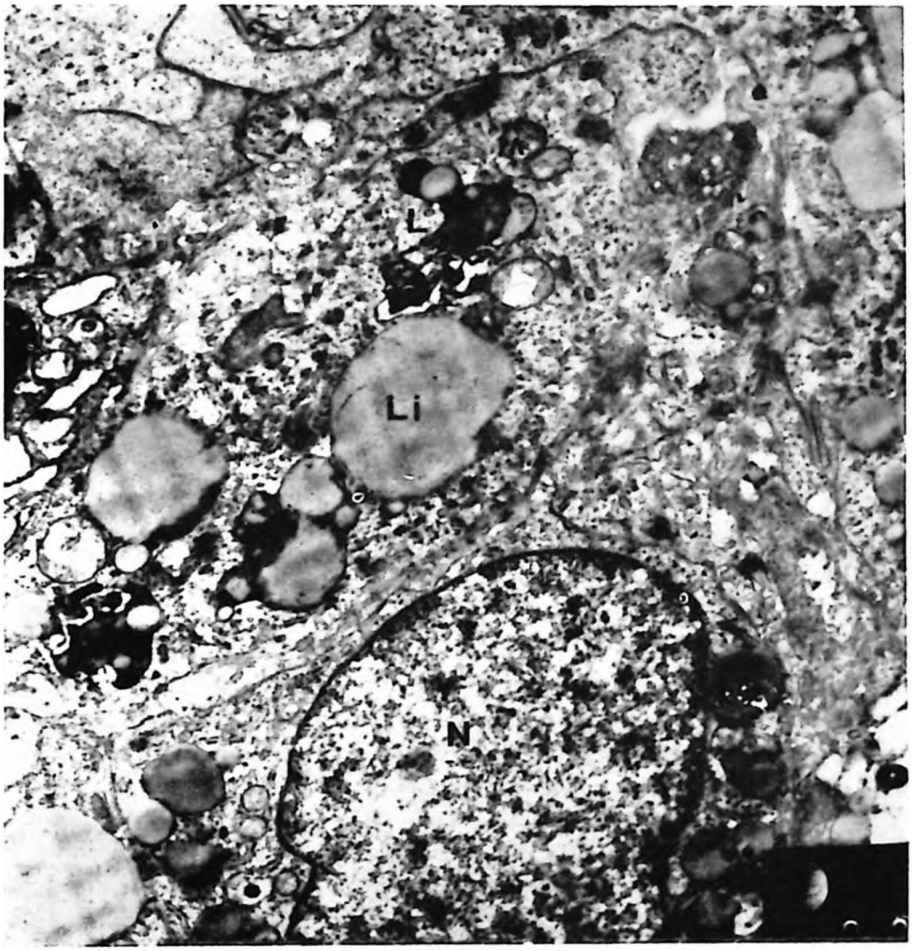

Figure 7
A part of a macrophage cytoplasma in spleen. L. Lysosome; Li Lipid droplets; N, Nucleus; M, Mitochondrion. X 18.000.

Discussion

Wolman's disease is probably an autosomal recessively inherited, fatal familial disease, characterized by the accumulation of large amounts of cholesterol esters, neutral lipids, and glycerides in the adrenal cortex, liver, spleen, upper part of the instestine, thymus and in limited quantity in the thyroid, testicles, ovaries, central nervous system, heart, lung, and kidneys. Vomiting and diarrhea, hepatosplenomegaly, steatorrhea, failure to thrive, and marked abdominal distension are usually observed in the first weeks of life; death usually occurs by 6 months of age. The finely-stippled or punctate calcification of the enlarged adrenal glands, which

are consistently demonstrated by X-ray examination, is the most striking feature of this disease. Anemia usually appears early and becomes more severe as the disease progresses. Intracytoplasmic and intranuclear vacuolization of peripheral lymphocytes, lipid laden histiocytes and large number of foam cells in the bone marrow are usual findings. Liver function tests are frequently abnormal, malabsorption, especially fat malabsorption, and depressed adrenal responsiveness are well documented. Although the clinical picture is fairly characteristics, the diagnosis should be made by chemical analysis of the stored lipids.[7]

Patricks, Lake and Young have demonstrated a deficiency of acid esterase activity in the liver, spleen and leukocytes of these patients.[4-6] Because of the deficiency of this enzyme it is probable that the catabolism of cholesterol ester and triglycerides is blocked at the step at which fatty acids are cleaved from the molecules; so cholesterol esters and triglyrerides accumulate in many cells, especially in lysosomes throughout the body. It seems that heterozygotes could also be detected by the assay of acid esterase activity in the leukocytes.[6] Its activity is so low in cord blood leukocytes of the patients that the diagnosis can be madde at the time of birth. Prenatal evaluation of the offspring of known carriers by the application of acid esterase activity assays to the aminiotic fluid after 14 weeks of pregnancy is a new approach for diagnosis.[6] Data obtained from skin fibroblast culture of the patients might be adopted for intrauterine diagnosis of the disease by amniocentesis.[13]

Low values of α and β-lipoproteins[8, 13, 14] and some changes of fatty acid pattern in the tissue lipids[13, 15] have been reported. These findings do not seem to be related to the pathogenesis of the disease; most likely it is the result of the metabolic changes, or diet.[13] In two patients with this disease greatly reduced concentrations of bile acids, phospholipid, and cholesterol in the bile were also shown.[13]

Chemical analysis of the lipids in the liver, clinical, his tological, and X-ray findings of this patient does not leave any doubt that she had Wolman's disease. It should be pointed out that triglycerides and cholesterol ester concentrations in the liver of this patients were found to be much higher than the previously reported values. Abnormalities of liver function tests are expected and jaundice was observed in a few papatients.[2, 3, 15] But in this case, as in the case of Kamalian et. al.[10] jaundice was associated with liver failure and hemorrhage. Death in Wolman's disease, unlike in our patient, is generally related to bronchopneumonia or impairment of instestinal absorption, with diarrhea, and nutritional failure.

This patients' parents were first cousins, which is supportive of recessive inheritance of the disease as mentioned in only two families[2, 16] Chromosomal analysis of the patient was normal as was reported only once previously.[15] Ultrastructural studies of the spleen and liver were very similar to the findings of two reported cases.[5, 17] Although Marshall etal.[8] and Barson[9] demonstrated large amounts of ceroid in the involved tissues, the electron microscopical findings of this entity do not look like other ceroid storage diseases, such as sea-blue histiocyte disease.[18] Wolman's disease by electron microscopy is quite easily distinguished from Niemann-Pick's disease,[19] which should be considered in the differential diagnosis. Of course, chemical determinations which were carried out in this patients are more essential in the differential diagnosis of these entities. Unfortunately acid esterase could not be assayed either in the patients' liver and spleen, or in the leukocytes of the parents to provide evidence for their heterozygosity. Since their first baby most probably had also had Wolman's disease, the occurence of disease, in this patient was not a new mutation.

Summary

A case of Wolman's disease is reported in a Turkish infant, in whom the diagnosis was verified by the chemical composition of the lipid in the liver and the spleen. The parents were first cousins and the patient died due to hemorrhages and liver failure. Hepatosplenomegaly was documented on the 6th day of life and a protuberant abdomen was noticed at birth. Typical adrenal calcifications were shown at 45 days of age. Chromosomal studies did not disclose any abnormality and electron-microscopic examination of the liver and the spleen showed typical lysosomal lipid storage and cholesterol crystals.

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