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GM_1 Gangliosidosis Type I (Norman-Landing Disease)*

(Report of a Case with Enzymatic Analysis of the Family)

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GM_1 gangliosidosis type I is a progressive, uniformly fatal sphingolipidosis of infancy.[1] It is transmitted as an autosomal recessive disorder and is characterised by widespread tissue accumulation of GM_1 ganglioside and a keratin sulfate-like acid mucopolysaccharide.[2-4]

In 1951, Smith described two neonates with many of the features of Hurler's syndrome but of prenatal onset.[5] Landing[6] and associates have summarized clinical characteristics and pathological findings in eight infants who fit into the group displaying what Caffey termed infantile Hurler's syndrome. Okada-O'Brian[7] have detected a severe deficit of the lysosomal enzyme B-galactosidase in the liver of these patients.

The purpose of this report is to document the clinical appearance, and laboratory findings, and to determine the beta galactosidase activity in the leukocytes of both the patient with this syndrome and his parents, and no compare these values with the controls.

Case Report

A 2 day-old boy was referred to the Hacettepe Children's Medical Center with a poor sucking reflex, abnormal face appearance and presence of bony swellings at the wrists and ankles.

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His parents were first cousins. The patient weighed 2.1 kg., and was the product of an uncomplicated 38-week gestation. The mother was 22 years old, gravida 3, para 2.

Physical examination revealed coarse facial features resulting largely from a depressed nasal bridge, frontal bossing marked hypertrophy of the alveolar ridges and maxillary processes. The tongue was large. The ears were low set (Figure 1). At the Fundus the "cherry-red spot" appearance was present. Extention of the upper extremities was limited to 140 degrees. The chest was deformed and narrow. Examination of the lungs revealed, bilateral crepitant rales. Hepatosplenomegaly was markedly prominent. Neurologic examination revealed poor sucking, moro and grasping reflexes. Bilaterally distal ulna and radius metaphysis enlargement was determined.

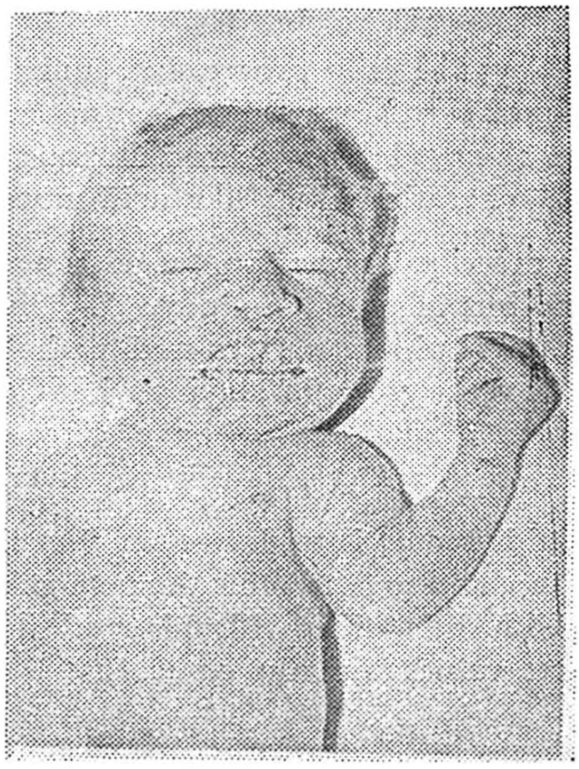

Figure 1

Showing appearance of the Patient. Marked hypertrophy of the alveolar ridges.

Initial laboratory findings were: normal urinalysis, Hb: 19.20 gr/dl, WBC: 9000/mm3, vacuolated lymphocytes were seen in peripheral smear. Total bilirubin 9.3 mg/dl, direct bilirubin 0.9 mg/dl, P: 5.2 mg/dl, Ca: 9 mg/dl. The mucopolysaccarides in urine were 0.9 mg/dl, (N: up to 3 mg/dl). Three bone marrow aspiration attempts were unsuccessful.

X-ray examination demonstrated generalized abnormality, especially in the long bones, most markedly in the humerus and femur. Periosteal thickening, increased irregular trabeculation and moderate demineralization were seen (Figure 2). The ribs were thick and flattened. "The shoe shape" appearance was demonstrated in the lateral skull roentgen. Bilateral bronchopneumonic infiltration was seen in the chest X-ray. The patient died within 3 days in spite of vigorous supportive treatment.

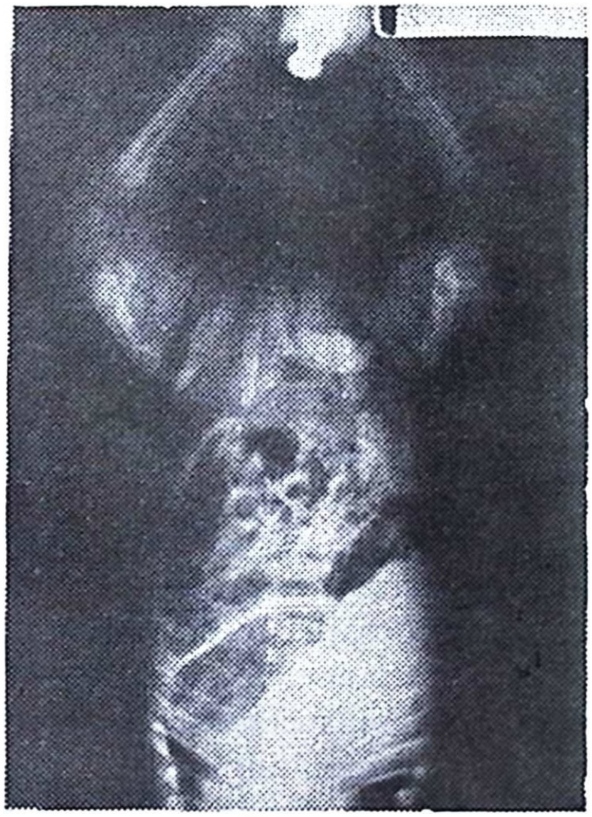

Figure 2

Bone radiograph demonstrating periosteal thickening increased irregular trabeculation

Necropsy Findings

The brain was found to be reduced in size weighing 330 gm, (Normal weight for this age 382 gm) and increased in consistency. The cortices of the cerebrum and cerebellum were atrophic and the white matter was pale. The deep gray matter was not remarkable, and the ventricular system was not dilated.

The neurons of the cerebrum and cerebellum showed cytoplasmic swelling. The white matter exhibited varying degrees of demyelinisation and axonal loss with reactive astrocytosis.

Microscopical examination revealed many foamy histiocytes in various organs; particularly in the lymph nodes, spleen, bone marrow, thymus, lamina propria of the intestine and liver. In the kidneys, the

glomerular mesangial cells, the lining cells of Bowman's capsule and the tubular epithelial cells, also contained numerous fine vacuoles.

Enzymatic Assay of the Family Member and the Control Subjects:

Materials-Leukocytes for beta-galactosidase activity were obtained from patients, family members, healthy adults and newborns. Leukocyte suspension containing approximately 85 % polymorphonuclear (PMN) leukocytes was prepared from 6 to 12 ml heparinized venous blood by using the standard technique of dextran sedimentation.[8] The erythrocyte contamination was reduced by hypotonic lysis.[9] Cells were suspended in 1 or 2 ml of saline and stored frozen as 200 µl portions at-20°C.

Enzyme Assay-Beta-galactosidase activity was determined by the modification of the method of Hindman and Cotlier using 4-methylumbelliferyl-beta galactosidase (4-MU-gal) as substrate.[9] The 200 µl leukocyte suspension was diluted to 1 ml with distilled water to which Triton x-100 µl had been added to reach the final concentration of 0.1 % (V/V). Twenty microliters of leukocyte sample were added to 100 µl of the subtrate solution prepared in 0.05 M sodium citrate sodium phosphate buffer with 0.1 M sodium chloride, at pH 4.3. The concentration of the substrate in the solution was $5x10^{-4}M$. The mixture was incubated at 37°C for two minutes. The reaction was terminated by the addition of 1 ml of 0.5 M glisin-NaOH buffer, pH 10.4 At this pH, maximum fluorescence of the liberated 4-methylumbelliferone (4-MU) was obtained and measured in an Amino-Bowmann spectrofluorometer. The solution was excited at the 365 nm and the resulting fluorescence was measured at 450 nm emission. Control experiments were performed where the substrate was incubated in the absence of enzymen. Four methylumbelliferon standard was used. The beta-galactosidase activity was expressed as M mol, 4-MU. per gm leukocyte protein per hour.

TABLE I

ACTIVITY OF BETA-GALACTOSIDASE IN PMN LEUKOCYTES

Subject	Beta-galactosidase*
Patient	72.5
Father	51.9
Mother	58.4
Newborn control	298.5–16.7 (6)
Adult control	214.4–24.7 (11)

* Beta-galactosidase activity was expressed as µ mol 4-methylumbelliferyl ester hydroyzed/g leukocyte protein/hr. The results were the mean ± S. E. M. of (n) number of experiments.

Protein concentration in leukocyte preparation was determined by the method of Lowry et al.[11]

The results of the activity of beta-galactosidase in the leukocytes of the patient and his parents were shown in (Table I).

Discussion

GM, gangliosidosis type I is a recessively inherited galactosidase enzymatic black acusing accumulation of gangliosides in various organs, especially in the brain.[2,5] The clinical signs and symptoms of this disease are generally present at birth. Patients with this syndrome have abnormal facial appearances with frontal bossing, depressed nasal bridge, marked hypertrophy of the alveolar ridges and maxillary processes, a large tongue, and low-set ears,[5] as evident in our case.

The "Cherry red spot" appearance was present at the fundus in 50 % of the reviewed cases.[4,5] Examination of the extremities indicated extension contractures and deformities similar to those observed in our case. Hepatosplenomegaly was found in some of these cases.[5] X-ray examinations, especially of the long bones, showed periosteal thickening, increased irregular trabeculation and demineralization. The "shoe shape" appearance was observed in the lateral skull X-ray[1,5,12] Laboratory findings of all the reviewed cases demonstrated vacuolated lymphocytes, as did the findings in our case.

In order to confirm our clinical diagnosis, blood samples were taken from the patient prior to his death as well as from his parents, normal newborns, and adults, for determining the activity of beta galactosidase in leukocytes. At the conclusion of the experiments, decreased enzymatic values were found in the patient and his parents. However, leukocyte beta galactosidase activity was noted to be higher in the patient than the parent. Similar difference was noted in the control subjects of newborn and adults (Table I). To explaine the differences between the newborn and adults we postulated that beta galactosidase activity was higher during the newborn period due to high sphingolipid turnover during this period.

GM_1 ganliosidosis type 1 has recently been detected in utero by Kaback et al.[12] One pregnancy was interrupted on the basis of amniotic fluid cell analyses, and diagnosis of GM gangliosidosis was confirmed in fetal tissues. Therefore, it is hoped that high risk pregnancies would easily be identified in the future.

Summary

This paper documents GM_1 gangliosidosis type I with clinical, radiological and necropsy findings, and reports the activity of beta galactosidase in the leukocytes of the patients and his parents as compared to the normal newborn and adults. We have postulated that beta galactosidase activity is higher in the newborn period due to high sphingolipid turnover.

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