CASE REPORTS

GM₁-generalized gangliosidosis variant with cardiomegaly

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Summary

A female infant with generalized GM₁-gangliosidosis differing in several aspects from previously reported cases is described. Clinically she is the first case to have cardiomegaly, unilateral congenital dislocation of the hip and normal facial appearance. She had a higher residual leucocyte β -galactosidase activity towards two synthetic substrates, namely *p*-nitrophenyl- β -D-galactoside (PNP- β -gal) and 4-methylumbelliferyl- β -D-galactoside (MU- β -gal) than in previously reported cases. Total β -hexosaminidase activity was raised in cultured fibroblasts, leucocytes, brain, kidneys and liver of the patient, and in fibroblasts from the parents. The parents' leucocytes had normal β -galactosidase activity towards PNP- β -gal but slightly reduced activity towards MU- β -gal. The parents' cultured fibroblast enzyme, however, had moderately low activities towards both substrates.

It is suggested that she may represent a genetic variant of generalized GM₁-gangliosidosis and that assay of leucocyte β -galactosidase activity should be added to the list of investigations for infantile cardiomegaly of unknown origin.

Introduction

Infantile generalized gangliosidosis (GM₁-gangliosidosis type 1) is an autosomal recessive disorder characterized by progressive cerebral degeneration, blindness and death before the age of 2 years. Hepatosplenomegaly, bilateral macular cherry red spots, extensive osseous changes and pitting subcutaneous oedema occur in most patients (O'Brien *et al.*, 1965; O'Brien, 1972). The monosialoganglioside GM₁ and its asialo derivative, GA₁, accumulate in the cytoplasm of neurones, histiocytes, hepato-

cytes and of renal glomerular epithelial cells (Ledeen *et al.*, 1965; O'Brien *et al.*, 1965; O'Brien, 1972). This has been attributed to a profound deficiency of lysosomal β -galactosidase which is required for the next step in the breakdown of GM₁ (Okada and O'Brien, 1968). Twenty-five cases have been reviewed by O'Brien (1972). A female infant who had certain features not previously described in the disorder is now reported. These features include cardiomegaly with endocardial fibroelastosis and unilateral dislocation of the hip.

Moreover, although she had a severe deficiency of β -galactosidase, the higher degree of residual enzyme activity in leucocytes, together with other features, suggests that she may represent a different genetic entity from that responsible for other reported cases.

Patient and methods

Case report

She had been born after a normal pregnancy and delivery but was small for dates, weighing 2500 g at 40 weeks' gestation. She had two half sisters aged 11 and 13 years from her father's previous marriage and a full sister aged 4 years. The parents were not consanguineous and there was no history of family illnesses.

At the age of 10 weeks she began to smile, vocalize and reach out for objects. There had been swelling of her hands, legs and feet and noisy breathing from birth, but otherwise she was considered to be normal.

At 4 months of age she became reluctant to feed and developed a temperature of 38.5°C. She appeared to be moderately ill, had plagiocephaly with a corresponding chest deformity, lumbar kyphosis, bilateral flexion deformity of the thumbs, limitation of abduction of the left hip and pitting oedema of the

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hands and feet. Her facies were unremarkable (Fig. 1a and b).

She had bilateral macular cherry-red spots surrounded by grey swollen retina. The liver and spleen were firm, smooth and enlarged to 4.0 cm below the costal margins. The heart sounds were normal but there was an intermittent grade 1 apical systolic murmur. The blood pressure was 95/70 mmHg. There was respiratory obstruction, mainly in the upper naso-pharyngeal region, tachypnoea, and costal recession. At 6 months of age her weight was 6.7 kg (25 percentile) (Tanner, Whitehouse and Takaishi, 1966) and her head circumference 44.0 cm (75th–90th percentile). Her mental development was assessed as being approximately at the 2-months level.

Radiographs revealed moderate bilateral cardiac enlargement and prominent pulmonary vascular markings radiating from both hila (Fig. 2a). There was lumbar kyphosis, anterior notching of the first lumbar vertebra (Fig. 2b) and subluxation of the left hip with hypoplasia of the acetabular roof. The periosteum of the humeri was slightly thickened. The ECG showed moderate right ventricular hypertrophy with upright T waves in V1–V6. Other investigations were as follows: sex chromatin, positive; CSF, normal; blood culture, no growth; serum proteins, total 5.7 g/100 ml, albumin 3.0 g/100 ml, globulin, 2.7 g/100 ml.

A 24-hr sample of urine (560 ml) contained 11.65

mg of 9-amino-acridine-precipitable polymeric glycosaminoglycan (GAG). On acid hydrolysis this yielded glucosamine : galactosamine : uronic acid : galactose with approximate molar ratios of 1.95 : 0.95 : 1.2 : 1 respectively; the 24-hr excretion of GAG uronic acid (0.5 mg) being within the normal limits.

She became increasingly listless, apathetic, anorexic and required tube feeding intermittently until death at 8 months of age. Post-mortem examination revealed bronchopneumonia to be the immediate cause of death. There was gross hepatosplenomegaly, moderate hypertrophy of both ventricles (Fig. 3a and b) and endocardial fibroelastosis, especially in the left ventricle but the valves and cordae tendinae appeared normal. There was no septal defect and the ductus arteriosus was closed. Histology of the liver revealed hepatocytes distended by cytoplasmic vesicles and Kupffer cells which were enlarged and contained PAS-positive, diastase-fast material. Many renal glomeruli contained swollen cells with pale foamy cytoplasm and swollen Bowman's capsules. The connective tissue of the adrenal cortex contained many macrophages whose cytoplasm was strongly PAS-positive and diastase-fast. Neurones of posterior root ganglia and of Auerbach's plexus of stomach (Fig. 4) and colon were swollen and contained fine basophilic granules. The haemopoietic marrow of the costo-chondral junction was severely distorted and contained very occasional cells.

FIG. 1a and b. Facies of the patient at 5 months of age. Tube feeding was because of progressive lethargy.

FIG. 2. (a) Radiograph of chest at 4 months of age showing moderate cardiomegaly; (b) lateral radiograph of spine at 4 months of age showing anterior notching and breaking of first lumbar vertebra.

FIG. 3. Anterior cardiac aspect (a) and the left ventricle (b) showing moderate hypertrophy of both ventricles and fibroelastosis of left ventricular endocardium.

FIG. 4. Auerbach's plexus of stomach showing large neurone with coarse granular material. Stain PAS. Magnification: microscopic $\times 320$, photographic $\times 2.5$, total $\times 800$.

with bulky or foamy cytoplasm. The brain was used for biochemical study and was not examined histologically.

Biochemical studies

Leucocytes from 15 ml of heparinized blood were isolated by sedimentation in dextran (Mellman *et al.*, 1964) and disrupted by alternate freezing and thawing three times. After mixing, aliquots were taken for assay of protein (Lowry *et al.*, 1951) β -D-hexosaminidase A and B (Kaback, 1972), or β -galactosidase, using either *p*-nitrophenyl- β -D-galactoside (PNP- β -gal) (Singer, Nankervis and Schafer, 1972) or 4-methylumbelliferyl- β -D-galactoside (MU- β -gal) as substrate. The last assay was a modification of the method of Leroy *et al.* (1972) using lactate buffer, 0.05 mol/l (instead of citrate-phosphate 0.018 mol/l) and MU- β -gal, 0.46 mmol/l (instead of 0.41 mmol/l). Fibroblasts were cultured as previously described (Babarik *et al.*, 1974). Starch gel electrophoresis was carried out by the method of Ho and O'Brien (1969).

Liver, brain and kidneys were frozen within a few hours of death and stored below -15°C until enzyme assay as described above or for extraction of gangliosides by the method of Suzuki, Suzuki and Kamoshita (1969) and assay after thin layer chromatography.

FIG. 5. Activities of fibroblast 4-methylumbelliferyl- β -galactosidase of (○), patient; (●) mother; (■), father and ---, 4 controls assayed simultaneously at different pHs.

Storage material

Total cerebral grey matter ganglioside NANA was 825 $\mu\text{g/g}$ wet weight (control 500 $\mu\text{g/g}$). Assays after thin layer chromatography revealed that in the patient GM₁ represented 70.6% of gangliosides (control 14.1%). Thus, in the patient there was an approximate 8.3-fold increase in grey matter GM₁.

Enzyme activities

β -galactosidase activities of cultured fibroblasts from the patient, her parents and four controls are shown in Fig. 5. At the optimal pH, 4.0, fibroblast 4-methylumbelliferyl- β -galactosidase activity of the patient was 4.15 nmol of 4-methylumbelliferone released/h/mg of protein (units) or 0.56% of the control mean (744 units; range 630–830 units). The activity of the father's cells (519 units) was clearly below that of the normal range (69.8% of control mean), but that of the mother (608 units) was only slightly below (81.7% of control mean).

Fibroblast total β -hexosaminidase activities (Fig. 6) in the patient were 15.2×10^3 nmol of 4-methylumbelliferone released/h/mg of fibroblast protein (units) or 145% of the control mean (10.44 units; range 8.0–12.30 units). Activities of both the A and B components were elevated. Intermediate rises in activity were observed in the parents (father 12.42 units; mother 14.34 units).

Leucocyte β -galactosidase was assayed using both PNP- β -gal and MU- β -gal as substrates. Although the two methods give similar results for controls

FIG. 6. Fibroblasts β -hexosaminidase activities of patient, parents and four controls assayed simultaneously.

(Table 1) (mean activities for the two methods 105.9 and 107.7 units respectively), appreciably greater values were obtained for the patient and parents when PNP- β -gal rather than MU- β -gal was the substrate. Indeed for the parents, the mild reduction of activity towards MU- β -gal (68.5% and 78% of control mean by father and mother respectively) was not detected towards PNP- β -gal. The patient had 7.8% of control activity towards the former substrate and 17.0% towards the latter.

Considerable reduction of 4-methylumbelliferyl β -galactosidase and elevation of total hexosaminidase activities were also observed in the patient's liver, brain grey matter and kidney (Table 2).

Discussion

The patient has several clinical features which are typical of generalized GM₁-gangliosidosis. These include cherry-red macular spots, vertebral changes characteristic of the Hurler syndrome, pitting

TABLE 1. Leucocyte β -galactosidase activities

	<i>p</i> -nitrophenyl β -galactosidase (units) *	4-methylumbel- liferyl β -galacto- sidase (units) †
Control mean (n = 12)	105.9	107.7
Control range	93.6-118.2	88.8-139.2
Patient	18.0 (17.0%)†	8.54 (7.8%)
Mother	108.0 (102.0%)	84.0 (78.0%)
Father	106 (100.8%)	73.8 (68.5%)

* 1 unit of activity is defined as nmol of either *p*-nitrophenol or 4-methylumbelliferone released/hr/mg of leucocyte protein.

† Figures in parentheses indicate percentage of control means.

TABLE 2. Tissue enzyme activities

	β -galactosidase (units)*		β -glucosaminidase (A+B) (units) * $\times 10^{-3}$	
	Control	Patient	Control	Patient
Liver	237.0	4.5 (1.90%)*	2.44	22.68 (931%)†
Brain	93.0	3.7 (4.00%)	3.90	10.26 (263%)
Kidney	243.6	12.1 (4.98%)	11.64	45.96 (395%)

* One unit of activity of either enzyme is defined as nmol of 4-methylumbelliferone released/hr/mg of tissue protein.

† Figures in parentheses indicate percentage of controls.

oedema, since birth, upper respiratory tract obstruction and a progressive course with early death. Cardiac enlargement, however, which is characteristic of some storage diseases due to lysosomal enzyme deficiency, has not been reported in other infants with GM₁-gangliosidosis (Craig, Clarke and Banker, 1959; Norman *et al.*, 1959; Landing *et al.*, 1964; O'Brien *et al.*, 1965; Gonatas and Gonatas, 1965; Sacrez *et al.*, 1967; Scott, Lagunoff and Trump, 1967; Attal *et al.*, 1967; Grossman and Danes, 1968; Seringe *et al.*, 1968).

It is invariable in Pompe's disease (glycogenosis type 2; α 1,4-glucosidase deficiency) and has been described in Sandhoff's disease (β -hexosaminidase A and B deficiency) (Bliden *et al.*, 1974). At post-mortem examination both ventricular walls were found to be thickened and there was endocardial fibroelastosis.

Other unique clinical features of the patient included unilateral dislocation of the left hip (bilateral dislocation has been reported (Craig *et al.*, 1959; Seringe *et al.*, 1968) in two infants) and absence of abnormal facies (Fig. 1a and b). A 'coarse'

facial appearance has been noted in all eighteen patients where the facies have been described (O'Brien, 1972).

The histological findings included neuronal lipidosis (Fig. 4), visceral histiocytosis, foam cells in the marrow and glomerular changes, all of which are characteristic of GM₁-generalized gangliosidosis.

β -galactosidase activity of leucocytes was reduced, but the mean residual activity was higher when PNP- β -gal rather than MU- β -gal was the reaction substrate. The scant number of values in the literature tend to support this finding, although reported reductions of activity towards both substrates are appreciably lower than in our patient. Thus, in a patient with generalized GM₁-gangliosidosis when PNP- β -gal was the substrate, Singer *et al.* (1972) found residual leucocyte β -galactosidase activity of 10.4% of control mean (present patient, 17.0%); and in six patients when MU- β -gal was the substrate, Young, Ellis and Patrick (1972) found 0.2-8% (present patient, 8.8%). A similar trend may be noted in two reported parents who had levels of 36.4% and 44.2% towards the PNP substrate (Singer *et al.*, 1972) (parents of present patient 100.8% and 102.0%) and in both parents from two families, 35.3% to 47.6% towards the MU substrate (Young *et al.*, 1972) (parents of present patient 68.5% and 78.0%). In the present patient there was a greater reduction of fibroblast MU- β -galactosidase (0.56% of control mean) than of the leucocyte enzyme (7.8% of control mean).

The occurrence of genetic heterogeneity is now well recognized among inborn errors of metabolism previously considered to represent one entity. The criteria which suggest heterogeneity include variation in the amount of residual activity of the mutant enzyme in patients from different families (Benson, 1973). The higher levels of β -galactosidase activity in the leucocytes of the present patient than in cases previously reported, together with bizarre clinical features not previously described in generalized GM₁-gangliosidosis, suggest that this patient may be a new genetic variant of this disorder.

An elevation of β -hexosaminidase activity previously noted in liver and brain (O'Brien, 1972) was observed in this patient, also in fibroblasts, leucocytes and kidney, and is unexplained. In fibroblasts, enhancement was due to both the A and B enzymes. An intermediate rise was observed in the parents' fibroblasts. This suggests that demonstration of raised fibroblast β -hexosaminidase activity could aid detection of heterozygotes when β -galactosidase levels are found to be only slightly decreased as was the case in the parents of the patient. Assay of leucocyte β -galactosidase activity should be added to the list of investigations for infantile cardiomegaly of unknown origin.

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Acute rheumatic fever following streptococcal wound infection

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Summary

A case of acute rheumatic fever with pancarditis secondary to infection of abrasions on the hand with β -haemolytic streptococci is described.

Introduction

Despite the decline in recent years in the incidence of rheumatic fever, interest in the disease is unabated because of its unique relationship to streptococcal infections. Epidemiological evidence suggests that rheumatic fever occurs only after streptococcal pharyngitis, but not in association with streptococcal skin infections such as impetigo.

The purpose of this paper is to describe a patient who developed acute rheumatic fever following infection of abrasions, and to draw attention to the possible danger of acute rheumatic fever in meat handlers who are liable to develop streptococcal infection of cuts on the hand.

Case report

A 17-year-old butcher was admitted to Bedford General Hospital with a history of fever and

generalized joint pains. Between 2 and 3 weeks earlier he had sustained small cuts on both hands at work. These cuts had become infected and had failed to heal. Five days before admission he developed a fever and left shoulder pain. The following day he had pain and stiffness in both knees, ankles and feet, and 2 days later sharp stabbing retrosternal pain aggravated by movement, deep inspiration and coughing. There was no family or past history of rheumatic fever.

On admission, he was flushed and pyrexial with rapid shallow breathing. There was a single infected abrasion on the extensor surface of each hand. Pulse regular—110/min. Blood pressure—140/90. Apex beat was displaced outside the left mid-clavicular line. Heart sounds normal. Loud pericardial friction rub audible over the praecordium. The jugular venous pressure was raised to 4 cm with patient erect. Movement of shoulders, knees, elbows, wrists, fingers and toes was painful and restricted. There was no abnormality in the respiratory, alimentary or nervous systems.

Investigations. Haemoglobin 13.6 g/100 ml. White Cell Count 7,200/mm³. ESR 75 mm/hr (Westergren). ASO titre 500 Todd units. Throat swab

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