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BRIEF REPORT

LIVER TRANSPLANTATION FOR TYPE IV GLYCOGEN STORAGE DISEASE

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TYPE IV glycogen storage disease is a rare autosomal recessive disorder (also called Andersen's disease[1] or amylopectinosis) in which the activity of branching enzyme alpha-1, 4-glucan: alpha-1, 4-glucan 6-glucosyltransferase is deficient in the liver as well as in cultured skin fibroblasts and other tissues.[2,3] This branching enzyme is responsible for creating branch points in the normal glycogen molecule. In the relative or absolute absence of this enzyme, an insoluble and irritating form of glycogen, an amylopectin-like polysaccharide that resembles plant starch, accumulates in the cells. The amylopectin-like form is less soluble than normal glycogen, with longer outer and inner chains and fewer branch points. The clinical onset of the disease is insidious, with nonspecific gastrointestinal symptoms at first, followed by progressive hepatosplenomegaly, portal hypertension, ascites, and hepatic failure. Children with this disorder usually die of hepatic cirrhosis by the age of two to four years.[4-8] In exceptional cases, cardiomyopathy,[5-7,9] neurologic syndromes — including tremors, seizures, and dementia[10,11] — or variable manifestations of myopathy[5,12,13] have been reported. In patients with these unusual symptoms, the clinical onset is frequently later than in typical cases, and death most often results from cardiac failure.

Liver transplantation for Type IV glycogen storage disease was attempted in 1972; the recipient died 110 days later after the rejection of the first liver transplant and attempted retransplantation.[14] Liver transplantation was first performed successfully in September 1984 in Patient 1 of this series; since that time we have treated six more such patients. Our experience with these seven patients forms the basis of this report.

CASE REPORTS

All the patients were boys, including two sets of brothers (Table 1). All had progressive liver failure with massive hepatomegaly, splenomegaly, and ascites. (Because patients with Type IV glycogen storage disease but without progressive liver disease have occasionally been reported,[16] progressive liver disease was a condition for transplantation in each case.) Two of the seven patients died 7 and 36 days after liver transplantation — from a bowel perforation and thrombosis of the hepatic artery, respectively. The five other recipients (71 percent) are healthy and have normal liver function 16 to 73 months after transplantation. The five transplant recipients who survived were hospitalized for 26 to 49 days (mean, 40). The mean (±SD) cost of transplantation in these five patients, including all professional fees, was $107,900±30,000. Rehospitalization was

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not required except for follow-up percutaneous needle biopsies of the liver, performed 1 to 45 months after transplantation, or transjugular endomyocardial biopsies, performed 3 weeks to 54 months after transplantation (Table 2). The transplantations and follow-up biopsies were undertaken with informed parental consent. The biopsy procedures were considered essential for optimal care of the patients and were not part of a prospective experimental design.

Height, weight, gross and fine motor development, and language ability were retarded in all patients before transplantation. The physical strength and motor skills of the five survivors improved steadily after transplantation. All have had a normal growth rate, two are performing well in elementary school, and the three in preschool have no apparent abnormalities in intellectual development. None has had any cardiac symptoms or any evidence of abnormalities on chest films or electrocardiographic and echocardiographic examinations. A magnetic resonance imaging scan of the heart in one child was normal more than four years after transplantation.

Methods

Liver Transplantation

The diseased liver was totally excised and replaced with a size-matched cadaveric liver, which was revascularized in an anatomically normal manner and provided biliary drainage with a Roux-en-Y choledochojejunostomy.[17] Immunosuppressive therapy consisted of cyclosporine and prednisone. The daily maintenance dose of prednisone in the five surviving patients was 5 mg or less.

Tissue Studies

Cultured skin fibroblasts and homogenates of liver tissue from the patients were assayed at Washington University for branching-enzyme activity. The phosphorylase-coupled assay measured the rate of formation of inorganic phosphate from glucose-1-phosphate as glucose was polymerized to glycogen by the tissue homogenates.[3,6,15] The branching-enzyme activity in skin fibroblasts averaged 0.08 μmol of inorganic phosphate per minute per milligram of protein, which was less than 10 percent of the activity in normal subjects (Table 1).

Amylopectin was sought on light-microscopical examination in the excised livers, in liver-biopsy samples obtained after transplantation, and in other tissues obtained at the time of transplantation or at autopsy. The characteristic inclusions were positive on periodic acid–Schiff staining after digestion with diastase (PAS-D). Electron-microscopical studies demonstrated the fibrillar aggregations that are typical of amylopectin.[18-20]

Morphometric studies were performed on PAS-D–stained myocardial tissue obtained at autopsy from one patient and endomyocardial-biopsy samples from three others obtained at various times after transplantation (Table 2), with the Bioquant System IV (R&M Biometrics, Nashville) used for automated image analysis. The biopsy samples were also separately evaluated with the point-count technique. To eliminate artifacts caused by vessels or compression, interstitial and vascular spaces in the field were eliminated from the final calculations of the area of amylopectin deposition. Variation within samples was studied in 24 fields from a large piece of left ventricle obtained at autopsy from Patient 3. All the fields had much the same amylopectin distribution, with the single exception of a superficial subendocardial field that contained little muscle.

Table 1. Clinical Data on Patients with Type IV Glycogen Storage Disease Who Underwent Liver Transplantation.

Patient No.*	Age at Transplantation	Skin-Fibroblast Branching-Enzyme Activity†	Date of Operation	Length of Survival
	mo	*μmol/min/mg*		
1	31	0.10	9/6/84	73 mo
2	11	0.07	8/28/85	Died 7 days after transplantation
3	36	0.11	9/5/85	Died 36 days after transplantation
4	46	0.11	11/25/85	58 mo
5	22	0.08	10/24/87	35 mo
6	20	0.06	3/21/89	18 mo
7	34	0.09	5/26/89	16 mo

*Patients 1 and 6 were brothers, as were Patients 3 and 5.

†Normal branching-enzyme activity in skin fibroblasts, 1 to 2 μmol per minute per milligram of protein.[7,15]

Table 2. Deposits Positive on Periodic Acid–Schiff Staining after Digestion with Diastase (PAS-D) in the Myocardium of Four Patients with Type IV Glycogen Storage Disease after Liver Transplantation.

Patient No.	Time after Transplantation	No. of Inclusions	Mean Area Occupied by Inclusions	Mean Size of Inclusions
	mo	*per 3.6×10³ μm²*	*%*	*μm²*
1	54.0	24	0.5	140
3	1.2 (at autopsy)	46	2	141
6	0.9	332	13	146
	14.0	129	5.9	164
7	1.0	Too few to quantitate	—	—
	11.0	Too few to quantitate	—	—

Results

There was no trace of amylopectin in the graft of the child who died 36 days after transplantation from hepatic-artery thrombosis or in the liver-biopsy specimens obtained from the five surviving children 1 to 45 months after transplantation. The biopsy specimens also showed no evidence of rejection.

Amylopectin deposits were found in all biopsy specimens of skin (including arrectores pilorum muscles), jejunum, and skeletal muscle obtained at the time of transplantation. Postmortem studies in Patient 3, who died 36 days after transplantation at 36 months of age, showed amylopectin in the esophagus, bowel, bladder smooth muscle, skeletal muscle, central nervous system, peripheral nerves, and heart. In this patient, amylopectin occupied about 2 percent of the myocardial area (Table 2).

Myocardial samples were available from three other patients (Table 2). Patient 1, whose liver was replaced when he was 36 months old, had a nearly amylopectin-free myocardial-biopsy specimen 54 months later. His younger brother (Patient 6) had involvement of 13 percent of the myocardial area three weeks after transplantation (Table 2). A year later, the amount of amylopectin had been reduced to 5.9 percent (Fig. 1, Table 2). In Patient 7, the myocardial-biopsy specimen obtained one month after liver transplantation showed very few deposits in the myocytes; 10 months later, there was no change.

Discussion

The absence of amylopectin deposition in the liver grafts was expected, but the freedom from neuromuscular or cardiac morbidity associated with extrahepat-

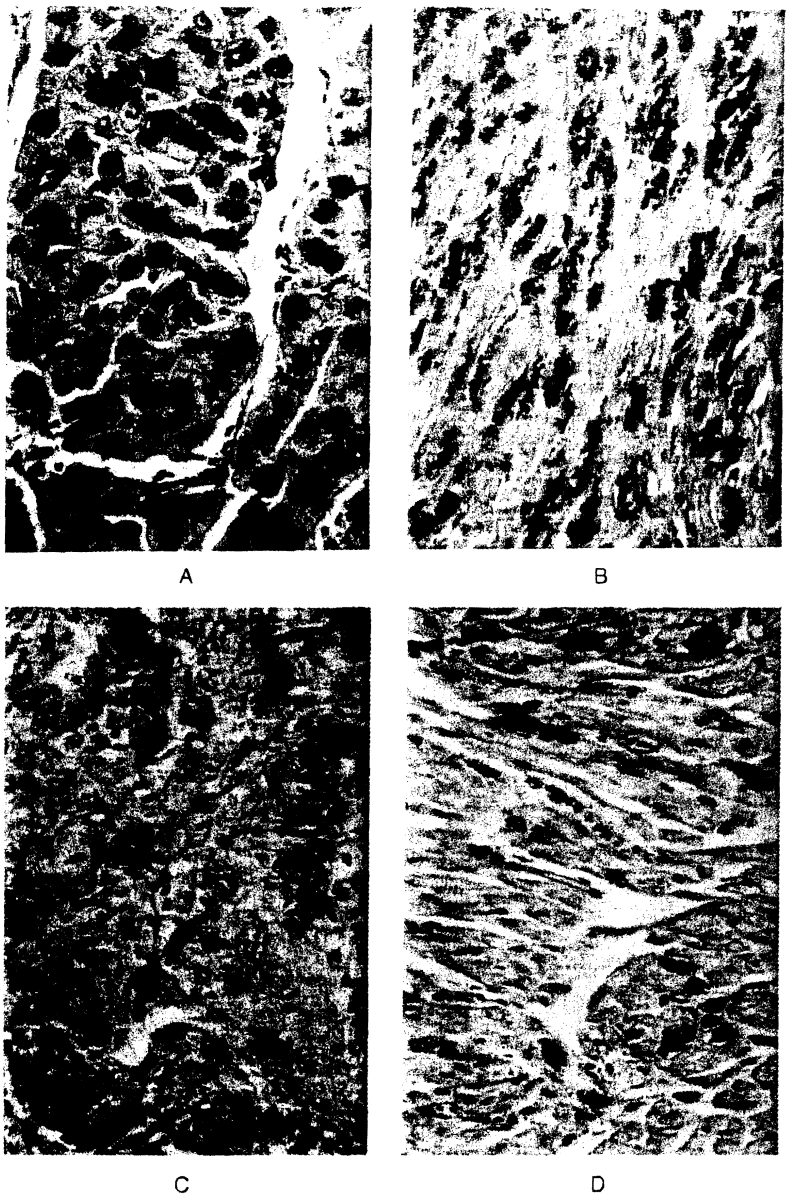

Figure 1. Myocardial Biopsy Specimens from Patient 6 (PAS-D, ×405).
Transverse sections (Panel A) and oblique sections (Panel B) of myocardial fibers in a biopsy specimen obtained 3½ weeks after liver transplantation show abundant amylopectin inclusions (red material). Panels C and D show comparable fields from a biopsy specimen obtained 14 months after transplantation, in which there is evidence of a marked decrease in the number of inclusions.

ic amylopectin deposits was more noteworthy. No neuromuscular complications were seen, and the retarded growth that was characteristic of the patients before transplantation was restored to more nearly normal values in all the survivors. More important, none of the surviving patients has had cardiac complications during follow-up periods as long as six years. Because a wide variety of cardiac and noncardiac tissues collected perioperatively from different patients contained cytoplasmic amylopectin, it must be assumed that all the patients had cardiac involvement, as was demonstrated in the four patients studied. However, there was little amylopectin in myocardial-biopsy specimens obtained a year or more after liver transplantation. Particularly striking was the decrease in myocardial amylopectin over a period of 13 months after liver transplantation in Patient 6, whose older brother had minimal heart involvement 54 months after transplantation. We speculate that the older brother may have cleared amylopectin from his extrahepatic tissues by the time this first heart biopsy was performed. That amylopectin disappears slowly is illustrated by the cases of our patients and may account for unpublished observations in a 14-month-old Belgian child who died of a respiratory infection superimposed on congestive heart failure 11 months after liver transplantation. Amylopectin was found post mortem in this child's myocardium and in other organs (Otte JB, University of Louvain Medical School, Brussels: personal communication).

These observations could have implications for other enzyme deficiencies that affect multiple organs, particularly if cell-to-cell transfer of the deficient enzyme can be demonstrated. However, any explanation for extrahepatic amylopectin clearance must await a better understanding of the paradoxical enzymologic features of Type IV glycogen storage disease, in which a branching-enzyme defect results in glycogen with fewer than normal branches.[2] Furthermore, the distribution of the normal and abnormal branching enzymes may be different in various tissues.[21]

Finally, it is possible that long-term cyclosporine therapy was an ameliorating factor in our patients. This drug,[22] as well as the new immunosuppressive agent FK 506,[23] has hepatotrophic qualities similar to those of insulin,[24] including the ability to increase hepatic glycogen stores. Both these immunosuppressive drugs attach to cytosolic binding sites that are rich in the ubiquitous enzyme peptidyl-prolyl isomerase,[25,26] and both cause wide-ranging immunologic and nonimmunologic effects, including alterations in carbohydrate, cholesterol, and uric acid metabolism.[23] Thus, it is conceivable that the early treatment of patients with Type IV glycogen storage disease could obviate the need for transplantation.

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