FOETAL LIVER TRANSPLANT IN FABRY'S DISEASE

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Summary

Two patients, 33 and 26 years old, presented with Fabry's disease and minimal renal involvement. They were treated with a transplant of foetal liver cells exhibiting normal enzymatic activities, after plasma transfusions and symptomatic therapies had proved ineffective. In the first patient, objective and subjective clinical symptoms were significantly improved: sweating appeared, cutaneous lesions seemed slightly decreased and pains disappeared. In the second patient, pains were also seemingly decreased. The mechanism which may be held responsible for improvement of our patients, as of recipients of a kidney transplant, is not completely elucidated. The cells, rather than steroids or azathioprine, seemed to be responsible for the improvement.

Introduction

Fabry's disease is a recessive X-linked inborn error of glycosphingolipid catabolism characterised by the systemic accumulation of the neutral glycosphingolipid, ceramide dihexoside and trihexoside in organs and tissues. This accumulation is due to a deficiency of the specific lysosomal α galactosidase-A isoenzyme. This metabolic disorder results in systemic manifestations and the involvement of the kidney usually leads to chronic renal failure during the fourth decade of life.

Transplantation of a normal kidney has been suggested partly to correct the metabolic disorder [1-4]. Two of our patients, with as yet only slight renal involvement, have been treated with foetal liver transplantation.

Case—Reports

As shown in Table I, the two young male patients presented with typical clinical manifestations of Fabry's disease. The diagnosis was confirmed by ophthalmologic,
TABLE I. Case Reports

1 LAG. J.P. Born on August 15, 1944
   Age (yrs)
   5    painful crisis of limbs
   21   left deafness and initial cutaneous lesions
   24   bilateral sympathectomy without effect on pains
   26   haematuria on two occasions
   27   diagnosis of Fabry’s disease
   28   left hemiparesis disappearing without sequelae
   28–31 increased cutaneous involvement, complete anhidrosis, frequent
   pains and subsequent psychological manifestations
   31   foetal liver transplant (October 1975)

2 SCH. C. born on July 14, 1952
   Age (yrs)
   14   appearance of typical cutaneous angiomas
   16   polyarthralgia
   21   diagnosis of Fabry’s disease
   26   aphasia and hemiparesis which progressively disappeared with few sequelae
   26   foetal liver transplant (June 1978)

histological, and enzymatic analyses. On slit lamp examination, the first patient
had tortuous vessels with aneurysms, and the second patient’s sisters had corneal
opacities.

Cutaneous biopsy demonstrated polymorphic lipid inclusions in the arterial
walls. In particular dense inclusions with periodical laminations at 65–75 Å intervals
were seen [5]. The kidney histology also showed lipid inclusions in glomeruli,
in the cytoplasm of the proximal and the distal tubular epithelium and in the
wall of small arteries. By electron microscopy, inclusions were comparable with
those seen in the cutaneous specimen [6,7]. Enzymatic studies, performed ac-
ccording to methods of Hó et al and Desnick et al [8,9], proved the lack of thermo-
labile α galactosidase A. The results in the two families are shown in Table II
and support the view that the mothers of the two patients and sisters of the
second patient are heterozygotes, while the two patients are hemizygotes. Enzym-
atic studies gave confirmatory results in serum, urine, fibroblasts and lympho-
blastoid cell lines from both patients. Trihexoside ceramide in urine was increased.
The renal function was virtually unaltered. In the first patient, the creatinine

TABLE II. Alpha-Galactosidase Activity in Leucocytes

<table>
<thead>
<tr>
<th>Leucocytes</th>
<th>LAG.</th>
<th>SCH.</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mother</td>
<td>Propositus</td>
</tr>
<tr>
<td>α galactosidase activity (nanomoles/min/mg of protein)</td>
<td>0.56</td>
<td>0.06</td>
</tr>
</tbody>
</table>

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clearance was slightly reduced (75ml/min), and there was moderate proteinuria with a glomerular-type electrophoretic pattern. In the second one, the renal function was normal, without proteinuria.

**Foetal Liver Transplant**

After symptomatic treatment had proved ineffective a foetal liver transplant was performed in October 1975, in the first patient, and in June 1978, in the second patient. Foetal liver cells from a 15 week-old female foetus and from a 17 week-old male foetus were resuspended in culture medium and injected under the aponeurosis of the rectus abdominis muscle.

At the time of injection, the cell viability was greater than 95%, as judged by the trypan blue exclusion assay. The patients received azathioprine (3mg/kg/day) and prednisone (0.25 then 1mg/kg/day progressively tapered), started one week prior to transplantation. The first patient was also given 4 vials of antilymphocyte globulins.

**Results**

Improvement in the first patient included the following changes: sweating appeared, became and remained normal, pains completely disappeared, cutaneous lesions seemed slightly decreased. This improvement has persisted for 3½ years after foetal liver transplantation.

In the second patient, treated one year ago, it is too early to evaluate the possible clinical benefit from the procedure, especially since this patient had almost normal sweating and relatively few pains except in the cold season. However the patient has mentioned a single and very mild pain over the whole year following transplantation. In addition no neurological manifestation has occurred since foetal liver transplantation. The viability of the transplanted foetal liver cells was monitored by measuring circulating α foetoprotein (A.F.P.) - (Figure 1). Levels of A.F.P. rose sharply (1–50ng/ml) and then decreased progressively over more than 6 weeks while the cells matured. In the first patient a dramatic diminution of A.F.P. level was interpreted as evidence of rejection and was treated with an increased dosage of steroids which resulted in a further rise of the A.F.P. level.

The α galactosidase A activity was not significantly increased in either leucocytes or sera. Dihexoside and trihexoside ceramides were only mildly altered in urines and slightly decreased in the sera. Renal function seemed stable during the observation period, with serum creatinine levels below 120μmol/L and unmodified proteinuria in the first patient.

**Discussion**

The two patients reported had typical Fabry’s disease with clinical, ophthalmological, enzymatic and histological manifestations.

Plasma transfusions and symptomatic therapy were of virtually no benefit.
The first patient had neurological involvement, very frequent pains and developed ideas of suicide. He demanded a kidney transplantation. Because of his nearly normal renal function, and because of the normal enzymatic activities in foetal liver [10], the patient was transplanted with foetal liver cells [11]. He was informed of the uncertainty of the result and of the need for prolonged immunosuppressive therapy. The second patient was also treated after informed consent.

Monitoring of AFP levels proved useful to ascertain persisting function of the transplanted cells over the first month. Efficacy of treatment was mostly documented by the clinical improvement: decreased pains, reappearance of sweating, and absence of any neurological manifestations following the transplant. Corticosteroids and azathioprine do not appear to be responsible for this effect [12] and when given alone to the second patient for a short period no modification was noted. The transplanted cells themselves therefore may be held responsible for the apparent clinical improvement.

A lack of major improvement in histological lesions would be expected since a kidney with glycosphingolipid deposits is not cleared even after transplantation into a non-Fabry Recipient [13]. The modifications of α-galactosidase-A activity in leucocytes or sera and of di- and trihexoside ceramides were moderate, inconstant, and due to various factors (e.g. correction of haemolysis) following kidney transplantation, [1,12,14–16]. Major alterations of these parameters were not expected in our patients and we were not surprised to observe only moderate
modifications of biochemical results.

The possible mechanism for this improvement in our patients is not yet fully elucidated. Several hypotheses can be formulated: activity in situ in the transplanted cells themselves, enzyme induction in other cells, or, more likely, 'colonisation' by lysosomal enzymes as suggested by in vitro experiments [17]. Clinical follow-up for several years and further in vitro analyses in co-culture experiments are needed to determine the significance of this therapeutic approach to enzyme-deficiency diseases, an approach that may be needed until other methods (incorporation of modified enzyme molecules or use of D.N.A. recombinant techniques) are developed.

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References

4 Simmons, RL, Desnick, RT, Najarian, JS and Krivit, W (1973) Cours international de transplantation, Page 59
5 Perrot, H, Schmitt, D and Thivolet, J (1973) Lyon Medical, 229, 581
7 Malik, MC (1977) Thèse Médecine Lyon
8 Ho, MW, Beutler, S, Tennant, L and O'Brien, JS (1972) Amer. J. Human Genet., 239, 207

Open Discussion

WESTBROEK (Co-Chairman) Have your two patients been on immunosuppression with steroids before the foetal liver transplant?
MALIK  Yes, one week prior.

WESTBROEK  One week. The question you bring up is whether it is the transplant or the steroids and immunosuppressive treatment. Was the schedule of prednisone and azathioprine the same dosage as before the transplant?

MALIK  Yes, it was.

WOODS (Leicester, United Kingdom)  In pancreatic islet transplantation, direct injection of cells into the portal vein has been used. Have you considered this as possibly a more effective way of giving your foetal liver transplant?

MALIK  No.