BODY IRON STORES IN CHILDREN WITH CHRONIC RENAL FAILURE IN RELATION TO HLA PHENOTYPES

D E Müller-Wiefel, V Lenhard*, K Schärer

Children’s Hospital and *Institute of Immunology,
University of Heidelberg, FRG

Summary

In 57 children with chronic renal failure (19 on conservative treatment, 25 haemodialysis and 13 after transplantation) body iron stores were determined by an immunoradiometric assay with a heterologous antibody system in relation to haemochromatosis alleles, HLA A3 and B7. Iron overload, predominantly found during haemodialysis, depended on the number of erythrocyte transfusions given and was found to be more pronounced in patients with HLA A3 and/or B7.

The frequency of these antigens was significantly higher in patients with iron overload (91%) than with normal (43%) or decreased (44%) iron stores. The relative risk of iron overload was calculated to be 2.0 for HLA A3 and 8.7 for HLA B7. The results suggest that erythrocyte transfusion therapy should be minimised in children with haemochromatosis alleles in order to avoid organ damage by haemosiderosis.

Introduction

Body iron stores (BIS), specially those of the liver [1], consisting of ferritin and haemosiderin are well reflected by serum ferritin (SF) concentrations. The strong correlation between storage iron and SF is disturbed only by liver disease, infections and parenteral iron therapy [2]. In chronic renal failure (CRF) BIS are increased by reduced erythropoiesis, haemolysis, blood transfusions and uncritical iron therapy. They are decreased by blood loss from the gastrointestinal tract, through the dialyser and blood sampling. BIS were recently reported to be genetically determined by the HLA antigens HLA A3 and B7 [3,4], predisposing to idiopathic haemochromatosis.

The aim of our study was to evaluate the genetic influence on BIS in CRF comparing SF levels with haemochromatosis alleles.

524
Patients and methods

We investigated 57 children with CRF over a mean period of 14 months on conservative treatment (CT), on intermittent haemodialysis (HD) 5 hours thrice weekly (Haemoflow C 0.8 or 1m²) and after renal transplantation (TP) (Table I).

<table>
<thead>
<tr>
<th>TABLE I. Patients</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td>Conservative</td>
</tr>
<tr>
<td>treatment (CT)</td>
</tr>
<tr>
<td>Haemodialysis</td>
</tr>
<tr>
<td>(HD)</td>
</tr>
<tr>
<td>Transplantation</td>
</tr>
<tr>
<td>(TP)</td>
</tr>
</tbody>
</table>

* Median value of blood transfusions per patient: * = 2.4, † = 3.0

The patients were subdivided into those with HLA A3 and/or B7 and those without these HLA alleles. Both groups were comparable with respect to age, haemoglobin, serum creatinine and mean transfusion rate. At the time of investigation none of the children had evidence of active liver disease, infection or was being treated by oral or parenteral iron.

SF levels were determined at an average interval of three months by an immunoradiometric assay with a heterologous antibody system (Het IRMA) applying an unlabelled antibody directed against liver ferritin and a labelled antibody (125I) directed against placenta ferritin (Riagnost, Behring-Werke, Marburg, FRG). The SF determination by Het IRMA seems to be more representative of the sum of isoferritins pooled in the serum than the determination by a homologous antibody system with antibodies usually directed against spleen ferritin [5].

HLA type was determined by a standard microlymphocytotoxicity technique. The statistical evaluation was based on t-test after logarithmic transformation; differences of percentages were calculated by the χ²- (n>60) or the Fisher exact test (n<60). According to our own previous results in childhood with this assay [6] iron deficiency (ID) was defined by SF values below 40μg/L, normal iron stores by SF between 40 and 300μg/L and an evident iron overload (IO) by SF above 1000μg/L.

Results

On CT as well as on HD and after TP only half the children had persistently normal iron stores (58%, 52% and 47%, respectively). ID was most frequent on CT with 42%, less pronounced after TP (38%) and of minor importance on HD (12%). On the contrary IO was mainly a problem on HD (36% of all children); it was less important
after TP (15%) and never detected in children on CT. The percentages of HLA A3 and/or B7 positive patients among the different treatment groups were more or less comparable: 47% (CT), 52% (HD) and 62% (TP). The geometric means of SF levels in the patients in relation to HLA A3 and B7 are given in Figure 1.

Figure 1. Geometric means of serum ferritin (SF) values of haemochromatosis alleles positive and negative patients on conservative treatment (CT), on intermittent haemodialysis (HD) and after renal transplantation (TP)

On CT and after TP the mean SF values of the two different HLA groups ranged within normal limits and did not significantly differ from each other. However, on HD a significant SF elevation up to 509μg/L in the haemochromatosis alleles positive patient group was found, in contrast to HLA A3 or B7 negative patients with a mean value of only 180μg/L. On the other hand HLA A3 and/or B7 were normally distributed in children with decreased or normal BIS (Figure 2). In IO the haemochromatosis alleles were significantly more frequent compared to patients with normal or decreased iron stores.

Discussion
Body iron stores in children with CRF depend on the mode of treatment. Whereas ID is a major problem on CT and after TP, overloading of iron stores is important mainly on HD and only of minor interest after TP and absent in CT. A strong correlation exists between SF values and the number of erythrocyte transfusions given on HD (Figure 3). It is of interest that in the presence of IO (SF>1000μg/L)
Figure 2. Proportions of haemochromatosis alleles (HLA A3, B7) positive patients (black) in different degrees of iron stores

Figure 3. Positive correlation between longitudinally measured serum ferritin (SF) values and the number of erythrocyte transfusions (250ml) given at the time of investigation in children on intermittent haemodialysis
the linear relationship between these parameters is lost. Therefore other factors seem to influence the degree of IO. Bregman et al found in adult patients on HD a genetic influence on BIS by comparing the frequency of haemochromatosis alleles (HLA A3, B7, B14). The presence of these alleles was associated with iron overload. Our results confirm these findings for paediatric patients on HD, but not on CT. In addition they demonstrate that this genetic influence has no bearing on BIS during the period of CT and is of questionable interest after TP.

From our data the antigen frequency for HLA A3 and B7 in iron overload patients was 91%. This antigen frequency was significantly higher not only compared with patients with normal and decreased iron stores (43%) but also with the normal European population (39%) [8]. The total relative risk of IO in CRF children was calculated for HLA A3 as 2.0 and for HLA B7 as 8.7. On CT there was no risk of IO. Under HD the relative risk of IO was 1.8 for HLA A3 and 7.1 for HLA B7.

The clinical importance of IO is mainly based on its dangerous potential damage on the liver [7] and perhaps on other organs (endocrine system, heart, muscle) which can be reduced by lowering the frequency of transfusions. Our data indicate that in CRF children with HLA A3 and, especially with B7, are at a high risk to develop IO on HD and therefore a restricted transfusion policy seems to be indicated in patients possessing these alleles.

References

6 Müller-Wiefel DE. Ped Res 1980; 14: 1427
7 Ali M et al. JAMA 1980; 244: 343
8 Terasaki PT. Histocompatibility testing 1980. The Reprints of the University of California 1980: 962

Open Discussion

MICHALK (Erlangen, FRG) Did you compare iron overload with the underlying disease, which may influence the need for blood transfusion?

MÜLLER-WIEFEL We did not find any correlation between the primary renal disease and iron overload. On the other hand, we could find a relationship between iron deficiency and the primary renal disease. Especially with conservative treatment the incidence of iron deficiency was much higher with urinary tract obstruction. One can explain this fact by better erythropoiesis in these children in whom the renal parenchyma is not so damaged. There was one patient in our study who was bilaterally nephrectomised and this was the only patient who showed iron overload, although he did not possess HLA A3 and B7.
CHANTLER (London) If I understood your slide correctly you have some patients with very high serum ferritin levels, and we do, as well. We are worried about it but not quite sure what we should do and I wonder if you have got any experience of using desferrioxamine as iron chelation therapy in children who are on haemodialysis. I understand that desferrioxamine does have another route of excretion from the body, other than in the urine and I wonder if you have looked into the kinetics of this.

MÜLLER-WIEFEL Yes, we have tried to look for the kinetics but it is very difficult to determine the desferrioxamine levels, so you have to determine the iron excreted in the dialysate or in the ultrafiltrate. We are performing our desferrioxamine treatment with a dose of 1gm per m² by infusion at the end of the dialysis, and with the next dialysis the iron is eliminated in a quantity of about — it depends on the iron stores, but if the serum ferritin levels are very high — 50mg/dialysis. But the problem is that it is very difficult to get a negative iron balance in children who already have iron overload and therefore we start desferrioxamine therapy at serum ferritin levels around 2000µg/L. Otherwise negative iron balance cannot be achieved.

FINE (Los Angeles) Have you had any correlation between the serum ferritin levels and deposition of iron in liver, intestine, etc? Second, if you follow the serum ferritin levels after successful transplantation, do the serum ferritin levels decrease or do they stay elevated for long periods of time and is there any difference as to whether the patients are HLA A3 and B7 as to what happens to ferritin levels after transplantation?

MÜLLER-WIEFEL There was correlation between the semi-quantitative measurement of iron in liver specimens and serum ferritin levels but unfortunately we did not measure it quantitatively but there is one more method to show a good correlation between iron stores and the serum ferritin. This is the measurement of the density of the liver by computer tomography and there is a good correlation too. And to the second question, indeed, we could demonstrate a decrease of serum ferritin levels after successful renal transplantation and after 1½ years the mean serum ferritin levels had normalised. So, renal transplantation is certainly the best method to avoid iron overload in chronic renal failure in children.

BROYER (Paris) Did you observe any case with clinical symptoms of haemochromatosis in these patients with iron overload?

MÜLLER-WIEFEL It is very difficult to differentiate between the symptoms of chronic renal failure sometimes and iron overload, but up to now we can say there was no patient in our hospital who died from massive iron overload and I do not remember the patients suffering from manifest clinical symptoms of iron overload. But we want to prevent this state.

De SANTO (Naples) Have you any experience with ferritin levels and intermittent PD or CAPD?
MÜLLER-WIEFEL We have no experience with ferritin levels in CAPD and IPD.

DRÜEKE (Paris) What is the mechanism for this increased iron reservoir? Would you speculate that there is increased intestinal absorption or would you speculate that there is increased iron retention in the reticulo-endothelial system?

MÜLLER-WIEFEL You are quite right, we have to speculate because this problem is not solved just as it is unsolved in patients with idiopathic haemochromatosis. Because we saw an increase of the serum ferritin levels only on haemodialysis, only in the form of treatment where blood transfusions have to be given and not on conservative treatment, I speculate that it is dependent on the reticulo-endothelial system and not on exaggerated absorption of iron by the intestine.