No regression of renal AL amyloid in monoclonal gammopathy after successful autologous blood stem cell transplantation and significant clinical improvement

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Abstract

Background. High-dose chemotherapy followed by autologous blood stem cell transplantation induces remission of plasma cell dyscrasia in patients with AL amyloidosis. The impact of this treatment on the glomerular amyloid mass is still unknown.

Methods. In the present study, the quantity of the renal amyloid mass before and more than 3 years after high-dose melphalan treatment and autologous blood stem cell transplantation was assessed in two patients. At the time of the second renal biopsy, both patients were in complete remission without detectable serum and urinary monoclonal IgA-C21 and a normal percentage of plasma cells in the bone marrow.

Results. In both patients with biopsy-proven AL amyloidosis, urinary protein excretion decreased from 7 g/24 h to <2 g/24 h more than 3 years after autologous blood stem cell transplantation. In contrast, glomerular amyloid deposits persisted, as shown in the second biopsy.

Conclusion. Despite complete remission of the plasma cell dyscrasia and improvement of glomerular permeability, the amount of glomerular amyloid mass did not regress.

Keywords: AL amyloidosis; autologous stem cell transplantation; nephrotic syndrome; quantification of amyloid

Introduction

AL amyloidosis is characterized by the accumulation of proteinaceous deposits in various tissues. The deposits are formed by monoclonal immunoglobulin light chains and light chain fragments. They also hold other constituents, including the amyloid P component. These insoluble proteins consist of fibrils with β-pleated configurations which bind Congo red and display the characteristic green birefringence [1]. Renal structures (e.g. glomeruli, pre-glomerular arterioles) are commonly affected by amyloid deposition in AL amyloidosis, with clinically evident renal disease occurring in 48–82% of patients [2–6]. AL amyloidosis is usually characterized by the nephrotic syndrome [7], and amyloid is located predominantly in the glomerular mesangium [8]. The natural course of renal involvement in AL amyloidosis is persistence of the nephrotic syndrome and a progressive decline in glomerular filtration rate (GFR) [9].

Previous reports on amyloid A (AA amyloidosis) documented the resorption of amyloid when the stimulus for amyloid formation was eliminated [10,11]. This was also reported for AL amyloidosis by van Buren et al. [12]. A decrease in amyloid deposits followed syngeneic bone marrow transplantation applying 125I-labelled serum amyloid P scintigraphy. However, there was no bioptic control. These observations are in contrast to a report by Kyle et al. [2], who found an increase of glomerular amyloid in a second renal biopsy despite the remission of nephrotic syndrome after treatment with oral melphalan and corticosteroids.

We report on two patients who received autologous blood stem cell transplantation for AL amyloidosis due to IgA-λ monoclonal gammopathy, resulting in subsequent remission of the nephrotic syndrome but
no regression of glomerular amyloid by quantitative measurement. Because of encouraging results [13], we have chosen the blood stem cell transplantation as a treatment option in our patients.

Case reports

Patient 1. A 50-year-old male patient presented with nephrotic syndrome (7 g/24 h) in August 1997. Renal biopsy revealed AL amyloid by Congo red stain and immunohistochemistry predominantly in the glomerular mesangium, and urinary excretion of free λ light chains and intact IgA-λ monoclonal proteins. In addition, cardiac AL amyloid was proven by biopsy. GFR was normal and he was treated from October to December 1997 with a combination chemotherapy (vincristine, adriamycin and corticosteroids). In June 1998, he received high-dose chemotherapy with ifosfamide (9.6 g/m²) followed by high-dose melphalan (200 mg/m²) and an autologous blood stem cell transplantation (July 1998). Urinary protein excretion declined to 1.6 g/24 h in February 2002 with neither free light chains nor IgA-λ excretion by immunfixation after isoelectric focusing.

Because of persistent proteinuria and the recent onset of microhaematuria, the patient underwent a second renal biopsy after informed consent.

Patient 2. A 52-year-old female patient presented with nephrotic syndrome (8 g/24 h) and renal failure (serum creatinine 2.0 mg/dl) in June 1997. Renal and cardiac biopsy revealed AL amyloidosis. She also had amyloidosis-associated nephropathy with recurrent syncopal episodes. Urinary excretion documented free λ light chains and IgA-λ monoclonal proteins. AL amyloid was predominantly found in the glomerular mesangium.

She was initially treated from July to September 1997 with a combination chemotherapy (vincristine, adriamycin and corticosteroids). This treatment was followed by high-dose ifosfamide (12 g/m²) chemotherapy which was stopped due to ifosfamide-induced encephalopathy. From January to March 1998, the M2 chemotherapy protocol was administered; this was followed by a high-dose melphalan (100 mg/m²) treatment in May 1998 and autologous blood stem cell transplantation. Urinary protein excretion decreased to 0.6 g/24 h in October 2001 with no excretion of free light chains and IgA-λ. Because of persistent proteinuria, a second renal biopsy was performed after informed consent.

Subjects and methods

The second renal biopsies in October 2001 were compared with the initial biopsies taken in 1997 by conventional histology and immunohistology. A panel of polyclonal antibodies against λ- and κ-light chains (Behring, Dako) as well as a purified amyloid fibril protein [1] was applied. The time between blood stem cell transplantation and a second renal biopsy was 39 months in patient 1 and 40 months in patient 2. Paraffin sections were studied following Congo red staining in bright and birefringent light. In addition, Congo red was used as a fluorochrome; this is more sensitive than Congo red light microscopy [1]. The amount of amyloid within the glomerulum was measured by a computer-aided device in a blinded fashion [1]. In patient 1, the biopsy of 1997 revealed five glomeruli (12 sections were examined) and the biopsy taken in 2001 included six glomeruli (seven sections examined). In patient 2, the biopsy of 1997 contained eight glomeruli (14 sections were available for quantification) and the biopsy taken in 2001 included seven glomeruli with 19 slides. The amount of amyloid was expressed as the amyloid-positive area of the glomerular tuft relative to the total glomerular area. Analysis was performed on individual glomerular measurements. Renal biopsies from three different patients with AL amyloidosis were taken as controls [14]. The number of biopsy sections were 121 (control 1), 19 (control 2) and 19 (control 3), respectively. Data are given as median and ranges and have been compared using the Wilcoxon test as appropriate.

Results

Results of patients 1 and 2

The amount of amyloid in patient 1 was 5.5% of the glomerular area prior to blood stem cell transplantation and 5.3% at the second biopsy. However, in patient 2, the amyloid mass increased from 17.7% at the time of the initial biopsy to 25%, despite clinical improvement with a reduction in proteinuria and normalization of GFR.

Table 1 summarizes the results of the quantitative measurements of amyloid mass in patients 1 and 2 before and after bone marrow transplantation.

Apart from AL amyloidosis, the second renal biopsy of patient 1 revealed mesangial deposits of IgA and C3 compatible with IgA glomerulonephropathy. IgA and C3 deposits were not present in the renal biopsy in 1997. This finding corresponded to the recently detected microhaematuria. Immunostainings with antibodies against amyloid A protein, β2 microglobulin and transthyretin were negative.

Results of controls

The amount of amyloid (%) in the three control patients was 15 (5.7–22.7), 8.8 (4–20.5) and 15.1 (8.7–31), respectively.

Table 1. The results of the quantitative measurements of amyloid mass in patients 1 and 2 before and after bone marrow transplantation

<table>
<thead>
<tr>
<th>Patient</th>
<th>% amyloid 1997</th>
<th>% amyloid 2001</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patient 1</td>
<td>5.5 (0.8–16.9)</td>
<td>5.3 (1.4–16)</td>
<td>NS</td>
</tr>
<tr>
<td>Patient 2</td>
<td>17.7 (14.1–24.4)</td>
<td>25 (9.5–34.8)</td>
<td>0.2</td>
</tr>
</tbody>
</table>
Discussion

We report on patients in whom AL amyloid did not regress despite high-dose chemotherapy with autologous blood stem cell transplantation and improvement of nephrotic syndrome. We chose the high-dose chemotherapy in combination with blood stem cell transplantation since recent studies suggest that this treatment option offers the best chance for haematological remission and prolongation of survival [15]. However, treatment-associated mortality of >10% should be taken into account and therapy should be limited to patients with good performance status and minor cardiac involvement [15]. Therapeutic success was documented by a significant fall in proteinuria and normalization of renal function (GFR) (patient 2). In addition, remission of plasma cell dyscrasia was repeatedly proven by the exclusion of serum and urinary IgA-λ and λ light chains. The absence of IgA-λ and λ light chains in serum and urine was accompanied by a remarkable improvement of the patients' well being. The presence of mesangial deposits of IgA and C3 in the second biopsy of patient 2 (IgA deposits were definitively not seen in biopsy 1, 3 years before) was unforeseen but correlated with the new appearance of microscopic haematuria in this patient. The association of IgA nephropathy and AL amyloidosis is exceptional. The finding may be a completely incidental association or related to the underlying lymphoproliferative disease and its complications [16]. Furthermore, it remains speculative whether deposition of IgA could influence or impair the glomerular clearance of amyloid deposits. While the clinical progress after treatment of AL amyloidosis is in line with several previous reports [2,6,12,16], the exact quantification of the amyloid mass 3 years after blood stem cell transplantation is new and needs further comment.

An earlier report by Kyle et al. [2] described an increase of amyloid deposition in two patients after oral treatment with prednisone and melphalan in a second renal biopsy or autopsy [2]. The amount of amyloid, however, was not assessed quantitatively. In our report, we assessed the amyloid mass more precisely by quantitative measurement using a computer-aided technique [1,14]. We confined our assessment of AL amyloid to the glomerulus because the glomerular area is a well-defined structure with a bordering membrane (Bowman’s capsule). Furthermore, there is a strong interaction of glomerular structures (e.g. mesangium, basement membrane) with AL light chains [8]. In our study, it is notable that amyloid mass remained virtually unchanged despite a significant fall in proteinuria and normalization of renal function (GFR) (patient 2).

The paradox of the objective improvement of the amyloid disease in the absence of biopsy-proven regression of amyloid mass needs further clarification. One explanation could be that the amyloidogenic light chain is the pathogen and not the amyloid mass per se. In both of our cases, the reduction of the λ light chains was associated with clinical improvement. Reduction of the light chain concentration does induce a shift in the balance from amyloid growth to regenerative processes in the glomerulus and does not possess the assumed nephropathogenic property as shown in our patients and in the report of Kyle et al. [2]. In addition, the shifted balance allows clinical improvement in spite of slow amyloidal apposition as in patient 2, since the pathogenic light chain is significantly reduced. The slight increase of amyloid mass could be explained by a minimal concentration of the light chain and/or the slow redistribution from the extracellular space. However, there are no specific tools (serum measurement, radioactive scan) to support this hypothesis. This hypothesis is supported by a recent report of Mandreoli et al. [17] who documented the disappearance of proteinuria in a patient with localized Castleman’s disease 30 months after surgical resection, despite persistence of glomerular amyloid mass. Electron microscopy revealed an even subtle reparative phenomenon at the epithelial site of the basement membrane [17].

A suitable hypothesis would be that the disappearance of amyloidogenic components (e.g. amyloid A, light chains) could restore cellular regeneration despite the local presence of amyloid mass. The pathogenetic property of amyloidogenic protein was described recently by Yan et al. [18] who found an interaction of amyloid A component and RAGE (receptor for advanced glycation end-products) which resulted in cellular perturbation. In a mouse model, amyloid A accumulation, cell stress and expression of RAGE were closely linked. Antagonizing RAGE suppressed cell stress and amyloid deposition in mouse spleen [17].

In summary, these direct quantitative results prove that a mechanism other than the amyloid mass per se may be responsible for the improvement of AL amyloid disease after blood stem cell transplantation. This mechanism is most probably the amyloidogenic light chain.

Conflict of interest statement. None declared.

References


*Received for publication: 10.4.03
Accepted in revised form: 9.7.03*