Serum amyloid A levels in human renal allograft rejection

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Summary
1. Serum amyloid A (SAA) levels were studied in 35 recipients of cadaveric renal transplants. Marked SAA elevations were seen during all acute allograft rejection episodes. The mean peak SAA level in well-documented rejections was 446 mg/l (median 415 mg/l, range 132-1040 mg/l; controls <1 mg/l).
2. Rejections in patients receiving cyclosporin-A alone as post-transplantation immunosuppressive medication were characterized by a significantly higher peak SAA level than rejections in patients receiving cyclosporin-A in combination with methylprednisolone (539 ± 53 mg/l, mean ± SEM, vs 226 ± 9 mg/l, P < 0.01).
3. Excluding surgery-induced SAA elevations in the immediate postoperative period, seven significant SAA peaks not related to allograft rejection were observed. These were associated with surgical complications and infections, and in one case probably with the underlying rheumatic disease, which was complicated by amyloidosis.
4. The results show that acute renal allograft rejection induces a dramatic acute phase SAA response. Since SAA is an easily measured serum component and the rejection-induced elevation is an early event, monitoring of SAA in kidney transplant patients may have considerable clinical significance.

Key words: amyloid A protein, cyclosporin-A, renal allograft rejection.

Abbreviation: SAA, serum amyloid A.

Introduction
Early recognition of allograft rejection remains a major problem in clinical renal transplantation. Monitoring of inflammatory and immune responses [1-6] in the postgrafting stage provides valuable information on the rejection process, may facilitate an early diagnosis of rejection and, in some instances, even predict rejection.

Recent studies have implicated serum amyloid A (SAA) in the early inflammatory response [7-9]. It is a very sensitive acute phase reactant; its serum concentration can rise rapidly by a factor of up to 1000 and it has a short half-life in serum [10-12]. In this paper we describe the effect of allograft rejection on SAA levels in patients undergoing kidney transplantation. The influence of two different post-transplantation immunosuppressive regimens on the acute phase SAA response was also assessed.

Patients and methods
Nine female and 26 male recipients (mean age 42 years, range 19-66 years) of cadaveric renal transplants were studied. There was no selection of patients for this study. The underlying renal disorders were chronic glomerulonephritis (16 cases), diabetic nephropathy (seven cases), unclassified chronic nephritis (four cases), polycystic renal disease (three cases), chronic pyelonephritis (two cases), focal glomerulosclerosis (one case), congenital renal hypoplasia (one case) and secondary amyloidosis (one case). The patients received immunosuppressive medication in the form of either cyclosporin-A alone (10 mg day⁻¹ kg⁻¹ [on the day of operation 20 mg day⁻¹ kg⁻¹]), tapered to a dose corresponding to a plasma level of 150-250 ng/ml within the first week, 19 patients) or
cyclosporin-A (as above) in combination with methylprednisolone (3.5 mg day\(^{-1}\) kg\(^{-1}\), reduced to zero over 9 days, 16 patients). Peroral methylprednisolone (3 mg day\(^{-1}\) kg\(^{-1}\)) was used for antirejection treatment.

Thirty-three antirejection therapy episodes were monitored by SAA determinations. Retrospective analysis revealed a classical acute allograft rejection episode in 27 instances (impairment in renal function tests, local and systemic signs and symptoms of rejection, positive finding in transplant aspiration cytology [2]. In six instances antirejection therapy had been started on the basis of a positive finding in transplant aspiration cytology without clinical signs of rejection ("subclinical rejection").

Three hundred and eighty post-transplant sera taken at 1–4 days intervals were analysed for SAA by using the radial immunodiffusion method. Specific antiserum and purified tissue amyloid A protein were used in the assays [13]. Controls were 50 healthy blood donors.

Results

The SAA level rose significantly during all rejection episodes; the mean peak level was 377 mg/l (median 276 mg/l, range 45–1040 mg/l, \(n = 33\); controls < 1 mg/l). In the well-documented rejections (i.e. clinical criteria + positive transplant aspiration biopsy, \(n = 27\)) the mean peak level was 446 mg/l (median 415 mg/l, range 132–1040 mg/l). In the ‘subclinical rejections’ (i.e. positive transplant aspiration biopsy without clinical signs of rejection, \(n = 6\)) the mean peak SAA level was 67 mg/l (median 66 mg/l, range 45–90 mg/l). The SAA response during two renal allograft rejection episodes is illustrated in Fig. 1. Both rejection episodes are accompanied by marked SAA elevations.

In the immediate postoperative period surgery-related SAA elevations with a peak on the second post-transplantation day were seen. Excluding these predictable changes in the SAA concentrations [14], serial measurements revealed seven significant SAA elevations not related to rejection, i.e. clear SAA peaks (in these instances \(> 90\) mg/l) rising above the patient’s post-transplantation baseline values. The probable causes of these elevations are summarized in Table 1.

Rejections in patients receiving post-transplantation immunosuppression by cyclosporin-A alone were characterized by significantly higher peak SAA levels than rejections in those receiving cyclosporin-A in combination with methylprednisolone (Table 2).

![Graph of serum creatinine and SAA levels over time](image)

**Fig. 1.** Serum amyloid A (SAA) concentration, inflammatory score in fine needle aspiration biopsy of the graft [2] (TAC), and serum creatinine concentration during two renal allograft rejection episodes. MP, Commencement of antirejection therapy with peroral methylprednisolone (3 mg day\(^{-1}\) kg\(^{-1}\)).

<table>
<thead>
<tr>
<th>Patient no.</th>
<th>Peak SAA (mg/l)</th>
<th>Probable cause</th>
</tr>
</thead>
<tbody>
<tr>
<td>21</td>
<td>90</td>
<td>Severe stomatitis</td>
</tr>
<tr>
<td>15</td>
<td>115</td>
<td>Urinary infection (Proteus vulgaris)</td>
</tr>
<tr>
<td>28</td>
<td>135</td>
<td>Amyloid disease associated with spondylarthitis</td>
</tr>
<tr>
<td>6</td>
<td>185</td>
<td>Perirenal haematoma</td>
</tr>
<tr>
<td>33</td>
<td>195</td>
<td>Necrosis of ureter with extravasation of urine</td>
</tr>
<tr>
<td>3</td>
<td>360</td>
<td>Febrile urinary infection (Escherichia coli)</td>
</tr>
<tr>
<td>8</td>
<td>415</td>
<td>Subcutaneous wound infection</td>
</tr>
</tbody>
</table>
TABLE 2. Comparison of peak serum amyloid A levels during rejection episodes in patients receiving cyclosporin-A alone or in combination with methylprednisolone as post-transplant immunosuppression

Both groups received the same antirejection treatment with methylprednisolone. Values are means ± SEM.

<table>
<thead>
<tr>
<th>Immunosuppression</th>
<th>Rejection episodes</th>
<th>Mean peak SAA (mg/l)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cyclosporin-A</td>
<td>19</td>
<td>539 ± 53*</td>
</tr>
<tr>
<td>Cyclosporin-A + methyl-</td>
<td>8</td>
<td>226 ± 9</td>
</tr>
<tr>
<td>prednisolone</td>
<td></td>
<td></td>
</tr>
</tbody>
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* P < 0.01; Student’s t-test.

Discussion

The results show that acute renal allograft rejection in man induces a dramatic acute phase SAA response. Peak SAA levels in the well-documented rejections were similar in magnitude to those seen in extensive tissue trauma [14], and were about five to ten times the mean levels encountered in rheumatoid arthritis or secondary amyloidosis [13, 15]. The mechanism leading to the high SAA levels in acute rejection is not known, but it is probably related to the host’s general response to tissue injury and inflammation. SAA is an acute phase reactant synthesized by the hepatocytes [16] and perhaps by other cells as well [17–19]. Morrow et al. [7] studied acute phase liver tissue in culture and showed that the concentration of m-RNA for SAA increases to values at least 500 times greater than normal and that SAA synthesis comprises about 2.5% of the total hepatic protein synthesis in the acute phase response. Experimental studies have provided evidence that mononuclear cells/macrophages release a factor which induces hepatic SAA synthesis [9, 16, 20]. Two stages are recognized in SAA induction: a latent period of 1–3 h, during which the SAA concentration remains at baseline values and in which the SAA inducer appears in the blood, and a period of SAA synthesis which lasts for approximately 24 h after induction. The high SAA response seen in renal allograft rejection may also be mediated by monocyte/macrophage-derived factor(s). It has been shown that the continued influx of monocytes and appearance of macrophages in the transplant correlate with the severity of rejection [2]. It is noteworthy in this context that SAA suppresses the antibody response of spleen cells in vitro, and a normal function of SAA may be an immunoregulatory one [21].

Serial determinations showed that the SAA elevation in rejection is an early event. Since the SAA test is not a renal function test [22] it can also be carried out in transplant recipients with initially nonfunctioning grafts. These characteristics suggest that SAA measurements may be of considerable value in the monitoring of renal transplant patients. This view is also supported by the demonstration that the serum levels of another liver-produced protein, C-reactive protein, seem to predict the onset and termination of renal allograft rejection [3].

However, with respect to rejection diagnosis, the SAA character of a sensitive acute phase reactant must be kept in mind. Surgical complications and infections influence SAA levels. Surgery itself also increases SAA levels [14], but since these elevations are predictable and occur in the immediate postoperative period, they do not limit the use of the SAA determinations to any great extent. The most useful information is derived from serial, daily SAA determinations. They allow determination of the patient’s post-transplantation baseline SAA level (which may vary individually), from which a significant elevation suggests rejection or some other inflammatory complication.

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References

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