PRETRANSPLANT ANTIBODIES AND RENAL ALLOGRAFT SURVIVAL

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Summary

Fc receptor blocking antibodies directed against autologous lymphocytes were present in pretransplant sera from only one of a group of 24 cadaver donor renal transplant recipients. Such antibodies were present in recipient sera against B lymphocytes from the donor in 11/24, against normal B lymphocytes in 12/24 and against leukaemic B lymphocytes in 15/24 cases. There was a significant correlation between EA inhibiting antibodies directed against donor (p<0.01), normal (p<0.05) and leukaemic B lymphocytes (p<0.001) and improved one year allograft survival. No autoantibodies were detected. Fc receptor blocking antibodies detected by the Erythrocyte Antibody Inhibition assay may thus have a beneficial effect on graft outcome but do not appear to be autoantibodies.

Introduction

The presence of antibodies in recipient sera is generally thought to have a deleterious effect on transplant survival. In the standard crossmatch test, for example, the presence of recipient anti-donor lymphocytotoxic antibodies correlates with almost inevitable graft rejection. In the mid 1970s, however, sera from certain recipients were shown to contain cytotoxins directed against their own, as well as donor lymphocytes [1,2], and in these cases transplantation was successfully carried out. Subsequent larger studies confirmed the harmless nature of these autoreactive antibodies [3,4] and further work [5] showed that allograft survival was slightly, although not significantly, better in those with a positive B lymphocyte crossmatch due to autoantibodies than in those with a negative crossmatch. These antibodies were shown to react with normal panel lymphocytes but not with leukaemic (CLL) lymphocytes [5] and to be most active at 5°C [6].

In a previous study we showed that the presence of Fc receptor blocking
antibodies to B lymphocytes in pretransplant serum (detected by the erythrocyte antibody inhibition assay) correlated with improved allograft survival [7]. In this study, therefore, we aimed to determine whether such antibodies were autoantibodies.

Materials and methods

Sera were obtained from 24 recipients of cadaver donor renal transplants within the 12 hours prior to transplantation. All patients had been transfused with at least four units of blood prior to transplantation. An aliquot of each serum was heat inactivated at 56°C for 45 minutes and a further aliquot was absorbed with pooled human platelets. The sera were ultracentrifuged at 100,000g for one hour prior to use.

The target cells used were B lymphocytes from a) the transplant recipient, b) the transplant donor, c) 12 normal panel members and d) 12 patients with chronic lymphatic leukaemia (CLL).

Lymphocytotoxic antibodies were detected by the standard long NIH lymphocytotoxicity assay at 5°C, 23°C and 37°C [8]. EA inhibition was performed by a modification of the rosette technique [7].

Statistical analysis was performed using Fisher's exact test for four-fold tables.

Results

No lymphocytotoxic antibodies were detected in any of the 24 untreated or treated sera when the recipient's own B lymphocytes were used as targets and the assays were carried out at 5°C, 23°C and 37°C. B lymphocytotoxic antibodies were however present in two cases against donor B lymphocytes, in

<table>
<thead>
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<th>Target cell</th>
<th>Success</th>
<th>Failure</th>
<th>Total</th>
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<tr>
<td>Recipient B lymphocytes</td>
<td>EAI positive</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>EAI negative</td>
<td>14</td>
<td>9</td>
</tr>
<tr>
<td>Donor B lymphocytes</td>
<td>EAI positive</td>
<td>10</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>EAI negative</td>
<td>4</td>
<td>9</td>
</tr>
<tr>
<td>Normal Panel B lymphocytes</td>
<td>EAI positive</td>
<td>10</td>
<td>2</td>
</tr>
<tr>
<td></td>
<td>EAI negative</td>
<td>4</td>
<td>8</td>
</tr>
<tr>
<td>CLL Panel B lymphocytes</td>
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nine cases against normal panel B lymphocytes and in 14 cases against CLL panel B lymphocytes. There was no statistically significant correlation between the presence of such lymphocytotoxic antibodies in pretransplant sera against any of the types of target cells used and allograft survival (Table I).

EA inhibiting antibodies directed against the recipient’s own lymphocytes were detectable prior to transplantation in sera from only one recipient; they were not removed by platelet absorption and the graft failed within three months of transplantation. EA inhibition was not present against donor lymphocytes in this case. In the group of 24 patients as a whole, however, EA inhibiting antibodies were detected in 11 cases against donor, in 12 cases against normal panel and in 15 cases against CLL B lymphocytes. The presence of EA inhibition against donor normal and leukaemic B lymphocytes correlated significantly with improved allograft survival (p<0.01, p<0.05, and p<0.001 respectively).

Discussion
This study shows that Fc receptor blocking antibodies in pretransplant recipient sera (detected by the EA inhibition assay) were found in only one case when autologous lymphocytes were used as target cells, and in that case the graft failed. The sera, however, did contain such antibodies directed against the donor in around 50 per cent of cases and against panel lymphocytes even more frequently, and the correlation with improved allograft survival previously shown [7] was maintained.

Anti-donor cytotoxic antibodies, as occur in a positive crossmatch test, are harmful to allograft survival whereas similar antibodies which are also cytotoxic to autologous lymphocytes, particularly B lymphocytes, appear harmless [3,5]. They occur in the IgM fraction of serum [5] and since they react with normal panel B lymphocytes but not with leukaemic B lymphocytes (both of which possess HLA-A, -B, -C, and DR antigens) they are thought not to be directed to the products of the MHC. The Fc receptor blocking antibodies, however, which we have demonstrated occur in the IgG fraction of serum as shown by DEAE column chromatography and can react both with normal panel and leukaemic lymphocytes. We have shown that, as well as being indicative of favourable graft outcome, they are generated by blood transfusions [9] and therefore may represent a possible mechanism for the blood transfusion effect in renal transplantation. Antibodies detected by EA rosette inhibition has also been demonstrated in the sera of primigravid women but not in the sera of women undergoing spontaneous abortion [10]. Family studies using sera from primigravidae suggested that these antibodies were directed against HLA-linked antigens. They therefore have very different properties from previously determined autoantibodies.

Fc receptor blocking antibodies detected by the EA inhibition assay

a) are rarely directed against autologous B lymphocytes
b) correlate with improved allograft survival when present in pretransplant serum
c) are generated by blood transfusion, and
d) may be HLA-linked.
References

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7. MacLeod AM, Mason RJ, Stewart KN et al. Transplantation 1982; 34: 273
9. MacLeod AM et al. Lancet 1982; ii: 468