Impact of cyclosporin on the incidence and prevalence of chronic rejection in renal transplants

I J Beckham FRCS
Senior Surgical Registrar

J S O’Rourke MCh FRCS
Senior Surgical Registrar

S R Stubington FRCS
Transplant Research Fellow

M Hinwood RGN
Clinical Research Sister

M C Bishop MD FRCS
Consultant Transplant Surgeon

K M Rigg MD FRCS
Consultant Transplant Surgeon

Nottingham City Hospital, Nottingham

Key words: Renal transplantation; Chronic rejection

Over a 14-year period, 435 patients underwent renal transplantation. Chronic rejection has occurred in 58 (13%) of all grafts and has accounted for 18% of all graft losses. After the first 6 months following transplantation, chronic rejection was the most common cause of graft failure, accounting for 40% of losses. The median time (interquartile range) from transplantation to graft failure was 3 years (2–5.5 years). Comparison of azathioprine versus cyclosporin treated patients showed no significant difference in the incidence of graft loss (Cox regression score 2.55, \(P = 0.11\)). Furthermore, there were significantly more grafts with deteriorating function owing to chronic rejection in cyclosporin-treated patients \((n = 16, 11\%\) of surviving grafts) than in azathioprine-treated patients \((n = 2, 3\%\) of surviving grafts). These data suggest that cyclosporin does not prevent the development of chronic rejection in renal transplants.

One-year graft and patient survival figures after renal transplantation are well documented. Unfortunately, long-term allograft survival results have not matched the improvement in 1-year survival. Although increased short-term survival contributes to improved long-term success, the annual rate of graft loss after the first year has changed very little. The Eurotransplant Collaborative Study analysed 13 000 renal transplants based on grafts that were still functioning at 1 year after transplantation \((1)\). Between 1981 and 1987, patients receiving cyclosporin had an expected half-life that was little different than for patients receiving azathioprine performed between 1971 and 1975 (11.6 compared with 9.7 years). Similar results have been found by the UKTSSA and UCLA registries \((2)\).

There have been very few studies of the incidence of biopsy-proven chronic rejection, and none comparing the incidence in azathioprine and cyclosporin treated patients. We have analysed the results of 435 transplants performed over a 14-year period in a single centre.

Patients and methods

An analysis of all renal transplants performed in Nottingham between January 1978 and January 1992 was undertaken. Data concerning risk factors, immunosuppressive regimens and outcome measure were collected from the renal unit and pathology computer databases, hospital records and from the UKTSSA.

Patients

There were 435 transplants performed in this period, of which 335 were first cadaveric transplants, 85 were second or subsequent transplants and 15 were transplants from living related donors. Details of patients according to immunosuppressive regimens are shown in Table I.
**Table I.** Comparison of patients within each immunosuppressive group

<table>
<thead>
<tr>
<th></th>
<th>Azathioprine</th>
<th>Cyclosporin</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total number</td>
<td>211</td>
<td>224</td>
</tr>
<tr>
<td>Male : female ratio</td>
<td>1.54</td>
<td>1.24</td>
</tr>
<tr>
<td>Number 1st cadaveric grafts</td>
<td>173</td>
<td>162</td>
</tr>
<tr>
<td>Number 2nd/3rd cadaveric grafts</td>
<td>33</td>
<td>52</td>
</tr>
<tr>
<td>Number LRD</td>
<td>5</td>
<td>10</td>
</tr>
<tr>
<td>Local/imported donors</td>
<td>122/73</td>
<td>125/79</td>
</tr>
<tr>
<td>Mean no. HLA-DR</td>
<td></td>
<td></td>
</tr>
<tr>
<td>mismatches</td>
<td>0.82</td>
<td>0.52</td>
</tr>
<tr>
<td>Mean number total mismatches</td>
<td>3.39</td>
<td>2.64</td>
</tr>
</tbody>
</table>

N.B.— Data on the donor source were unavailable in 21 patients (10 in the cyclosporin-treated group and 11 in the azathioprine-treated group). Data on HLA matching was complete in 95% of the cyclosporin group (DR data unavailable in nine, no HLA data available in four) and 82% of the azathioprine group (DR data unavailable in 26, no HLA data available in 12).

**Immunosuppression**

Two main immunosuppressive regimens were used:

1. 1978–1986: Azathioprine 2.5 mg/kg/d and prednisolone 20 mg/d, decreasing by 2.5 mg/month starting after the 3rd month after transplantation, to achieve a maintenance dose of 10 mg/d.
2. 1986–1992: Cyclosporin 5 mg/kg/d and prednisolone as in (1). Doses were adjusted to maintain a trough level of 100–200 ng/ml measured by radio-immunoassay (CYCLO-Trak SP, Incstar Ltd, Wokingham, Berks).

During this study period, 211 patients received azathioprine- and 224 cyclosporin-based immunosuppression. The cyclosporin group included 21 patients who received triple therapy (cyclosporin 3.5 mg/kg, azathioprine 2 mg/kg, and prednisolone as in (1) above). A preliminary analysis of this small subgroup (not shown) showed no differences from the main group of cyclosporin-treated patients.

Patients who changed immunosuppressive regimen within the first month because of a precipitous fall in white cell count with azathioprine (n = 3), were analysed according to the predominant immunosuppressive regimen, i.e. cyclosporin.

Acute rejection episodes were treated with intravenous methyl prednisolone 500 mg/d for 3 days. Steroid-resistant rejection and acute vascular rejection were treated with antilymphocyte globulin (ALG, Merieux, Maidenhead, Berks) for 10–14 days after 1983, or by ALG or OKT3 after 1986.

**Cause of graft loss**

Graft failure was classified according to clinical and histopathological evidence in the following groups:

1. Early technical failure—within the 1st week after transplantation, usually from venous or arterial thrombosis, with no evidence of acute rejection on histopathological examination.
2. Acute rejection—histologically proven.
3. Death with a functioning graft within the first 6 months.
4. Later death with functioning graft.
5. Chronic rejection—defined according to the definition first proposed by Foster et al. (3) and subsequently accepted by the Alexis Carrel conference on chronic rejection (4) (Table II).
6. Other/unknown.

**Functioning grafts**

All grafts with deteriorating renal function were identified from computer-generated graphs of plasma reciprocal creatinine versus time. Patients in whom the slope of the reciprocal of the plasma creatinine was significantly different from zero (over a period of more than 3 months observation, with at least 10 consecutive creatinine values) were biopsied. Only one patient with these criteria (a Jehovah’s Witness receiving warfarin for pulmonary emboli) was not biopsied. This allowed the number of patients with chronic rejection causing deteriorating graft function to be determined. The estimated time to graft failure was calculated by extrapolation of the graph of reciprocal plasma creatinine versus time (assuming a plasma creatinine concentration of 1000 µmol/l to be indicative of graft failure). This method has been shown to predict the time to end stage renal failure to within 4 months in 82% of cases (5).

**Analysis**

Patient follow-up was complete to January 1994. Actuarial graft survival was analysed by Kaplan–Meier life survival analysis and groups were compared by Cox regression analysis. Actual and predicted times to graft failure were compared by the Mann–Whitney U test.
Table III. Outcome of renal transplants at end of follow-up

<table>
<thead>
<tr>
<th></th>
<th>Azathioprine</th>
<th>Cyclosporin</th>
</tr>
</thead>
<tbody>
<tr>
<td>Early technical failure</td>
<td>18</td>
<td>29</td>
</tr>
<tr>
<td>Acute rejection</td>
<td>43</td>
<td>19</td>
</tr>
<tr>
<td>Death with functioning graft</td>
<td>16</td>
<td>3</td>
</tr>
<tr>
<td>Death with functioning graft (&gt; 6/12)</td>
<td>17</td>
<td>12</td>
</tr>
<tr>
<td>Chronic rejection</td>
<td>28</td>
<td>12</td>
</tr>
<tr>
<td>Other/unknown</td>
<td>16</td>
<td>8</td>
</tr>
<tr>
<td>Still functioning</td>
<td>73</td>
<td>141</td>
</tr>
</tbody>
</table>

Results

Analysis of graft losses at all times after transplant

Follow-up was complete to January 1994, and ranged from 7–15 years in the azathioprine-treated group compared with 2–8 years in cyclosporin-treated patients. Actuarial graft survival was better for cyclosporin- than azathioprine-treated patients (Cox regression score 8.14, P < 0.004). A total of 138 (65%) grafts had failed in the azathioprine-treated group, and 83 (37%) in the cyclosporin-treated group. Causes of graft loss, irrespective of length of follow-up, are shown in Table III. Chronic rejection accounted for 18% of all graft losses (20% in the azathioprine group, 15% in the cyclosporin group).

Analysis of graft loss after first 6 months after transplantation

An analysis of causes of graft loss after the first 6 months following transplantation (ie in those grafts at risk of loss from chronic rejection) is shown in Fig. 1. Chronic rejection accounted for 40% of all graft losses after the first 6 months (42% azathioprine, 36% cyclosporin). Graft losses listed as ‘Other’ were made up of the following mixed causes: recurrent renal disease or de novo glomerulonephritis (8), diabetic nephropathy (2), pyonephrosis or recurrent kidney infection (3), chronic cyclosporin toxicity (2), ureteric stenosis (1), renal artery stenosis (1), lymphoma (1), recurrent stone disease (1). The diagnosis of chronic cyclosporin toxicity was made when vascular changes were confined to arterioles according to the generally accepted criteria (6). Although cyclosporin results in slightly elevated mean creatinine levels, few allograft losses are attributable to chronic cyclosporin toxicity alone (7). The cause of graft loss was unknown in five cases (two azathioprine-treated, three cyclosporin-treated) owing to loss of follow-up or inadequate data.

The incidence of graft failure from chronic rejection over time is shown in the Kaplan–Meier survival table (Fig. 2). Comparison by Cox regression analysis shows no significant difference in the incidence of chronic rejection between immunosuppressive regimens. Total numbers of grafts lost, and failing from chronic rejection are shown in Fig. 3. Actual and predicted numbers of grafts lost from
chronic rejection on a yearly basis are shown in Fig. 4. The median time (interquartile range) to graft loss based on actual graft losses in azathioprine-treated patients is 3.5 years (2–7 years) and 3.0 years (2–4.5 years) in the cyclosporin group (Mann–Whitney U test, Z = 0.5, P = 0.6). The median (IQR) time to graft loss from transplantation based on actual and predicted graft loss for azathioprine-treated patients is 4 years (2–7.5 years) compared with 5 years (3–9 years) in cyclosporin-treated patients (Mann–Whitney U test, Z = 0.17, P = 0.17).

Discussion

Multicentre studies of long-term graft survival show a continued attrition of grafts after the first year of transplantation. Yet the incidence of chronic rejection as a cause of graft losses is still largely unknown. In the few studies where causes of later graft losses are mentioned, comparison of data is difficult because of the lack of a standard definition of chronic rejection, inadequate evidence of chronic rejection as the cause of graft failure or short follow-up. Knight et al. (8) found 69 cases of chronic rejection (11%) in 643 patients receiving cyclosporin, with a mean follow-up of 44 months. The diagnosis was made in patients >3 months after transplantation, with evidence of a progressive non-acute renal function deterioration, confirmed in most cases by biopsy. Graft failure had occurred in 38 patients by 3 years, although the proportion of all graft failures attributable to chronic rejection is not given. MacDonald et al. (9) reviewed the outcome of 276 renal transplants surviving beyond 6 months, but do not state their criteria for diagnosing chronic rejection. Chronic rejection accounted for 13% of all graft failures (ie 5% of all transplants performed) and 38% of grafts surviving beyond 6 months. Other studies often give information only about specific cohorts of patients. Rao et al. (10) reported that 86% of failures after 10 years were because of chronic rejection. Kirkman et al. (11) found 81% of failures after 5 years were owing to chronic rejection, although histology was only available in 36% of cases. The need for histological evidence is supported by a finding that 38% of biopsies yield a histological diagnosis contradicting the clinical pre-biopsy diagnosis of allograft dysfunction (12). We believe that the data presented in this study are the first from a single centre using standardised diagnostic criteria to compare the incidence of chronic rejection in azathioprine- and cyclosporin-treated patients; histological confirmation of the diagnosis of chronic rejection (either from biopsy or graft nephrectomy) was available in all but one patient in this series.

Graft losses in the first 6 months were predominantly from vascular thrombosis or acute rejection. Since the currently accepted definition of graft loss from chronic rejection requires grafts to be more than 6 months after transplantation, re-analysis of the data was performed to exclude graft losses occurring in the first 6 months (Fig. 1). Chronic rejection was the most common cause of graft failure after this time, accounting for 40% of graft losses, with a similar number of losses owing to patient death.

Graft losses, irrespective of cause, were more common in azathioprine-treated patients, reflecting in part the longer follow-up of this group. The proportion of graft losses after 6 months from chronic rejection are similar in each group (Fig. 1). Analysis of graft loss by Kaplan–Meier survival tables shows a trend towards azathioprine-treated patients losing more grafts from chronic rejection. However, any improvement in the cyclosporin group is small and is not statistically significant (Fig. 2). Furthermore, there are significantly more grafts with deteriorating function because of chronic rejection in cyclosporin-treated patients (n = 16, 11% of surviving grafts) than in azathioprine-treated patients (n = 2, 3% of surviving grafts) (Figs 3, 4).

Cyclosporin did delay the onset of chronic rejection which occurs at any time after the first 6 months with a
median time to graft loss (irrespective of immunosuppressive regimen) of 3.0 years.

The findings of this study are supported by a number of experimental models of chronic rejection, where cyclosporin in a variety of dosages and routes did not prevent the development of chronic rejection in heart (13,14), aorta (15,16) or kidney transplants (17–19). Low-dose cyclosporin, with or without prednisolone, is insufficient to prevent chronic rejection and the search continues for the optimal immunosuppressive agent.

References

7 Mihtsch MJ. Selectivity has its price: personal experience with cyclosporin over the last 10 years. Trans Proc 1988; 24 (Suppl 2): 67–70.

Received 20 June 1996