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Posttransplant Administration of Donor Leukocytes Induces Long-Term Acceptance of Kidney or Liver Transplants by an Activation-Associated Immune Mechanism

Yiqun Yan,* Suma Shastry,* Craig Richards,* Chuanmin Wang,* David G. Bowen,* Alexandra F. Sharland,* Dorothy M. Painter,* Geoffrey W. McCaughan,* and G. Alex Bishop²

Donor leukocytes play a dual role in rejection and acceptance of transplanted organs. They provide the major stimulus for rejection, and their removal from the transplanted organ prolongs its survival. Paradoxically, administration of donor leukocytes also prolongs allograft survival provided that they are administered 1 wk or more before transplantation. Here we show that administration of donor leukocytes immediately after transplantation induced long-term acceptance of completely MHC-mismatched rat kidney or liver transplants. The majority of long-term recipients of kidney transplants were tolerant of donor-strain skin grafts. Acceptance was associated with early activation of recipient T cells in the spleen, demonstrated by a rapid increase in IL-2 and IFN-γ at that site followed by an early diffuse infiltrate of activated T cells and apoptosis within the tolerant grafts. In contrast, IL-2 and IFN-γ mRNA were not increased in the spleens of rejecting animals, and the diffuse infiltrate of activated T cells appeared later but resulted in rapid graft destruction. These results define a mechanism of allograft acceptance induced by donor leukocytes that is associated with activation-induced cell death of recipient T cells. They demonstrate for the first time that posttransplant administration of donor leukocytes leads to organ allograft tolerance across a complete MHC class I plus class II barrier, a finding with direct clinical application. The Journal of Immunology, 2001, 166: 5258–5264.

Donor leukocytes, in particular donor dendritic cells within a transplanted organ, are the major stimulus for its rejection, and depletion of these passenger leukocytes leads to long-term graft acceptance in animal models (1, 2). In contrast, donor leukocytes can promote survival of transplanted organs, and infusion of donor leukocytes before transplantation often leads to prolonged survival of organs that would otherwise have undergone rapid rejection (3, 4). Previous studies in experimental models have shown that donor leukocytes only prolong allograft survival when administered at least 1 wk before transplantation (4–6). These findings have a limited application in the clinic, as cadaver donors are the major source of organs for transplantation and acceptance of the transplanted organ requires that MHC Ags are shared between the leukocyte donor and the organ donor (reviewed in Ref. 7). Furthermore, pretransplant infusion of donor blood is associated with a risk of presensitization of the recipient, resulting in hyperacute rejection rather than acceptance (7).

Despite these initial studies showing the lack of efficacy of donor leukocytes when infused at the time of transplantation, some evidence from animal models indicates that they can promote tolerance when present at the time of transplantation. Acceptance of liver allografts across complete histocompatibility barriers without immunosuppression is dependent on passenger leukocytes (8, 9). Also, spleen allografts in some completely MHC-mismatched strain combinations are accepted without requiring immunosuppression (10). In a model of rat heart transplantation, posttransplant administration of donor leukocytes slightly prolonged survival although it did not lead to long-term acceptance (11). Furthermore, transplantation of a heart together with organs rich in donor leukocytes such as lung or spleen (12), or of multiple organs supplemented with donor leukocytes (13), were found to promote long-term allograft acceptance. Posttransplant administration of donor leukocytes also led to tolerance of skin grafts across a minor histocompatibility barrier (14).

The aim of the studies presented here was to investigate whether administration of donor leukocytes at completion of the transplant operation could induce long-term acceptance of rat kidney or liver allografts across a complete major MHC barrier. Kidneys from PVG strain donors transplanted to completely MHC-mismatched DA recipients (PVG→DA) are rapidly rejected (13) although liver transplants in the same strain combination are spontaneously accepted without requiring immunosuppression (15). In contrast, high-responder Lewis strain recipients reject livers from PVG strain donors (15, 16). The effect of administration of donor leukocytes on rejection of PVG→DA kidney allografts and of PVG→Lewis liver allografts was examined. Previous investigations of the immune mechanism of spontaneous acceptance of liver allografts have shown activation and apoptosis of T cells (16, 17). Consequently, these processes were examined in the leukocyte-induced transplant acceptance reported here.

Materials and Methods

Animals

Inbred strains of rats were obtained from the Animal Resources Center (Perth, Western Australia) and were completely mismatched at the rat MHC (RT1) locus. The rat strains used were PVG (RT1a), DA (RT1b), and Lewis (RT1c). All experiments were performed with the approval of the Royal Prince Alfred Hospital Animal Care Ethics Committee.
Transplantation

Orthotopic liver or kidney grafts were performed, and in the case of kidney grafts, the contralateral kidney was removed 3 days after transplantation. Techniques for liver, kidney, and skin grafting have been reported previously (13). For some experiments, donors were given 10 Gy of whole body irradiation 1 wk before liver transplantation as described (9).

Immunohistochemical staining

Localization of leukocyte populations was by immunostaining of frozen sections by an indirect immunoperoxidase technique (18). Primary Abs were obtained from Dr. J. Sedgwick (DNAX Research Institute, Palo Alto, CA). They were OX6, reactive with MHC class II; OX27, reactive with donor (PVG) MHC class I; OX39, reactive with CD25; R73, reactive with α/β TCR; OX35 plus W3/25 1:1 mixture, reactive with CD4; OX42, reactive with CD11b/c expressed by monocytes/macrophages and dendritic cells (19); and MOPC 21 (Sigma, St. Louis, MO), negative control. Apoptotic cells were identified in frozen sections by TUNEL staining as described previously (20) with reagents obtained from Boehringer Mannheim (Mannheim, Germany).

Flow cytometry

Tissues were dissociated by mashing the organs through a 100-mesh steel sieve. Leukocytes were purified by centrifugation of the washed cell suspension on isoinct Percoll (Pharmacia Biotech, Uppsala, Sweden) 1.05 g/ml for 15 min at 840 × g. The leucocyte-containing pellet was washed and analyzed by four-color flow cytometry as described (17). Abs to CD4 or PVG MHC class I were detected by anti-mouse Ab conjugated to fluorescein (F2266; Sigma). R73-PE and OX39-biotin were obtained from Serotec (Oxford, U.K.); streptavidin-allophycocyanin was obtained from Molecular Probes (Eugene, OR). Dead cells were identified with propidium iodide at 1 μg/ml and excluded from analysis.

Measurement of cytokine mRNA expression

Cytokine mRNA in spleen was measured by quantitative RT-PCR as we have described previously (17). Total RNA was isolated and reverse-transcribed, and then aliquots of cDNA were measured for cytokine mRNA expression in a quantitative PCR. Quantification was by a noncompetitive method with external standards of known numbers of cytokine cDNA molecules. In some experiments, the number of cytokine mRNA molecules was adjusted to account for sample-to-sample variation with a G3PDH-positive internal control. In some experiments, quantitative PCR was performed by using a Model 7700 Sequence Detector (PE Applied Biosystems). Amplification primers were used at 300 nM each. The probe sequences were 6FAM-TTG CCC AAG CAG GCC ACA GAA TTG-CA 3′ and 6FAM-CCT TTT GCC AGT GTT AC 3′ (forward) and 6FAM-TTG CCC AAG CAG GCC ACA GAA TTG-CA 3′ and TAMRA, and the amplification primers were CCC CAT GAT GCT CAC AT 3′ and 6FAM-CCT TTT GCC AGT GTT AC 3′ (reverse). For IFN-γ, the probe sequence was 6FAM-CTG CCC AAG CAG GCC ACA GAA TTG-CA 3′ and 6FAM-TTG CCC AAG CAG GCC ACA GAA TTG-CA 3′ (forward) and ATT TTC CAG GCA CTG AAG ATG TTT (reverse). For IPN-γ, the probe sequence was 6FAM-CCT TTT GCC AGT TTC 3′ and 6FAM-TTG CCC AAG CAG GCC ACA GAA TTG-CA 3′ (forward) and ATT TTC CAG GCA CTG AAG ATG TTT (reverse). The leukocyte-containing pellet was washed and analyzed by four-color flow cytometry as described (17). Abs to CD4 or PVG MHC class I were detected by anti-mouse Ab conjugated to fluorescein (F2266; Sigma). R73-PE and OX39-biotin were obtained from Serotec (Oxford, U.K.); streptavidin-allophycocyanin was obtained from Molecular Probes (Eugene, OR). Dead cells were identified with propidium iodide at 1 μg/ml and excluded from analysis.

Preparation of spleen leukocytes and leucocyte subsets

Spleen leukocytes were aseptically prepared by meshing the spleen and then lysing the RBC in NH₄Cl buffer as described (17). Cells were washed and injected at a dose of either 2 × 10⁷ or 6 × 10⁷ i.v. in 1 ml of isotonic saline. This cell population consisted of a mixture of T cells, B cells, myeloid cells, and small numbers of NK cells, and RBC and platelets were not detectable. In some experiments, spleen cells were fixed by incubation in 2% paraformaldehyde (BDH, Poole, U.K.) in PBS for 20 min on ice, washed thoroughly with PBS containing FBS, and injected i.v. For separation of leucocyte subsets from recipient spleen, aliquots of 1.5 × 10⁷ spleen leucocytes were incubated with either OX27, R73, OX35 plus W3/25, or MOPC 21. After incubation for 30 min on ice and washing in PBS, the cells were incubated with 6 × 10⁷ magnetic beads (Dynal, Oslo, Norway), washed, and separated on a magnetic column as described previously (13) before RNA extraction.

Statistical analyses

Comparison of survival data was by log-rank analysis of the product-limit estimate of Kaplan and Meier as described previously (9). Cytokine mRNA molecule numbers were analyzed by Mann-Whitney U test, and infiltration and apoptosis in tissue sections were analyzed by unpaired t test. Error bars for cytokine mRNA expression and infiltration and apoptosis show the mean ± SD of three separate animals.

Results

Posttransplant administration of donor leukocytes induces allograft acceptance

Rat kidney allografts in the strain combination of PVG donor to DA recipient (PVG→DA) mismatched at all MHC class I and II loci were rejected with a median survival time of 7 days (Table I). Treatment of the recipients with donor leukocytes at completion of transplantation led to long-term acceptance of the transplanted kidneys. All recipients of 6 × 10⁷ spleen leukocytes survived for >300 days, and treatment with 2 × 10⁷ leukocytes resulted in long-term acceptance of the majority of kidneys (Table I). Long-term surviving recipients of PVG kidney allografts plus PVG leukocytes were tested 250 days after transplant with a PVG donor skin graft. In three of four animals so tested, the skin graft survived for >100 days, showing that donor leukocyte treatment led to tolerance. Donor leukocytes failed to prolong renal allograft survival when their viability was impaired by fixation (Table I).

Unmodified liver transplants were accepted in the PVG→DA strain combination without a requirement for conventional immunosuppression or donor leukocyte treatment (Table I). Irradiation of the liver donors abrogated tolerance, and administration of donor leukocytes at the time of transplantation of these irradiated livers resulted in restoration of graft survival (Table I), as we and others have shown previously (9, 13, 21). Liver allografts from PVG donors were rejected in the high responder Lewis rat strain.

Table I. Donor leukocytes induce long-term acceptance of kidney and liver transplants

<table>
<thead>
<tr>
<th>Organ Transplanted</th>
<th>Strain Combination*</th>
<th>Treatment†</th>
<th>Survival (days)</th>
<th>MST (days)</th>
<th>p (vs untreated)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Kidney</td>
<td>PVG→DA</td>
<td>6,6,6,7,9,10,13</td>
<td>168, 253, &gt;268, &gt;272</td>
<td>261</td>
<td>0.005</td>
</tr>
<tr>
<td>Kidney</td>
<td>PVG→DA</td>
<td>2 × 10⁷</td>
<td>&gt;300 × 4</td>
<td>&gt;300</td>
<td>0.005</td>
</tr>
<tr>
<td>Kidney</td>
<td>PVG→DA</td>
<td>6 × 10⁷</td>
<td>&gt;300 × 4</td>
<td>&gt;300</td>
<td>0.005</td>
</tr>
<tr>
<td>Kidney</td>
<td>PVG→DA</td>
<td>6 × 10⁷</td>
<td>&gt;100 × 6</td>
<td>&gt;100</td>
<td>0.99</td>
</tr>
<tr>
<td>Liver</td>
<td>PVG→DA</td>
<td>12,12,12,14,15,16,56, &gt;100 × 2</td>
<td>35,97, &gt;100 × 7</td>
<td>&gt;100</td>
<td>0.68</td>
</tr>
<tr>
<td>Liver</td>
<td>PVG→DA</td>
<td>14,14,15,17</td>
<td>&gt;100 × 5</td>
<td>&gt;100</td>
<td>0.004</td>
</tr>
</tbody>
</table>

* Strains used were PVG (RT1a), DA (RT1a), and Lewis (RT1b) and were mismatched at all MHC class I and class II loci.
† Treatment involved injection of cells into the recipient or irradiation of the donor or a combination of both.
‡ Pathology analysis showed hydronephrosis, probably due to ureteric obstruction.
although rejection could be prevented by administration of donor leukocytes to the recipient at the time of transplantation (Table I). Histological examination of the long-term accepted grafts showed little or no evidence of chronic rejection but did reveal occasional small perivascular aggregates of leukocytes that had not damaged the graft or blood vessels within it. The above results showed that large numbers of PVG donor leukocytes administered at the time of transplantation converted rejection of both kidney and liver allografts to long-term acceptance.

**Rapid migration of donor leukocytes to lymphoid tissues**

PVG leukocytes administered i.v. to DA recipients of PVG renal allografts at the time of transplantation migrated rapidly to recipient lymphoid tissues. Flow cytometric analysis showed that donor cells comprised 4.7 ± 0.5% (n = 3) of recipient spleen leukocytes 1 day after renal transplantation supplemented with 6 × 10⁷ leukocytes. This declined rapidly to 1.6 ± 0.6% on day 3. Immunohistochemical staining demonstrated the presence of large numbers of donor cells (849 ± 97 cells/mm²) in the periarteriolar lymphoid sheaths of recipient spleens on day 1. Many also migrated to the paraaortic lymph nodes draining the graft (288 ± 132 cells/mm²). By contrast, very few donor cells had migrated to the spleen and lymph nodes of recipients of kidney allografts transplanted without additional donor leukocytes, with only 15 ± 8 cells/mm² and 10 ± 6 cells/mm², respectively, being detected on day 1.

**Tolerance-associated immune activation**

The levels of IL-2 and IFN-γ mRNA expression were compared in the spleen of recipients of allografts that were undergoing either rejection or tolerance. Marked activation, as measured by these parameters, was observed early after transplantation in the spleens from recipients of tolerant but not rejecting grafts (Fig. 1). There was a significant (p = 0.05) increase of IL-2 mRNA 1 day after transplantation in the tolerant leukocyte-treated kidney or liver recipients compared with untreated kidney recipients undergoing rejection or normal nontransplanted animals (Fig. 1A). The increase in IL-2 mRNA in kidney-tolerant animals was rapid and transient, characterized by a peak on day 1 that had disappeared by day 3, similar to the transient increase in IL-2 mRNA observed in recipient lymphoid tissues of animals during liver allograft tolerance (16). No detectable increase in IL-2 expression was observed in the spleen of untreated kidney allograft recipients undergoing rejection or in syngeneic controls. IFN-γ mRNA showed a similar pattern of increase in tolerant kidney or liver allograft recipients that significantly exceeded that observed in untreated recipients or syngeneic controls (p = 0.05; Fig. 1B).

**Cellular source of the IL-2 and IFN-γ mRNA in tolerant allograft recipients**

The spleens of recipients of PVG→DA liver allografts were separated into subpopulations of leukocytes on day 1 after transplantation. The expression of IL-2 mRNA in these subpopulations is shown in Fig. 2A. Recipient cells expressed significantly (p = 0.05) more IL-2 than did donor cells. The activation-associated increase in IL-2 mRNA, compared with cells from a normal spleen, was confined to recipient cells from tolerant animals. Donor cells, analyzed by flow cytometry, comprised 1.4 ± 0.1% (n = 3) of the spleen leukocytes, and produced 3.0 ± 1.3% of the amount of IL-2 mRNA produced by recipient cells. T cells, which comprised 37.4 ± 2.7% of the spleen population, produced 97.6 ± 35% of the IL-2, whereas non-T cells only yielded 2.4 ± 0.9% of
pressed 71.6
6
liver-tolerant animals were followed by an extensive, diffuse in-

The early cytokine increases found in the spleens of kidney or

recipient compared with donor cells (p < 0.05), with only

4.4 ± 3% of the total IFN-γ being produced by donor cells. In

contrast to IL-2, IFN-γ was expressed by both T cells, which ex-

pressed 71.6 ± 41.5% of the total, and non-T cells, which ex-

pressed 28.4 ± 16.3% of the total. CD4 and non-CD4 cells pro-

duced approximately equivalent amounts of IFN-γ mRNA. Thus,

in quantitative terms, the rapid increase in cytokine mRNA in

spleens of tolerant animals was largely attributable to activation of

recipient CD4 T cells in the case of IL-2 and of recipient T cells

and non-T cells in the case of IFN-γ.

T cell activation and macrophage infiltration in tolerant kidney

allografts

The early cytokine increases found in the spleens of kidney or liver-tolerant animals were followed by an extensive, diffuse in-

filtrate of T cells and IL-2R-expressing (IL-2R+) cells in the tol-

erant kidney grafts with a high ratio of IL-2R+ cells to T cells on
day 3 after transplantation (Fig. 3; Table II). However, by day 5 after transplantation comparable numbers of T cells were present in the interstitial areas of both rejecting and tolerant kidney grafts. At the same time, the proportion of IL-2R+ cells in these areas had decreased in tolerant vs rejecting grafts (Fig. 3). Thus, the kinetics of the activated T cell response in tolerance and rejection differed markedly, with infiltration of large numbers of T cells and activated cells being seen mainly in the interstitial areas of tolerant grafts at least 2 days before their appearance in rejecting grafts. By contrast, the infiltrate in rejecting grafts was limited mainly to perivascular sites until day 5 after transplantation, when a marked increase in interstitial infiltrate occurred (Table II). Thus, it appears that tolerance is associated with altered patterns of T cell infiltration into the graft.

Graft-infiltrating leukocytes were stained for markers of T cells, IL-2R, and donor MHC class I to confirm that the infiltrate in day 3 tolerant kidneys was composed of recipient activated T cells and not injected donor cells that had localized to the transplanted kidney. The majority of T cells in the infiltrate was of recipient origin (Fig. 4A), donor cells comprising only 4.8 ± 2.6% of the T cell infiltrate. Many of the T cells in the infiltrate were activated as shown by the observation that 32 ± 5% of T cells expressed IL-2R α-chain. On further analysis of T cells, the majority (61.7 ± 0.3%) proved to be CD4+, a considerable proportion of which were IL-2R+ (43.7 ± 4.0%) (Fig. 4B).

The monocyte/macrophage and dendritic cell infiltrate was identified with the OX42 Ab reactive with CD11b/c expressed on these cells. Their pattern of infiltration was similar to T cells in that on day 3 there were significantly more (p = 0.004) monocytes/macrophages in the interstitial areas of tolerant than rejecting kidneys (Table II). By day 5 there were similar numbers of these cells in the interstitial areas of both tolerant and rejecting kidneys.

Early apoptosis in the lymphoid tissues and graft is associated

with tolerance

To examine whether activation of T cells in tolerant animals led to their subsequent death by apoptosis, sections of kidney allografts and of recipient spleen were stained by TUNEL to identify apoptotic cells. TUNEL staining of tolerant kidney transplants on day 3 identified large numbers of apoptotic cells in the same location as the interstitial infiltrate of T cells and IL-2R+ cells (Fig. 5A). In contrast, there were few detectable apoptotic cells in the rejecting kidneys at the same time (Fig. 5B). Examination of the spleen of kidney allograft recipients showed a corresponding large increase in apoptotic cells in the peritubular sheaths of tolerant (Fig. 5C) compared with rejecting animals on day 3 (Fig. 5D).

Table II. Comparison of graft infiltrates during kidney allograft acceptance and rejection

<table>
<thead>
<tr>
<th>T Cells</th>
<th>IL-2R+ Cells</th>
<th>CD11b+ Cells</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Perivascular</td>
<td>Interstitial</td>
</tr>
<tr>
<td>Normal PVG kidney</td>
<td>14 ± 6</td>
<td>6 ± 4</td>
</tr>
<tr>
<td>Day 3 KTx</td>
<td>865 ± 243**</td>
<td>71 ± 23**</td>
</tr>
<tr>
<td>Day 3 KTx + cells</td>
<td>2476 ± 758</td>
<td>604 ± 153</td>
</tr>
<tr>
<td>Day 5 KTx</td>
<td>1735 ± 269</td>
<td>546 ± 115</td>
</tr>
</tbody>
</table>

* Monocytes/macrophages and dendritic cells were identified by expression of CD11b/c.

** Number of cells/mm² of leukocyte subsets was identified by immunohistochemical staining of kidney tissue (mean ± SD of three replicate animals).

* PVG to DA kidney transplants (KTx) that reject were compared on day 3 and day 5 after transplantation to PVG to DA kidney transplants injected with 6 × 10⁷ PVG spleen leukocytes (KTx + cells) that are tolerant.

Statistically significant difference between KTx and KTx + cells by unpaired t test (*, p < 0.05; **, p < 0.005; ***, p < 0.001).
Counting of TUNEL$^+$ cells showed that there were significantly more of these cells in the spleen ($p = 0.0003$) and transplanted kidney (0.006) of tolerant compared with rejecting animals on day 3 (Fig. 6). By day 5 this difference had disappeared and there were similar numbers of apoptotic cells in both tolerance and rejection (Fig. 6). It is possible that the increase in TUNEL$^+$ cells in the rejecting kidney on day 5 might have been attributable to apoptosis of kidney parenchymal cells, reflecting damage to these cells as a result of rejection, which was usually complete by day 7. These results examining apoptosis in the transplanted kidney and recipient spleen indicate that early cell death was associated with tolerance rather than rejection.

**Discussion**

According to published evidence, donor leukocytes most effectively prevent rejection when given at least 1 wk before transplantation (5, 6). Such treatment is of little practical value in clinical transplantation programs, which depend on cadaver donors as the major source of organs. There is also the risk of patient presensitization by donor leukocyte pretreatment. The results presented here provide the first demonstration of markedly prolonged survival of either liver or kidney allografts when the recipients were given donor leukocytes at the time of transplantation, despite the use of completely MHC-mismatched strain combinations normally associated with rapid rejection. Recipients of kidney allografts treated in this way ultimately developed tolerance to subsequent skin grafts of the same strain as the kidney donor. Presumably, the explanation for the discrepancy between our studies and those of previous investigators is related to the choice of strain combination or organ transplanted. Previous studies have examined heart or skin allografts in mouse or rat models that are likely to present a greater barrier to induction of tolerance than kidney or liver allografts (5, 6, 13). Nevertheless, the transplant models examined here do reflect the strength of the barriers encountered in human transplantation, especially considering that human renal transplants, where possible, are matched at the MHC.

Our results show that donor leukocytes rapidly migrated to recipient lymphoid tissues, which are the site of initiation of the immune response to transplanted organs (22, 23). Within 24 h of their migration to the recipient spleen, rapid immune activation occurred, accompanied in the case of tolerance, but not rejection, with increased expression of IL-2 and IFN-$\gamma$ mRNA. These results for renal allograft tolerance induced by administration of donor leukocytes at the time of transplantation closely parallel previous findings in spontaneous tolerance of liver allografts that show a rapid increase in splenic IL-2 and IFN-$\gamma$ mRNA. This increase was not observed during rejection of liver (16), kidney (17), or skin (24) allografts.

Subsequent to immune activation in the recipient lymphoid tissues, an extensive diffuse infiltrate of activated recipient T cells, monocytes and macrophages appeared in the tolerant kidney grafts. This diffuse infiltrate differed markedly from the localized perivascular infiltrate that was initially observed in the untreated rejecting kidneys. At the same time, programmed cell death was
FIGURE 6. Time course of apoptosis in periarteriolar lymphoid sheaths of recipient spleens (A) or in transplanted kidneys (B) of PVG→DA kidney transplants. Apoptotic cells were identified by TUNEL staining of sections of untreated recipients (rejection) or of those that were induced to accept the kidney grafts by treatment with \(6 \times 10^7\) donor spleen cells (tolerance). Significantly more apoptotic cells were observed in tolerance compared with rejection on day 3 (*, \(p = 0.006\); **, \(p = 0.0003\); \(n = 3\) per group).}

observed in the tolerant kidneys and in the splenic periarteriolar lymphoid sheaths of the recipients of these kidneys, but not in those undergoing rejection. Thus, there was a close concordance between kidney allograft tolerance induced by donor leukocytes and spontaneous acceptance of liver allografts, which show an early infiltrate of activated T cells accompanied by activation-induced cell death (AICD) within the tolerant graft (17, 25) and in the recipient lymphoid tissues (17).

Analysis of the cell subsets producing IL-2 and IFN-\(\gamma\) in the spleen and of the activated cells in the graft showed that recipient and not donor cells were activated during induction of tolerance, suggesting exhaustion of the rejection response. The immune activation and subsequent exhaustive differentiation of recipient T cells culminating in AICD described here for leukocyte-induced transplant tolerance might also be important in other models of allograft tolerance. Once such model involves treatment of cardiac allograft recipients by blockade of the costimulatory interactions and not donor cells were activated during induction of tolerance, suggesting exhaustion of the rejection response. The immune activation and apoptosis is associated with this form of tolerance as well. Therefore, treatment of transplant patients might be improved by administration of donor leukocytes combined with immunosuppression that complements rather than inhibits tolerance associated with immune activation.

Acknowledgments

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References


Abbreviation used in this paper: AICD, activation-induced cell death.
tolerance and rejection are more marked in the B cell compared with the T cell or cytokine response. Transplantation 57:1349.


