Abstract

Objectives: Several donor and recipient factors are known to be associated with graft loss in a kidney transplant. In this retrospective single-center study, we analyzed the effect of clinical and immunologic factors on kidney transplant outcomes in our region in Italy.

Materials and Methods: The study included 245 transplanted recipients from deceased donors at Federico II University of Naples, Kidney Transplant Centre, between the years 2000 and 2006. Age, cause of death, history of hypertension, hypotension or cardiac arrest, length of time spent in the intensive care unit, serum creatinine levels and human leukocyte antigen typing all were evaluated in the donors. Age, time spent on the wait list, human leukocyte antigen typing, antibody sensitization, and allocation were evaluated in the recipients. Age, donor/recipient matching, and human leukocyte antigen mismatches also were evaluated.

Results: Cox regression analysis showed that in recipients, time spent on the wait list increased the risk of restarting dialysis (OR 1.019, 95% CI: 1.000-1.038; P = .050) and dying (OR 1.017, 95% CI: 1.000-1.038; P = .032). Patients who received a kidney from a donor with a history of hypertension presented a major risk of death (OR 3.212, 95% CI: 1.190-8.668; P = .021), while human leukocyte antigen-A mismatch increased the risk of restarting dialysis (OR 3.137, 95% CI: 1.255-7.842; P = .014).

Conclusions: In our study, in recipients, time spent on the wait list, and a history of hypertension were associated with a greater risk of death. Human leukocyte antigen-A mismatch is associated with a greater risk of restarting dialysis.

Key words: Human leukocyte antigen mismatch, Kidney transplant, Restarting dialysis

Introduction

A kidney transplant is still the best treatment for patients with end-stage renal disease. It results in better outcomes than dialysis regarding quality of life and risk of long-term mortality.1 Despite immunosuppressive and supportive therapies that have significantly reduced early graft loss, improving outcomes in the short and medium terms, long-term dysfunction still is associated with a return to dialysis and increased mortality.2,3 Long-term graft function is dependent on several donor/recipient clinical characteristics and immunologic factors.4,5 Indeed, it is well-known that the donor’s age, cause of brain death, prolonged cold ischemia time, hypertension, diabetes mellitus, or other conditions impair donor renal function and affect graft survival.6-9 Also, recipient factors such as
age, time spent on the wait list, cause of end-stage renal disease, and duration of dialysis treatment can influence outcomes.\textsuperscript{10-13}

Although it is well-established that a stringent human leukocyte antigen (HLA) compatibility improves graft survival,\textsuperscript{14} and deleterious effects of preformed anti-HLA antibodies are well known,\textsuperscript{15-18} it remains unknown whether HLA compatibility is mandatory in kidney transplant.\textsuperscript{19-21} Indeed, to limit HLA mismatching, organ sharing is frequently done, amplifying logistic difficulties and enhancing cold ischemia time.\textsuperscript{19,20} Recently, it was demonstrated that prolonged ischemia increases the risk of graft failure in the first year posttransplant.\textsuperscript{22} In contrast, other studies show that short cold ischemia time does not eliminate the benefit of better HLA matching.\textsuperscript{23} A recent multicenter study\textsuperscript{24} demonstrated that better HLA matching in renal transplant was associated with a lower dosage of immunosuppressive drugs; and therefore, a lower incidence of adverse effects from these drugs. Additionally, considering that HLA mismatch is the primary cause of allosensitization, good compatibility reduces acute and chronic detrimental effects by preventing anti-HLA antibodies development after transplant.\textsuperscript{25,26} Moreover, limited allosensitization may denote a great benefit to increase the possibility of further retransplant.\textsuperscript{27} We retrospectively analyzed the effect of the main clinical and immunologic factors on kidney transplant outcomes such as mortality or restarting dialysis treatment in a transplant center in a southern region of Italy.

Materials and Methods

From 2000 to 2006, at the “Federico II” University of Naples Transplant Renal Centre, we performed 308 single kidney transplants from deceased donors in 306 adult patients; 2 patients were retransplants.

Among these (306) patients, 245 of them were followed regularly at the time of analysis (June-December 2011) and were enrolled in this study. The study protocols were approved by the ethics committee of the institution before the study began, and the protocols conformed with the ethical guidelines of the 1975 Helsinki Declaration. Written, informed consent was obtained from all patients.

We recorded donor and recipient clinical and immunologic characteristics stored in a computer database and used for potential organ-donor reports and kidney allocation (Gedon-LURTO, La Traccia, Matera, Italy). Donor characteristics included age; sex; cause of death; history of hypertension and diabetes mellitus; cardiac arrest; or prolonged, low blood flow during an intensive care unit stay; and serum creatinine levels in the hospital and before organ donation. Donor data were repeated if both kidneys of a single donor had been transplanted. Recipient characteristics evaluated were age, sex, time spent on wait list, HLA typing, and antibody sensitization.

Recipient selection was performed according to allocation rules that include donor-compatible ABO blood group, absence of HLA antibodies against donor-HLA antigens, preventive negative cross-match, and number of HLA mismatches not greater than 4. All candidates who responded to these criteria were scheduled, according to a points-based scoring system that prioritizes a long waiting time, HLA match (number and types of HLA mismatch), and age match.

Patient and donor HLA typing were performed by serologic complement-dependent cytotoxicity assays and by molecular techniques: polymerase chain reaction with sequence-specific primers.\textsuperscript{28} Human leukocyte antigen antibody detection was performed periodically by complement-dependent cytotoxicity and by solid-phase assays such enzyme-linked immunosorbent assay and HLA-microbead assay (Luminex, Austin, TX, USA).\textsuperscript{29,30} The entity of allosensitization of each patient was expressed as percentage of panel reactive antibodies. Preventive complement-dependent cytotoxicity cross-match was performed for all transplanted recipients by standard method, with a long incubation, using historical and current recipient sera and isolated class I and class II donor lymphocytes.\textsuperscript{31}

To minimize adverse events related to over immuno-suppression, patients were treated with double immunosuppressive therapy (methylprednisolone and cyclosporine or tacrolimus) or triple (plus mycophenolate mofetil) at variable dosages, adjusted for each patient, relative to drug blood levels (except corticosterone) and time since the transplant.\textsuperscript{32}

Statistical analyses

Continuous variables are expressed as means ± SD. Categorical data are expressed as frequencies and percentages. During follow-up, characteristic...
differences in patients who had died, who had restarted dialysis, and free subjects were evaluated with univariate analysis or the chi-square test. Cox regression analysis was used on restarting dialysis and death, and combined endpoints were performed. In addition, HLA-A, HLA-B, and HLA-DR mismatches on different dialysis-free survival rates were analyzed by Cox survival curves. Statistical analyses were performed with SPSS software (SPSS: An IBM Company, version 17.0, IBM Corporation, Armonk, NY, USA). A value of $P \leq .05$ was considered statistically significant.

**Results**

Our sample consisted of 245 renal transplanted patients, age $44.1 \pm 10.7$ years (range, 18-68 y), mostly male (173; 70.6%), with time on the wait list of 25.8 months, followed for 84 ± 33 months.

As shown in Table 1, donor age was $40.4 \pm 15.5$ years, while mean difference in age between donors and recipients was $10.9 \pm 8.8$ years. Main cause of donor death was a cerebrovascular accident, and in particular, 55.5% for cerebral hemorrhage and 8.6% for cerebral ischemia. There were no cerebrovascular accidents, including traumatic brain injury and postanoxic encephalopathy (33.5% and 2.4%). Donor hospitalization in the intensive care unit was $6.1 \pm 5.7$ days, and reversible cardiac arrest occurred in 8.2% cases. Donor serum creatinine levels at the hospital and before donation were $79.6 \pm 26.5$ and $88.4 \pm 53.0$ μmol/L (0.9 ± 0.3 and 1.0 ± 0.6 mg/dL), a history of hypertension was present in 22.9% of the patients. Regarding immunologic compatibility, total HLA mismatch was 2.97 ± 1.1; specifically HLA-A mismatch was 1.12 ± 0.7, HLA-B mismatch was 1.25 ± 0.6, and HLA-DR mismatch was 0.60 ± 0.6.

### Table 1. Baseline Data From Kidney Recipients and Donors

<table>
<thead>
<tr>
<th>Variables</th>
<th>Recipients</th>
<th>Donors</th>
<th>Allocation</th>
</tr>
</thead>
<tbody>
<tr>
<td>n</td>
<td>18</td>
<td>31</td>
<td>49</td>
</tr>
<tr>
<td>Age (y)</td>
<td>44.6 ± 10.6</td>
<td>47.3 ± 14.1</td>
<td>44.4 ± 14.1</td>
</tr>
<tr>
<td>Time in the intensive care unit (d)</td>
<td>.553</td>
<td>.015</td>
<td>.023</td>
</tr>
<tr>
<td>Death for cerebrovascular accident (%)</td>
<td>.823</td>
<td>.774</td>
<td>.774</td>
</tr>
<tr>
<td>Hypertension (%)</td>
<td>.034</td>
<td>.023</td>
<td>.003</td>
</tr>
<tr>
<td>Hypotension or cardiac arrest (%)</td>
<td>.516</td>
<td>.774</td>
<td>.516</td>
</tr>
<tr>
<td>First creatinine level, μmol/L (mg/dL)</td>
<td>79.6 ± 26.5 (0.9 ± 0.3)</td>
<td>78.4 ± 26.5 (0.9 ± 0.2)</td>
<td>86.6 ± 26.5 (0.9 ± 0.3)</td>
</tr>
<tr>
<td>Last creatinine level, μmol/L (mg/dL)</td>
<td>88.4 ± 26.5 (1.0 ± 0.5)</td>
<td>79.0 ± 26.5 (1.0 ± 0.5)</td>
<td>86.6 ± 26.5 (1.0 ± 0.5)</td>
</tr>
<tr>
<td>Age donor/recipient matching (%)</td>
<td>9.4 ± 7.4</td>
<td>1.5 ± 0.6</td>
<td>1.2 ± 0.5</td>
</tr>
<tr>
<td>HLA-A mismatch (n)</td>
<td>44.0</td>
<td>1.5</td>
<td>1.2</td>
</tr>
<tr>
<td>HLA-B mismatch (n)</td>
<td>10.3 ± 7.9</td>
<td>1.0 ± 0.7</td>
<td>1.2 ± 0.6</td>
</tr>
<tr>
<td>HLA-DR mismatch (n)</td>
<td>645</td>
<td>264</td>
<td>595</td>
</tr>
<tr>
<td>Total HLA mismatch (n)</td>
<td>10 ± 9.1</td>
<td>1.2 ± 0.7</td>
<td>1.2 ± 0.6</td>
</tr>
</tbody>
</table>

Abbreviations: HLA, human leukocyte antigen

All $P$ values are tested vs no events.
1.25 ± 0.6, and HLA-DR mismatch was 0.60 ± 0.6 (Table 1). Of 245 transplanted subjects, only 20 (8%) presented anti-HLA antibodies before transplant, and only 12 had panel reactive antibodies greater than 30%.

Data were analyzed to evaluate the differences between transplanted patients who restarted dialysis, died, or combining these 2 events; and subjects who did not have these events (Table 2). At the end of the follow-up, 18 patients (7.3%) restarted dialysis, and 31 patients died with working graft (12.7%) after 47 ± 30 and 29 ± 28 months after the transplant. Patients who restarted dialysis had a longer time on the wait list, received kidneys from older donors, died predominantly of a cerebrovascular accident, had a history of hypertension, and presented more frequently with an HLA-A mismatch than did patients without events. Causes of death were cardiovascular diseases in 15 patients (48, 4%), infections in 10 patients (32, 3%, three were in triple immunosuppressive therapy), and cancer in 6 patients (19, 3%, one patient was in triple therapy). Patients who died during follow-up were generally older, had a longer time on the wait list, and received an organ from older donors with prevalence of hypertension.

If we consider combined endpoints, we find that donor age, time spent on the wait list, and history of hypertension were significantly different versus no-events group. It is noteworthy that in the combined endpoint group, recipient age was significantly higher and the cause of donor death was predominantly a cerebrovascular accident. Moreover, in these patients, we noted that HLA-A mismatch was not significantly higher compared with the no-event group’s.

Cox proportional regression model demonstrated that the more time spent on the wait list, the higher was the risk of restarting dialysis (OR 1.019, 95% CI: 1.000-1.038; P = .050) or dying (OR 1.017, 95% CI: 1.001-1.033; P = .032). There was a higher risk of death present for patients who received a kidney from a donor with a history of hypertension (OR 3.212, 95% CI: 1.190-8.668; P = .021). Interestingly, HLA-A mismatch increased the risk of restarting dialysis (OR 3.137, 95% CI: 1.255-7.842; P = .014) (Table 3). Moreover, dialysis-free survival curves during follow-up according to the number of HLA-A, B, and DR mismatches are shown in Figure 1. Human leukocyte
antigen-A, but not HLA-B and HLA-DR significantly predicted restarting dialysis.

Discussion

In the present, retrospective single-center study, time spent on the wait list increases the risk of restarting dialysis and death in recipients after transplant. Moreover, despite the fact that kidneys from hypertensive donors were not evaluated before transplant by a kidney biopsy, a history of hypertension in donors is associated with a major risk of death. In addition, we found that HLA-A mismatch is increase the risk of restarting dialysis, independent of the number of HLA mismatches.

Several studies have shown that renal graft outcomes are affected by such factors as donor and recipient age, cause of brain death, time spent on a wait list, ischemia time, impaired graft function, cause of end-stage renal diseases, immunologic compatibility, and recipient presensitization. Our study results confirm that a longer time on the wait list and a history donor hypertension are 2 significant risk factors affecting patients and graft outcomes.

In contrast to the literature, our data show that donor and recipient age do not affect the risk of recipient death and graft loss. Furthermore, we did not observe any effect of other important donor-related factors, such as time spent in the intensive care unit, severe hypotension, or reversible cardiac arrest and renal function. These findings probably are due to the young ages of our donors and recipients (about 44 and 47 y), and to our transplant center restricting criteria for donor and recipient eligibility for transplant during 2000 to 2006. Similarly, this study did not show the benefit of receiving the kidney of deceased donors who did not die of a cerebrovascular accident. We can hypothesize that this is because of the high percentage of noncerebrovascular death in our population (which is relatively young); however, it is remarkable that the cerebrovascular/noncerebrovascular ratio is 1.8 in all of our followed-up patients, and increases to 3.5 in persons who died, to 8.0 in persons who had to restart dialysis, while it was 1.5 in patients who had none of these events. These data confirm the favorable trend for patients who received kidneys from a deceased donor who died of traumatic or anoxic injuries.

Feeding the debate on role of an HLA compatibility-based allocation policy, especially in a kidney transplant from a living donor (in which excellent long-term survival rates have been observed independently from HLA incompatibility), is the fact that recent studies have confirmed the HLA-matching benefit on kidney transplant outcomes. Indeed, HLA matching is associated with a working graft with a lower incidence of death, posttransplant lymphoma, osteoporosis, and having a hip fracture.

In our single-center study, we found that HLA-A mismatches only increase the risk of restarting dialysis 3-fold, but they do not affect the risk of death. The lack of a statistically significant greater risk of death and combined endpoints may suggest an immunologic pathogenetic mechanism of short-middle damage that induces loss of graft function, and consequently, restarting dialysis. Although, we do not have the data on the incidence of donor-specific antibodies that might support this hypothesis, it should be considered a low percentage of preallosensitization recipients in our cohort.

Thus, in our study population, the time spent on the waiting list, combined with a history of hypertension, are associated with a higher risk of death in transplanted recipients who have received their grafts from deceased donors. Interestingly, HLA-A mismatches, also, are associated with a higher risk of restarting dialysis.

References