

Prolonged Cold Ischemia Time Obviates the Benefits of 0 HLA Mismatches in Renal Transplantation

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Hypothesis: Recipients of 0 HLA mismatch kidneys with prolonged cold ischemia times of longer than 36 hours do not have superior outcomes compared with recipients of kidneys with 1 or more mismatches.

Design: Retrospective review.

Setting: Transplantation centers.

Patients and Methods: A total of 63688 recipients who underwent transplantation between January 1, 1990, and July 31, 1998.

Main Outcome Measures: Delayed graft function, serum creatinine level, and patient and renal graft survival.

Results: Recipients of 0 HLA mismatch kidneys with fewer than 36 hours of cold ischemia time had better 5-year graft survival (75%) when compared with recipients with 1 or more mismatches (67%) ($P < .001$). However, recipients of 0 HLA mismatch kidneys with longer than 36 hours of cold ischemia time did not have any graft survival advantage (71% in 0 HLA mismatch kidneys vs 72% in 1 or more mismatches, $P = .24$).

Conclusions: Cold ischemia times of longer than 36 hours obviate the benefits of better graft survival conferred by better matching.

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AGGRESSIVE PROMOTION of organ donation has resulted in a steady increase in donors but, unfortunately, the disparity between the number of organs available and the number of patients awaiting transplantation continues to increase.¹ The waiting time for organs doubled from an average of 400 days in 1988 to 842 days in 1994 and regional differences in waiting time vary significantly.¹ Because regional discrepancies in waiting time are due to the sharing of kidneys on a regional basis, national sharing to ameliorate the perceived inequity of disparate waiting times has been proposed. Only “perfectly matched” kidneys are shared nationally because of the superior outcome attained with better matching. However, national sharing of kidneys tends to result in prolonged transport and cold ischemia times (CITs) that have been shown to compromise renal function. To determine the effect of both prolonged CIT and degree of HLA match on renal transplantation outcome, the United Network of Organ Sharing/Scientific Registry patient data were reviewed.

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RESULTS

Between January 1, 1990, and July 31, 1998, 63688 recipients of cadaveric renal transplants were reported to the United Network of Organ Sharing/Scientific Registry with sufficient CIT and HLA matching data. The distribution of patients with varying HLA mismatches is shown in **Figure 1**. The average (\pm SD) number of mismatches was 3.7 ± 1.5 and only 4% of patients had OMM. Average \pm SD CIT was 21.6 ± 9.8 hours with 62% of the kidneys being preserved for 0 to 24 hours, 30% for 24 to 36 hours, and 8% for longer than 36 hours. Increased transportation time owing to sharing nationally likely resulted in OMM kidneys having somewhat longer CITs compared with those with 1 or more mismatches (1 + MM) ($P < .001$) (**Figure 2**).

The effect of increasing CIT was first evaluated. Serum creatinine levels 4 years after transplantation was statistically, but not clinically, different in the 0- to 24-hour and the 24- to 36-hour groups (mean \pm SD, 156.8 ± 81 mmol/L [1.77 ± 0.92 mg/dL] for both) vs the longer than 36-

PATIENTS AND METHODS

Between January 1, 1990, and July 31, 1998, a total of 71 269 recipients of cadaveric renal transplantations were reported to the United Network of Organ Sharing/Scientific Registry. Of these, 63 688 had sufficient CIT and HLA matching data for retrospective comparison. To assess the effect of CIT on outcome measures, comparisons were made of patients with 0 to 24, 24 to 36, and longer than 36 hours of CIT. Donor and recipient HLA antigen profiles were analyzed to determine degree of match. "Perfect HLA matching" occurs when all 6 HLA loci are matched, but because it is possible to have undefined loci, a more accurate term is "0 mismatches" (OMMs). Six mismatches represents the worst donor-recipient matching situation. Delayed graft function (DGF) was defined as the patient's requiring dialysis in the early posttransplantation period. Graft loss was defined as the patient's permanent return to dialysis or graft loss owing to patient death. Death-censored graft survival excludes patients who died with a functioning graft.

Statistical calculations were performed using a commercially available software program (Statview 5; Abacus Concepts, Berkeley, Calif). Statistical significance was defined as $P < .05$. Differences were evaluated using either the unpaired t test or the χ^2 test, as appropriate. The probability of the difference between actuarial survival curves was determined by the Breslow (generalized Wilcoxon) method and the Mantel-Cox method. The Breslow method allows more weight to early differences that could relate to preservation duration and the Mantel-Cox method allows more weight to later differences. Multivariate analyses were performed using the Cox proportional hazards model. Covariates were considered to have an association with outcome when $P < .05$ and the relative risk ratio was greater than 1.20 or less than 0.85. Factors evaluated in the Cox proportional hazards model included data that may be important to the outcome.

hour group (161 ± 87 mmol/L [1.82 ± 0.98 mg/dL]) ($P = .02$). Shorter CITs were associated with improved graft survival. Five-year graft survival in the 0- to 24-, 24- to 36-, and longer than 36-hour groups was 68.3%, 63.1%, and 59.9%, respectively ($P < .001$ for all comparisons) (**Figure 3**). Five-year patient survival was slightly improved in the 0- to 2-hour group (85.4%) vs the 24- to 36-hour (82.2%) and the longer than 36-hour groups (81.3%) ($P < .008$ for the 0- to 24-hour group vs the 24- to 36-hour and >36 -hour groups; $P = .12$ for the 24- to 36-hour group vs the >36 -hour group).

Evaluation of the effect of mismatches showed that fewer mismatches portended improved outcome. Mean (\pm SD) 4-year posttransplant serum creatinine levels in patients with OMM was 144 ± 71 mmol/L (1.63 ± 0.84 mg/dL) vs 153 ± 82 mmol/L (1.78 ± 0.93 mg/dL) in patients with 1+MM ($P < .001$). Allograft survival was better in the OMM group compared with the 1+MM group. Five-year graft survival was 71.3% in patients with OMM and 63.9%

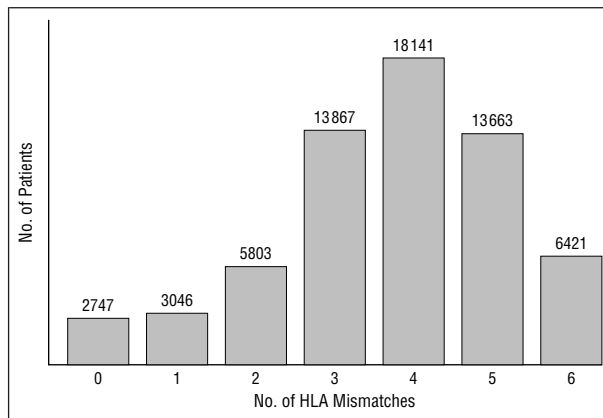


Figure 1. Distribution of HLA mismatches in 63 688 cadaveric renal transplantations reported to the United Network of Organ Sharing/Scientific Registry, 1990 to 1999.

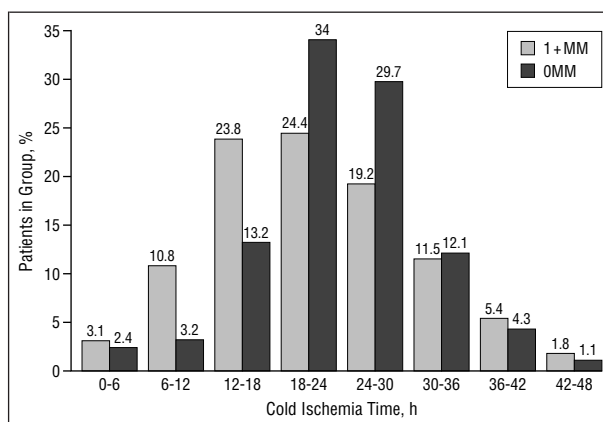


Figure 2. Recipients of 0 mismatched (OMM) kidneys were more likely to have longer cold ischemia times of 18 to 24 and 24 to 30 hours, while recipients of kidneys with more than 1 mismatch (1+MM) were more likely to have shorter cold ischemia times of 6 to 12 and 12 to 18 hours.

in patients with 1+MM ($P < .001$) (Figure 3). Patient survival was similar between the 2 groups up to 5 years after transplantation.

Relative risks showing the combined effect of CIT and HLA matching on renal graft survival are listed in the **Table**. The index group, with a relative risk of 1.0, consisted of patients who had 3 to 4MM and 0 to 24 hours of CIT (3-4MM/0-24 h) because 3.7 was the average number of mismatches and 22 hours was the average CIT. Increased CIT and more mismatches resulted in a greater risk for poorer graft survival (relative risk >1.0). Improved HLA matching can compensate for increased CITs and conversely, shortened CITs can compensate for poorer matching.

Evaluation of increasing CITs specifically in recipients with OMM was performed to examine the possible effect of nation sharing on outcome. Recipients of OMM kidneys with 0 to 24, and 24 to 36 hours of CIT had a 5-year graft survival advantage compared with all other subgroups ($P < .001$) (**Figure 4**). Recipients of OMM kidneys with longer than 36 hours of CIT (OMM/36+ h), however, did not have a survival advantage compared with patients with 1+MM with CITs of fewer than 24 hours ($P = .24$) (Figure 4). Patients with OMM/36+ h fared bet-

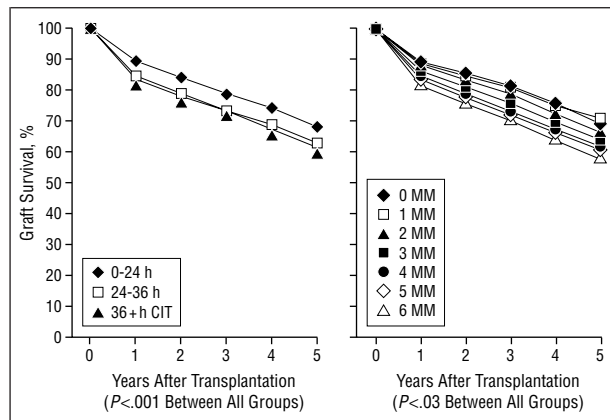


Figure 3. Actuarial graft survival decreases with progressively longer preservation times (left) and decreases with progressively more HLA mismatches (MMs) between the donor and the recipient (right). CIT indicates cold ischemia time.

Relative Risks for Renal Graft Survival Conferred by Degree of HLA Matching and Duration of Cold Ischemia Time*

No. of HLA Mismatches	Cold Ischemia Time, h†		
	0-24	24-36	36+
0	0.78 (0.69-0.89)†	0.78 (0.68-0.89)†	0.96 (0.72-1.28)
1-2	0.81 (0.75-0.87)†	0.93 (0.85-1.01)	1.10 (0.94-1.27)
3-4	1.0‡	1.16 (1.10-1.21)†	1.30 (1.21-1.39)†
5-6	1.17 (1.12-1.22)†	1.28 (1.21-1.36)†	1.37 (1.26-1.49)†

*There is increased risk for poorer graft survival with both longer cold ischemia times and with more HLA mismatches.

† $P < .001$ vs the 3 to 4 MM/0 to 24-hour group. Relative risk (95% confidence interval in parentheses) are determined using the Cox proportional hazards survival analysis model.

‡The index group is the patient group with 3 or 4 mismatches and kidney preservation times of 0 to 24 hours; the group is assigned a relative risk of 1.0.

ter than patients with 1+MM with longer than 24 hours of CIT ($P < .001$).

The effect of both CIT and HLA matching on short-term function was studied. Delayed graft function was found in 530 (19%) of the patients with 0MM and 14 687 (24%) of the patients with 1+MM ($P < .001$). Prolonged CITs were associated with increased incidence of DGF (15% at 0-24 hours, 29% at 24-36 hours, and 25% at >36 hours, $P < .001$ for all comparisons). The combined effect of both CIT and HLA matching is shown in **Figure 5**. Cold ischemia time appears to have a greater effect on DGF than does the number of HLA mismatches because, despite better matching, recipients of 0MM with longer than 24 hours of CIT had more DGF compared with recipients with less well-matched kidneys and shorter CITs. It seems that better HLA matching has a greater effect on graft survival, while prolonged CIT has a greater effect on DGF.

Multivariate analysis of other possible factors associated with decreased renal graft survival was performed. The index group, with a relative risk of 1.0, again consisted of patients who had 3 to 4MM and 0 to 24 hours of CIT. Risk for decreased graft survival was increased in donors younger than 18 years (1.55 times greater risk)

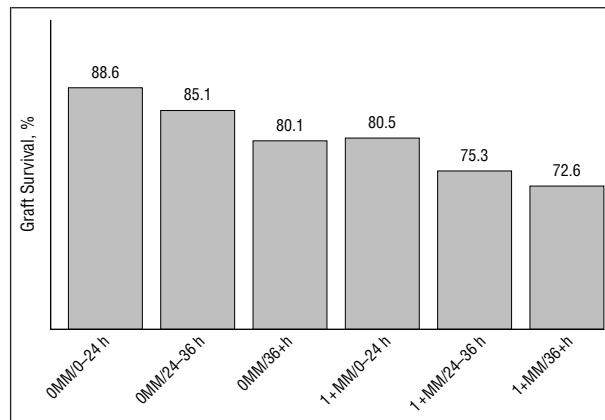


Figure 4. Comparison of actuarial graft survival of subgroups of patients with 0 mismatches (0MM) with varying cold ischemia times demonstrates that 0MM kidneys with preservation times of fewer than 36 hours (0MM/0 to 24 hours and 0MM/24 to 36 hours) confer a graft survival advantage compared with all other subgroups. Zero mismatched kidneys with cold ischemia times of longer than 36 hours (0MM/36+ hours), however, do not have improved graft survival compared with kidneys with more than 1 mismatch preserved for fewer than 24 hours (1+MM/0 to 24 hours). $P < .001$ for all comparisons except 0MM/36+ hours vs 1+MM/0 to 24 hours.

or older than 55 years (1.87 times risk), and recipients older than 55 years (1.3 times risk) ($P < .001$ for all groups vs index group). Donor hypertension did not increase risk.

COMMENT

The current policy of national sharing of 0MM kidneys is based on the belief that superior outcome can be achieved with better matching. However, national sharing tends to increase organ transportation time and thus CITs. Opinions vary widely in the literature regarding the effects of HLA matching and CIT. Some authors believe that HLA matching has a significant effect on graft outcome^{2,3} while others believe that it does not.⁴ Minimizing CIT is deemed so important that it has been suggested that HLA matching be omitted to reduce CIT,^{5,6} while others studies have concluded that the CIT is unimportant.⁷ The consensus is that fewer mismatches have been associated with improved graft and patient survival,^{8,9} and extended preservation has been associated with more DGF and decreased patient and graft survival.¹⁰

To evaluate the effect of the policy of national sharing of 0MM kidneys, the combined effect of both HLA matching and CIT needs to be determined. This study showed that the improved outcome as a result of better matching can be offset by prolonged CITs. If CIT was disregarded, patients with 0MM kidneys had less DGF and increased graft survival when compared with patients with 1 or more mismatches. When 0MM kidneys had more than 36 hours of CIT, however, the graft survival advantage was lost.

In the last few years concerns over the issue of equity in organ allocation have been raised. The fact that different procurement regions have widely disparate waiting times owing to regional distribution has been perceived by the general public as unfair. The possibility of moving to sharing all kidneys nationally, not just 0MM kidneys, has been proposed, but the issues of equity and utility need to be balanced.

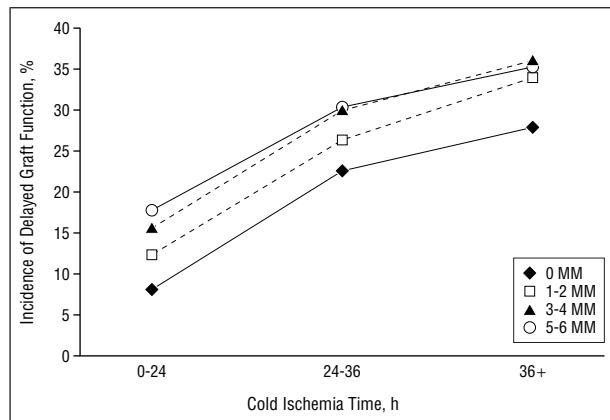


Figure 5. The incidence of delayed graft function increases as cold ischemia time increases and as the number of HLA mismatches (MMs) increases. Zero mismatched kidneys with cold ischemia times of longer than 36 hours have a higher incidence of delayed graft function compared with less well-matched kidneys with preservation times of less than 24 hours. $P < .05$ for all comparisons except between 3 to 4MM/24 to 36 hours and 5 to 6MM/24 to 36 hours, 3 to 4MM/36+ hours, and 5 to 6MM/36+ hours.

National sharing is beneficial not only in terms of equalizing disparate regional waiting times, but also because of (1) superior outcome if CIT is less than 36 hours, (2) shortened waiting times and improved outcome in highly sensitized patients, and (3) improved outcome in minority patients who on average are less well matched with the largely white donor population.

But while it may be more equitable to share organs on a national basis, it is unclear if the cost-benefit analysis will favor national sharing of all OMM kidneys, let alone all kidneys regardless of HLA match (as proequity groups are proposing).¹¹ The policy of national sharing of not only all kidneys but also OMM kidneys should be more closely examined because (1) as this article demonstrates, there are certain situations where national sharing of certain OMM kidneys does not result in improved outcome, (2) national sharing of kidneys has associated administration and transportation costs in an increasingly cost-conscious environment, and (3) the complexity of the situation increases further when other variables that may also affect outcome such as advanced donor age are considered. Advanced donor age has not only been associated with a negative effect on outcome in univariate analyses, but may also have an additive deleterious effect in conjunction with CIT.¹² While the ability to cold preserve organs for transplantation has improved dramatically over the last 3 decades, the current preservation techniques are, unfortunately, still a limiting factor and, as such, will continue to be a factor in the debate over national vs regional organ sharing.

CONCLUSIONS

Improved HLA matching improves outcome after cadaveric renal transplantation while prolonged CIT results in an increased incidence of DGF and decreased graft survival. Recipients of kidneys with 0MMs in general do better, but when CIT exceeds 36 hours the benefits conferred by improved HLA matching is obviated by the detrimental effects of prolonged CIT. Zero mis-

matches should not justify the acceptance of an otherwise inferior graft. Further collection and analysis of data on the combined effects of HLA matching and CIT is crucial in ascertaining the repercussions of proposed policies such as the national allocation of all organs for transplantation.

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DISCUSSION

Donald C. Dafoe, MD, Palo Alto, Calif: As the success and safety of renal transplantation has steadily improved, the gap between supply and demand has become an ever-widening chasm. In the United States, the shortfall exceeds 50000 kidney grafts annually. Until xenografts are a reality, the chasm will remain. Therefore, it is important to identify the factors associated with optimal success in the transplantation of a precious kidney graft. I would like to commend Dr Lee and her colleagues on their analysis. Such studies with a strong scientific foundation can be very influential in the sometimes rancorous debates that determine public policy. A major issue in transplantation is broader sharing of organs. In liver transplantation, this issue has spilled into the public arena where it has been highly politicized. In renal transplantation, a quieter argument is ongoing: should kidney

grafts that are well matched with distant recipients be shipped despite the detrimental effect of cold storage time? The negative effect of prolonged cold storage on graft survival is rooted in biology. We know that preservation begets nonspecific damage such as endothelial injury and cytokine release locally that induces immunologic rejection.

In general, the battle lines over national sharing are drawn between pragmatically minded transplant surgeons on the side of minimizing CIT and laboratory-oriented histocompatibility experts on the side of maximizing donor-recipient HLA match. Transplant clinicians prefer to keep the kidneys locally, sew them in as soon as possible for immediate function, and then rely on highly effective immunosuppression to overcome mismatches. Furthermore, it is contended that local use of grafts preserves community incentive for organ donation and avoids the expense for shipping. The tissue typers (and some clinicians) point to the undeniable better long-time survival when the donor and the recipient are well matched. They note the significant cost of a failed transplant and the other "cost" of recipient sensitization to mismatched donor HLA antigens that may preclude regrafting.

Dr Lee and coauthors have entered the debate armed with data from the United Network for Organ Sharing OPTN/Scientific Registry on over 63 000 kidney transplantations carried out from 1990 to 1998. They conclude that both sides are right. The current policy of national sharing of 0 HLA antigen mismatched renal grafts (about 16% of all renal transplants done in the United States) results in superior graft survival as long as the cold storage time is fewer than 36 hours. Their analysis is a valuable and welcome contribution.

However, there are some problems with their analysis. First, 10% of the transplantations were excluded because of insufficient cold storage and HLA matching data. This was not the authors' fault, but those approximately 8000 grafts might have changed the conclusion. They mention in the article that the mode of preservation, either simple cold storage or pulsatile machine preservation, was not identified in the database. A case can be made for pulsatile machine preservation in the setting of prolonged cold storage; this subset, though small, would be of interest. Do you think "old and cold" 0 antigen mismatched grafts can be resuscitated with pulsatile perfusion? Also, longer follow-up, 10 years or more, may be needed before the benefits of HLA matching, even when grafts were submitted to longer than 36 hours of cold ischemia, become evident.

Again, the efforts of the investigators to refine OMM sharing data are commendable, but they did not go far enough. Perhaps refinements of the analysis will be forthcoming. Donor factors such as age may be a crucial variable. Similarly, relevant recipient factors such as age, diabetes mellitus, level of anti-HLA antibodies and retransplantation status were not examined. The analysis did not include immunosuppression. This is a weakness of the study since potent induction agents are often employed when there is DGF from prolonged cold storage or when grafts are poorly matched. To what extent do the investigators think immunosuppressive strategies can overcome the immunologic consequences of prolonged cold storage? It is possible that the United Network of Organ Sharing OPTN/Scientific Registry computer will crunch the data and dictate to whom the kidney graft should be allocated for the highest success rate (although I suspect a seasoned transplant surgeon could do that if given half the chance). What data crunching suggests is best and what the clinician thinks is best for an

individual patient may differ. Dr Freise, would you accept a OMM kidney from an ideal donor for a recipient with high levels of anti-HLA antibody who has been waiting 8 years for his second transplant knowing this kidney will log 38 to 40 hours of CIT before transplantation?

Finally, in the future, the argument for or against national sharing based on HLA match may be swayed by advances such as more timely HLA typing (eg, wider use of donor peripheral blood), better preservation techniques, improved transport logistics, and new immunosuppressives.

Albert D. Hall, MD, Greenbrae, Calif: What is the present status of the pulsatile perfusion technique? At University of California–San Francisco, we were around when Folkert Belzer developed the perfusion technique that was used in those days.

Dr Roberts: I would like to thank Dr Dafoe for his comments. Whenever you take a national database such as the thousands of kidney transplants that are collected in the United Network of Organ Sharing OPTN/Scientific Registry system, you always have the problem of wanting more data to make sure that the data you do have are realistic. In particular, Dr Dafoe's points about the type of preservation solution, type of immunosuppression, and those kinds of things are important and probably will need to be tested down the road. Transplantation is one of the few areas where we actually have these giant databases to look at the outcome because of the interest by the government, transplant surgeons, and the transplantation community in general to try to improve outcome.

The question regarding whether or not by pulsatile perfusion, which was started really by Dr Belzer, we can add to the CIT and still have good outcomes: Dr Feduska at our institution and Dr Salvatierra published some years ago that for those kidneys that were already old you could get better outcomes by using pulsatile perfusion.

Dr Hall, pulsatile perfusion has been on the wane, primarily because the emphasis now is to try to get the kidneys transplanted in a relatively short period to maximize our outcome. In the older days when we had to wait longer to get our crossmatches back, and it was common to get the kidneys transplanted between 24 to 36 hours or longer, pulsatile perfusion made a difference. Now that we are trying to get the transplants done well under 24 hours, it seems to make less of a difference in outcome and the cost is substantial.

In terms of Dr Dafoe's question regarding how would we handle the patient who has been waiting eight years and his only chance would be to get this kidney that was 40 hours old, but perfectly matched: this is an ethical question where we talk about the balance of utility of the operation vs the justice of trying to transplant this patient who had been waiting 8 years and probably his only chance is today. That is one of the factors that has carried on in the debate about how do we decide to use organs best for transplantation. Do we focus completely on utility or do we say that for this patient, despite the fact that the graft outcome may be lower, we should go ahead and get this transplant because it is fair. These are decisions that can best be made on an individual basis.

To answer the last question, immunosuppression does make a difference in transplantation and with the multitude of new drugs that have appeared in the last several years, we have been able to decrease the chances of rejection early on. These new immunosuppressive drugs are going to make a difference in the slope of the curve of the half-life of cadaveric kidneys following transplantation.