

Protocol Core Needle Biopsy and Histologic Chronic Allograft Damage Index (CADI) as Surrogate End Point for Long-Term Graft Survival in Multicenter Studies

SERDAR YILMAZ,* STEVEN TOMLANOVICH,[†] TIMOTHY MATHEW,[†] EERO TASKINEN,[§] TIMO PAAVONEN,[§] MERCI NAVARRO,[¶] ELEANOR RAMOS,[¶] LEON HOOFTMAN,[¶] and PEKKA HÄYRY^{§#}

*Data Analysis Center, Division of Transplantation, Department of Surgery, University of Calgary, Calgary, Alberta, Canada; [†]US Renal Transplant Mycophenolate Mofetil Study Group, Transplant Service, University of California, San Francisco, California; [‡]Tricontinental Mycophenolate Mofetil Study Group, Queen Elizabeth Hospital, Woodville, South Australia; [§]University of Helsinki Hospital and Central Histopathology Reading Facility, Helsinki Transplantation R&D Ltd, University of Helsinki, Finland; [¶]Roche Global Development, Palo Alto, California; and [#]Rational Drug Design Programme, Biomedicum, and Transplantation Laboratory, University of Helsinki and University of Helsinki Hospital, Finland.

Abstract. This study is an investigation of whether a protocol biopsy may be used as surrogate to late graft survival in multicenter renal transplantation trials. During two mycophenolate mofetil trials, 621 representative protocol biopsies were obtained at baseline, 1 yr, and 3 yr. The samples were coded and evaluated blindly by two pathologists, and Chronic Allograft Damage Index (CADI) score was constructed. At 1 yr, only 20% of patients had elevated (>1.5 mg/100 ml) serum creatinine, whereas 60% of the biopsies demonstrated an elevated (>2.0) CADI score. The mean CADI score at baseline, 1.3 ± 1.1 , increased to 3.3 ± 1.8 at 1 yr and to 4.1 ± 2.2 at 3

yr. The patients at 1 yr were divided into three groups, those with CADI <2, between 2 and 3.9, and >4.0, the first two groups having normal (1.4 ± 0.3 and 1.5 ± 0.6 mg/dl) and the third group pathologic (1.9 ± 0.8 mg/dl) serum creatinine. At 3 yr, there were no lost grafts in the low CADI group, six lost grafts (4.6%) in the in the elevated CADI group, and 17 lost grafts (16.7%) in the high CADI group ($P < 0.001$). One-year histologic CADI score predicts graft survival even when the graft function is still normal. This observation makes it possible to use CADI as a surrogate end point in prevention trials and to identify the patients at risk for intervention trials.

A major problem in the development of new drugs to improve long-term graft survival is how to establish their efficacy. In clinical renal transplantation, 3 to 5 yr are needed to obtain a reliable estimate of graft half-life and to calculate the annual rate of transplant loss. In the analysis of Hunsicker and Bennett (1), based on UNOS registry data, the rate of late graft loss in cadaveric renal transplantation was about 6.9% per year. In trials of primary prevention, in which a reduction of 30% in graft loss is considered clinically significant, it is thus necessary to recruit 1500 patients to achieve statistical significance with 80% power (two-sided $P < 0.05$) or 4500 patients if 90% power is desired. On the other hand, if it were possible to define patients at high risk of progression to chronic rejection, a secondary intervention trial designed to detect a 50% reduction of graft loss rate with 80% power would require the entry

of only 126 patients; with 90% power, around 450 patients would be needed.

We have previously shown in the laboratory (2) and in the clinic (3) that incipient histologic findings in protocol core needle biopsy at 2 yr, quantitated as Chronic Allograft Damage Index (CADI), define patients who will proceed to clinical chronic rejection during the subsequent 4 yr. The early morphologic correlates to late graft outcome have since then been amply confirmed (4–10). These single-center studies suggest that the morphologic correlates may be used as surrogate marker for chronic rejection in primary prevention studies and possibly to identify those patients with normal and stable transplant function, who are prone to develop chronic rejection later.

During the design of two pivotal Mycophenolate Mofetil trials, the US trial (Roche #1866) (11,12) and the Tricontinental trial (Roche #023) (13,14), it was decided that certain centers participating in the trials, 13 centers in the US study and 16 centers in the Tricontinental study, would perform a protocol core needle biopsy at 1 yr posttransplantation and subject the biopsy slides for centralized blinded reading in Helsinki. In addition, some of these centers performed routine implantation biopsies; in two centers, an additional protocol

Received October 3, 2002. Accepted December 5, 2002.

Correspondence to Dr. Pekka Häyry, Transplantation Laboratory, POBox 21 (Haartmaninkatu 3), FIN 00014 University of Helsinki, Finland. Phone: 358-9-19126594; Fax: 385-9-2411227; E-mail: pekka.hayry@helsinki.fi

1046-6673/1403-0773

Journal of the American Society of Nephrology

Copyright © 2003 by the American Society of Nephrology

DOI: 10.1097/01.ASN.0000054496.68498.13

biopsy at 3 yr posttransplant was performed. These biopsies were also quantitated in Helsinki.

The aim of this study was to answer three questions. Is it technically possible to perform a multicenter study using mandatory protocol core needle biopsy and centralized reading of the biopsy slides? Does the 1-yr CADI score predict graft and patient survival at 3 yr also in multicenter studies. Which are the clinical risk factors that lead to high CADI score at 1 yr.

Materials and Methods

Patient Selection

Patient selection and immunosuppressive regimens for the US (Roche #1866) (11,12) and Tricontinental (Roche #0023) (13,14) studies have been described in detail in earlier reports.

Collection of Biopsies

Five microscope slides of each biopsy were collected by Roche Global Development, Palo Alto, coded, and sent to Helsinki for blinded quantitation of the CADI. After assessment, the slides were returned to Palo Alto and forwarded to the participating centers. Roche also collected the clinical data, and both the clinical and biopsy data were delivered together to University of Calgary, where the final analysis was completed.

Renal Biopsies

Sixteen centers in the Tricontinental study and 13 centers in the US study agreed to perform a 1-yr protocol core needle biopsy of all patients in their follow-up, and 388 biopsies were thus obtained. In addition, 122 patients with a 1-yr biopsy were also biopsied at baseline and 229 patients at 3 yr. The final evaluable biopsy material consisted of 739 biopsies.

One hundred seven of these biopsies contained fewer than seven glomeruli and were excluded from the analysis. Eleven additional biopsies were obtained for clinical causes, yet were submitted as “protocol.” These were also excluded, leaving 621 true protocol biopsies with seven or more glomeruli to evaluate.

The evaluable material contained 84 pairs with baseline and 1-yr biopsy, 146 pairs with 1-yr and 3-yr biopsy, and 43 triples where the same patient was followed with baseline, 1-yr, and 3-yr biopsy.

In a retrospective analysis for the actual timing of the biopsies, the baseline biopsies were taken at 0.27 ± 0.96 d (mean \pm SD), 1-yr biopsies at 386.5 ± 39.1 d, and for 3-yr biopsies at 1102.5 ± 25.8 d posttransplantation.

There was a recommendation to use the Uppsala spring-loaded device (15,16) to perform the biopsy. Though all centers did not follow this, no complications of the biopsy procedure were reported during the study.

Histologic Quantitation

For quantification of histologic changes in each biopsy, at least three stained slides were used: one with hematoxylin-eosin, one with Masson trichrome, and one with periodic acid-Schiff (D-PAS). All samples were coded and evaluated blindly with regard to randomization and clinical status. Each histologic sample was reviewed and rated independently by two pathologists. The CADI score obtained was based on the individual component scores for (a) diffuse or focal inflammation and (b) fibrosis in the interstitium, (c) mesangial matrix increase and (d) sclerosis in glomeruli, (e) intimal proliferation of vessels, and (f) tubular atrophy, each individual parameter being scored from 0 to 3 as described (3). If the total CADI score arrived at independently by the two pathologists differed more than one SD from mean, a consensus reading was performed. The data were filed using FileMaker Pro (Claris, California) software in the Helsinki Quantitative Renal Transplant Histopathology Database (17), and transferred to SPSS statistical software packaging (SPSS version 10.0.5) for further handling and statistical analyses.

Statistical Analyses

Spearman correlation coefficient was used to evaluate the correlation between the two pathologists in quantitation of CADI (Figure 1).

Multivariate logistic regression analysis was employed to identify independent predictors of 3-yr graft loss (including patient death). The model included variables that were significant in a univariate analysis ($P < 0.05$) or were of biologic interest. Independent variables in-

Table 1. Summary of baseline, 1-yr, and 3-yr Chronic Allograft Damage Index (CADI) scores

Patient Cohort	CADI Score		
	Baseline	12 mo	36 mo
All patients			
No. of biopsies	111	302	206
mean CADI score \pm SD	1.3 ± 1.1	3.3 ± 1.8	4.1 ± 2.2
range	0 to 5.5	0 to 10.8	0 to 11.3
Patients with biopsies at baseline, 12, and 23 mo			
No. of biopsies	43	43	43
mean CADI score \pm SD	1.4 ± 1.4	3.3 ± 2.1	4.3 ± 2.1
Patients with biopsies at baseline and 12 mo			
No. of biopsies	84	84	
mean CADI score \pm SD	1.2 ± 1.1	3.4 ± 1.6	
Patients with biopsies at 12 and 36 mo			
No. of biopsies		146	146
mean CADI score \pm SD		3.3 ± 1.8	4.1 ± 2.2

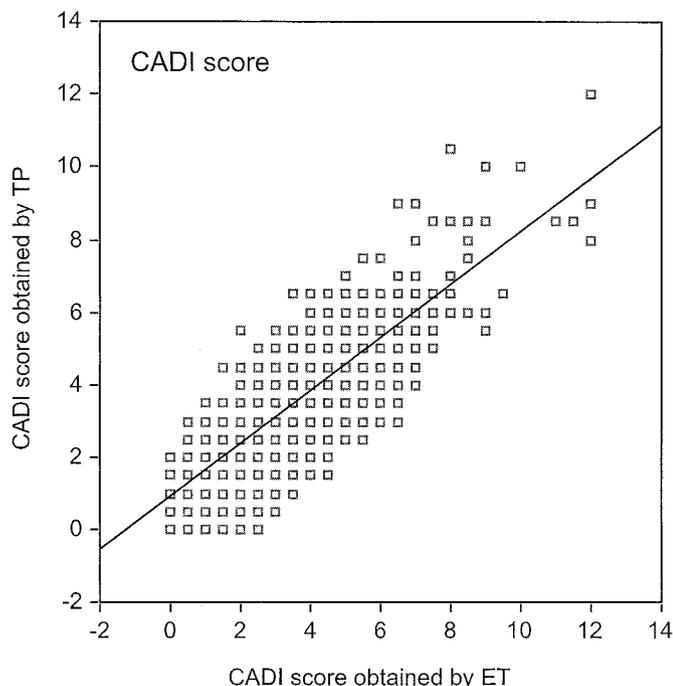


Figure 1. Correlation between the two pathologists in quantitation of the Chronic Allograft Damage Index (CADI) in 621 representative protocol core needle biopsies. Spearman correlation coefficient 0.85.

cluded in the model are listed in Table 2. Odds ratios (OR) and corresponding 95% confidence intervals were calculated. *P*-values less than 0.05 were considered to indicate statistical significance.

Multivariate linear regression analysis was performed to identify independent predictors of the 1-yr CADI score (Table 3). *P* values less than 0.05 were considered to indicate statistical significance.

A χ^2 test for multiple proportions was conducted for categorical data (Table 4).

Results

Interobserver Variation

The Spearman correlation coefficient in sum CADI score between the two pathologists was 0.85 (Figure 1). In case the

total CADI score arrived at independently by the two pathologists differed more than one SD from mean, a consensus reading was performed. Of all biopsies, 11.7% required consensus reading.

Progression of the CADI Score During Three Years Posttransplantation

Whereas the mean serum creatinine in all patients at 1 yr was still close to normal, 1.6 ± 0.7 mg/dl (mean \pm SD), the mean CADI score had already progressed to 3.3 ± 1.8 (Figure 2). At 1 yr, only 20% of patients had elevated (>1.5 mg/dl) serum creatinine, whereas 60% of the 1-yr biopsies demonstrated an elevated, (>2.0) (3) CADI score.

The progression of the CADI score during the first 3 yr is shown in Table 1. In the total cohort of 621 representative protocol biopsies, the mean CADI score was 1.25 ± 1.13 (SD) at baseline (111 biopsies), 3.27 ± 1.82 at 1 yr (302 biopsies), and 4.10 ± 2.15 (208 biopsies) at 3 yr. We also determined separately the progression of the CADI score in those patients within the cohort who were biopsied two or three times during the study (Table 1). The baseline and progression of the CADI score in these subgroups was not any different of the total cohort. Thus the histologic changes relevant to chronic rejection developed fastest during the first year, approximately 2 CADI units/yr, but less rapidly during the subsequent 2 yr of follow up, 0.5 CADI units per year.

Factors Predicting 3-yr Survival

Of the 302 patients with representative protocol biopsy at 1 yr, 23 patients experienced either graft loss or death at 3 yr when the two studies were combined. Multivariate logistic regression modeling revealed that the occurrence of graft loss or patient death at 3 yr posttransplant was significantly associated with the 1-yr CADI score ($P = 0.0003$; OR = 1.6), the presence of acute rejection during the first year ($P = 0.048$; OR = 2.8), and primary disease (hypertension or diabetes mellitus) as the reason of renal failure ($P = 0.036$; OR = 2.9) (Table 2). The data indicate, furthermore, that for every unit

Table 2. Parameters correlating (*P*) with 3-yr graft loss or patient death in the combined study^a

	OR	<i>P</i>	95% CI
CADI score at 1 yr	1.6	0.0003 ^b	1.3 to 2.1
Rejection during the first year	2.8	0.048 ^b	1.0 to 7.1
Primary disease (diabetes or hypertension)	2.9	0.036 ^b	1.1 to 8.3
HLA DR mismatches	0.8	0.594	0.4 to 1.6
CMV infection during the first year	0.6	0.392	0.2 to 2.0
Non-White ethnicity	0.7	0.602	0.3 to 2.3
Male gender	1.1	0.844	0.4 to 2.8
Cold ischemia time (>24 h)	0.8	0.581	0.3 to 2.0
Donor age (>60 -yr-old)	0	0.699	0 to 8.2
Patient with delayed function	1.8	0.352	0.5 to 5.8

^a *P* values were calculated from logistic regression model including all explanatory variables. Delayed graft function is defined as the need for dialysis during the first week posttransplant CMV infection is tissue invasive disease and/or viremia.

^b *P* < 0.05 are considered significant.

Table 3. Parameters correlating (*P*) with high CADI score at 1 yr in the US, Tricontinental, and combined studies^a

	US	Tricontinental	Combined
Donor age	0.0023*	0.00001 ^b	0.00001 ^b
Rejection during the first year	0.0091*	0.00003 ^b	0.00001 ^b
CMV infection during the first year	0.0056*	0.033 ^b	0.217
Cold ischemia time	0.407	0.085	0.922
Delayed graft function	0.330	0.193	0.053
No. of total mismatch	0.595	0.403	0.207
Primary disease (diabetes or hypertension)	0.638	0.719	0.485
Male gender	0.103	0.846	0.290
Non-White ethnicity	0.777	0.206	0.357
Panel reactive antibody (latest)	0.464	0.311	0.143

^a *P* values calculated from multiple linear regression model including all explanatory variables. Delayed graft function is defined as the need for dialysis during the first week posttransplant CMV infection is tissue invasive CMV.

^b *P* < 0.05 are considered significant.

Table 4. Correlation between 1-yr CADI score and patient/graft survival at 3 yr (patient with representative protocol biopsy)

	CADI (1 yr)			All	<i>P</i> ^a
	<2	2.0 to 3.9	>4.0		
S-creatinine	1.4 ± 0.3	1.5 ± 0.6	1.9 ± 0.8		
Number of patients	68	132	102	302	
Death	0	2	5	7	
Death-censored graft loss	0	4	12	16	<0.001
All graft losses %	0	6	17	23	<0.001
	0%	4.6%	16.7%		

^a χ^2 multiple proportion.

increase in CADI score, the odds of a graft loss or death increased by almost 50%.

Factors Predicting High 1-yr CADI Score

We also investigated whether the known risk factors for graft loss, previously defined in clinical studies (mostly retrospective), correlate with high 1-yr CADI score. Here the results of the two studies are given separately, as the data in the US and Tricontinental studies were somewhat different, as was the result when the two studies were combined. Multivariate linear regression analysis revealed that donor age (*P* = 0.0001) and rejection during the first year (*P* = 0.0001) were significantly associated with high CADI score. CMV infection during the first year correlated in the US (*P* = 0.006) and in the Tricontinental study (*P* = 0.033) but not when the studies were combined. The remaining recognized risk factors placed in the model, *i.e.*, cold ischemia time, delayed graft function, number of HLA-DR mismatches, primary disease, gender, ethnic background, and latest panel reactive antibody level, did not show correlation with high CADI score at 1 yr (Table 3).

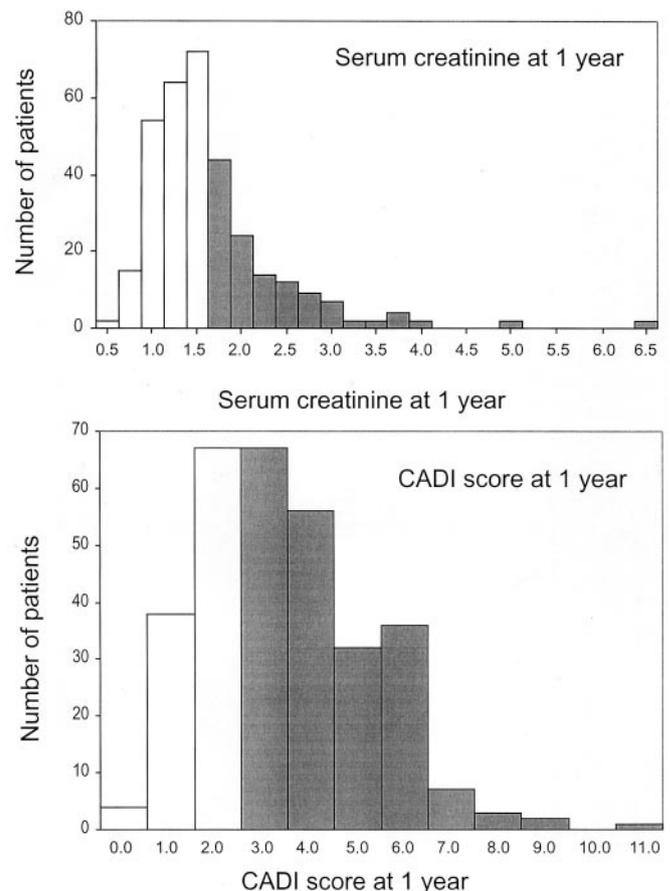


Figure 2. The histogram showing the distribution of CADI score and serum creatinine (mg/dl) at 1 yr, and the number of patients who fell within each interval. Elevated values are shaded.

Correlation of 1-yr CADI to Renal Function and Graft Survival at 3 yr

To investigate the predictive value of 1-yr CADI score for late graft loss, the 302 patients with a representative protocol biopsy at 1 yr were divided into three groups: those with CADI <2, those with CADI between 2 and 3.9, and those with CADI

>4.0. The respective serum creatinine levels were 1.4 ± 0.3 (SD), 1.5 ± 0.6 , and 1.9 ± 0.8 mg/dl. This analysis thus defined one group with normal serum creatinine and low CADI, one with normal serum creatinine and elevated CADI, and one with slightly elevated serum creatinine and high CADI. When the clinical and biopsy status at 1 yr was correlated with the clinical outcome at 3 yr, not a single patient lost their graft in the low CADI group, six patients (4.6%) lost their graft in the in the elevated CADI group and 17 patients (16.7%) lost their graft in the high CADI group ($P < 0.001$). When death with a functioning graft was censored, the outcome was no lost grafts in the low CADI group, four lost grafts in the elevated CADI group, and 12 lost grafts in the high CADI group ($P < 0.001$) (Table 4).

Discussion

In a recent analysis of UNOS Registry data from 1988 to 1996, 1-yr cadaveric renal allograft survival has improved from 75.7 to 87.7%, as has transplant half-life after the first year from 7.9 to 13.8 yr (18). On the other hand, the half-life of transplants with one or more acute episodes of rejection improved only modestly, from 9.1 to 11.1 yr (18). Here, acute vascular rejection (19), often manifesting as steroid-resistant rejection with a strong humoral component (20), is a major negative prognostic indicator. With the decrease of graft loss during the first year, late graft loss after the first year has become the major reason of renal allograft failure.

The most frequent reason for late cadaveric renal allograft loss is death with a functioning transplant (53%) followed by chronic allograft rejection (36%) (21). The main cause of death in patients with functioning transplant is cardiovascular (22). Thus the two main reasons for late allograft failure, chronic rejection, manifesting as fibroproliferative intragraft vascular disease (23), and accelerated atherosclerosis, are closely related and may potentially respond to similar therapy.

There is ample evidence from eight single-center studies (3–10) that incipient histologic changes characteristic of chronic rejection are visualized in the transplant before the transplant function deteriorates. Four of these studies have employed Banff histologic criteria in their evaluation (6–8,10), one study the amount of collagen/fibrosis in the biopsy (9), and the remaining three a sum score of various findings in the graft interstitium, glomeruli, vessels, and tubuli (3–5). The CADI score is the most extensive of these in regard to the number of parameters quantitated and, in contrast to the Banff criteria for chronic rejection, takes also into account the vascular and glomerular changes in the biopsy.

Implementation of protocol core needle biopsy in a multicenter study was met with two difficulties. There was clinical concern at performing protocol biopsies in a well-functioning transplant. Thus, only about one third of the centers complied with this, though within these centers all of the participating patients were biopsied at 1 yr (unless there was a clinical contraindication to performing the biopsy). Second, to monitor the progression of the lesion, two or preferably three biopsies would be needed from a single patient, but only five centers complied with two biopsies and only two centers were willing

to biopsy three times. Thus such follow-ups are clearly under-represented in this study. The biopsies were mostly of good quality; 86% contained at least seven glomeruli, and the timing of the biopsy procedures followed closely the protocol.

Both trials, (US and Tricontinental), were originally planned for 12 mo of follow-up, but they were extended thereafter to 3 yr. The original sample sizes chosen were adequate to detect a reduction in acute rejection rate at 6 mo. The studies were not designed to test the hypothesis of whether chronic rejection or graft loss at 3 yr were impacted; therefore, it is not surprising that there were insufficient numbers of patients in these trials to address these factors. For this reason and because the patients who provided biopsy data no longer represented the total population, the effect of treatment (*i.e.*, who received MMF versus placebo/azathioprine) was not addressed. The similar design of the two studies, however, enabled us to pool the data, when investigating the predictive value of protocol biopsy, as scored by the CADI criteria, for graft outcome, keeping in mind that the pool of patients with available biopsy data does not necessarily represent the total population in the two studies.

Most importantly, this study demonstrates that it is possible to perform a multicenter protocol biopsy-based trial with centralized biopsy scoring if the participating centers are selected for their willingness to perform the protocol biopsies. The study also confirmed the predictive value of the biopsy to subsequent graft outcome in a multicenter trial. Importantly, no major complications were observed in this study and no fatalities were reported as consequence of biopsy.

The results also show that high 1-yr CADI score correlates with known risk factors to chronic rejection, including donor age (24–26), acute rejection during first year (27,28), and CMV infection (29) (only in the separate studies but not in combined study). The fact that other frequently cited risk factors, such as cold ischemia time, delayed graft function, number of HLA mismatches, and others (for references, see references 30 and 31), did not correlate may be explained by too short interval between the transplant and the biopsy.

Additionally an elevated CADI score at 1 yr correlates with graft survival at 3 yr, both in patients with normal and elevated serum creatinine at the time of the biopsy. In other words, histopathologic findings in protocol biopsy are more sensitive markers than change in transplant function, as detected by serum creatinine level. At 1 yr, only 20% of patients had elevated (>1.5 mg/dl) serum creatinine, whereas 60% of the 1-yr biopsies demonstrated already an elevated (*i.e.* >2.0)(3) CADI score. When the clinical and biopsy status at 1 yr was correlated with the clinical outcome at 3 yr, not a single patient lost their graft in the low CADI group with normal serum creatinine, six patients (4.6%) lost their graft in the in the elevated CADI group also with normal serum creatinine, and (not unexpectedly) 17 patients (16.7%) lost their graft in the high CADI group displaying already an elevated serum creatinine level ($P < 0.001$). When death with a functioning graft was censored, the outcome was no lost grafts in the low CADI group, four lost grafts in the elevated CADI group, and 12 lost grafts in the high CADI group ($P < 0.001$). Although this analysis is based on a small cohort of patients with evaluable

biopsy data at 1 yr, that the number of lost grafts at 3 yr was still small, and that the results need confirmation after 5 to 10 yr follow-up, the trend is quite clear. Our findings in the present study and those reported earlier, suggest that protocol core needle biopsy and CADI can be used as surrogate end point in primary prevention trials and possibly to identify the cohort of patients at risk for chronic rejection in secondary intervention trials, unless the 1-yr time point is too late to interfere with the disease.

We observed a rapid progression of the CADI from baseline to 1 yr. The pace slowed down during the subsequent years. The fast progression of CADI during the first year is compatible with the paradigm that chronic rejection represents graft response to cumulative injury, occurring mostly during the first year, regardless whether it is immunologic or non-immunologic in origin (23).

The baseline CADI in this study indicates that most of the transplants were satisfactory in quality, with a mean CADI at baseline of only 1.25. The range (0 to 5.3) shows, however, that some suboptimal transplants were also included in the study. This observation emphasizes the need to obtain a baseline biopsy to calculate the progression of the histologic manifestations during the first year. Assessment of the Δ CADI, rather than an isolated measure of the CADI at 1 yr, is necessary to accurately differentiate the outcome between patients destined to progress (progressors) and those unlikely to do so (non-progressors). The number of grafts lost at 3 yr was too small to make meaningful comparisons between progressors and non-progressors.

The histologic findings characteristic of acute rejection and histologic sequelae of an immunologic insult do not appear to be different in the MMF era (where the data in this manuscript is derived from) compared with those resulting from the use of newer agents that have resulted in lower rejection rates. Therefore, we believe that the conclusion that higher CADI score leads to poorer graft survival is still valid.

Taken together, our results in two multicenter trials confirm the results of several single-center studies that early histologic alterations, quantitated here as CADI, predict chronic rejection, even when the graft function is still normal. This observation will make it possible to use the approach not only as a surrogate end point in prevention trials, but also to identify the patient cohort at risk for intervention trials. One important lesson of this study, however, is that it is more meaningful to investigate the progression of the lesion (Δ CADI) rather than the intensity of alterations at a single given time point.

Acknowledgments

We are grateful to the clinical centers contributing with the biopsies. These centers and investigators are listed in the original publications (references 11 to 14); because of lack of space, they are not listed here. The authors acknowledge the financial support from University of Helsinki Hospital Research Funds and F. Hoffmann, La Roche, Palo Alto, CA, and the work of Ms. Eerika Wasenius, RN (Lab), in expert handling of the biopsy slides and filing of the results.

References

- Hunsicker LG, Bennett LE: Design of trials of methods to reduce late renal allograft loss: The price of success. *Kidney Int Suppl* 52: S120–S123, 1995
- Yilmaz S, Taskinen E, Paavonen T, Mennander A, Hayry P: Chronic rejection of rat renal allograft. I: Histological differentiation between chronic rejection and cyclosporin nephrotoxicity. *Transpl Int* 5: 85–95, 1992
- Isoniemi H, Taskinen E, Hayry P: Histological chronic allograft damage index accurately predicts chronic renal allograft rejection. *Transplantation* 58: 1195–1198, 1994
- Kasiske BL, Kalil RS, Lee HS, Rao KV: Histopathologic findings associated with a chronic, progressive decline in renal allograft function. *Kidney Int* 40: 514–524, 1991
- Dimeny E, Wahlberg J, Larsson E, Fellstrom B: Can histopathological findings in early renal allograft biopsies identify patients at risk for chronic vascular rejection? *Clin Transplant* 9: 79–84, 1995
- Seron D, Moreso F, Bover J, Condom E, Gil-Vernet S, Canas C, Fulladosa X, Torras J, Carrera M, Grinyo JM, Alsina J: Early protocol renal allograft biopsies and graft outcome. *Kidney Int* 51: 310–316, 1997
- Legendre C, Thervet E, Skhiri H, Mamzer-Bruneel MF, Cantarovich F, Noel LH, Kreis H: Histologic features of chronic allograft nephropathy revealed by protocol biopsies in kidney transplant recipients. *Transplantation* 65: 1506–1509, 1998
- Nickerson P, Jeffery J, Gough J, McKenna R, Grimm P, Cheang M, Rush D: Identification of clinical and histopathologic risk factors for diminished renal function 2 years posttransplant. *J Am Soc Nephrol* 9: 482–487, 1998
- Nicholson ML, Bailey E, Williams S, Harris KP, Furness PN: Computerized histomorphometric assessment of protocol renal transplant biopsy specimens for surrogate markers of chronic rejection. *Transplantation* 68: 236–241, 1999
- Seron D, Moreso F, Ramon JM, Hueso M, Condom E, Fulladosa X, Bover J, Gil-Vernet S, Castela AM, Alsina J, Grinyo JM: Protocol renal allograft biopsies and the design of clinical trials aimed to prevent or treat chronic allograft nephropathy. *Transplantation* 69: 1849–1855, 2000
- Sollinger H W: Mycophenolate mofetil for the prevention of acute rejection in primary cadaveric renal allograft recipients. U.S: Renal Transplant Mycophenolate Mofetil Study Group. *Transplantation* 60: 225–232, 1995
- Mycophenolate mofetil in cadaveric renal transplantation. US Renal Transplant Mycophenolate Mofetil Study Group. *Am J Kidney Dis* 34: 296–303, 1999
- A blinded, randomized clinical trial of mycophenolate mofetil for the prevention of acute rejection in cadaveric renal transplantation. The Tricontinental Mycophenolate Mofetil Renal Transplantation Study Group. *Transplantation* 61: 1029–1037, 1996
- Mathew TH: A blinded, long-term, randomized multicenter study of mycophenolate mofetil in cadaveric renal transplantation: Results at three years. Tricontinental Mycophenolate Mofetil Renal Transplantation Study Group. *Transplantation* 65: 1450–1454, 1998
- Mackenzie JC, Mackay IG, Millar ND, MacIver AG, Penry JB: The Biopsy cut procedure for renal biopsies. *Br Med J (Clin Res Ed)* 296: 1128–1129, 1988
- Wahlberg J, Andersson T, Busch C, Fellstrom B, Frodin L, Larson A, Larsson E, Lindgren PG, Tufveson G: The biopsy technique: a major advance in the monitoring of renal transplant recipients. *Transplant Proc* 20: 419–420, 1988
- Hayry P, Yilmaz S, Taskinen E, Isoniemi, H: Intermediate efficacy endpoints for chronic kidney allograft rejection: Implica-

- tions of protocol core biopsy for early registration of new agents, In: *Principles of Drug Development in Transplantation and Autoimmunity*, edited by Lieberman R, Mukherjee A, Austin, TX, R.G. Landes Company, 1996
18. Hariharan S, Johnson CP, Bresnahan BA, Taranto SE, McIntosh MJ, Stablein, D: Improved graft survival after renal transplantation in the United States, 1988 to 1996. *N Engl J Med* 342: 605–612, 2000
 19. van Saase JL, van der Woude FJ, Thorogood J, Hollander AA, van Es LA, Weening JJ, van Bockel JH, Bruijn J A: The relation between acute vascular and interstitial renal allograft rejection and subsequent chronic rejection. *Transplantation* 59: 1280–1285, 1995
 20. Crespo M, Pascual M, Tolkoff-Rubin N, Mauiyyedi S, Collins AB, Fitzpatrick, D., Farrell ML, Williams WW, Delmonico FL, Cosimi AB, Colvin RB, Saidman SL: Acute humoral rejection in renal allograft recipients: I. Incidence, serology and clinical characteristics. *Transplantation* 71: 652–658, 2001
 21. Berthoux F, Abramowicz D, Bradley B, Ekberg H, Frei U, Morales J, Olgaard K, Paul L, Ponticelli, C: European Best Practise Guidelines for Renal Transplantation (Part II). *Nephrol Dial Transplantation* 17: MDNM, 2002
 22. Ojo AO, Hanson JA, Wolfe RA, Leichtman AB, Agodoa LY, Port F K: Long-term survival in renal transplant recipients with graft function. *Int Kidney* 57: 307–313, 2000
 23. Koskinen P, Lemstrom K, Hayry P: Chronic rejection. *Curr Opin Nephrol Hypertens* 5: 269–272, 1996
 24. Rao KV, Kasiske BL, Odlund MD, Ney AL, Andersen RC: Influence of cadaver donor age on posttransplant renal function and graft outcome. *Transplantation* 49: 91–95, 1990
 25. Pirsch JD, D'Alessandro AM, Sollinger HW, Hoffmann RM, Roecker E, Voss BJ, Lorentzen D, Knechtle SJ, Reed A, Kallayoglu M: The effect of donor age, recipient age, and HLA match on immunologic graft survival in cadaver renal transplant recipients. *Transplantation* 53: 55–59, 1992
 26. Alexander JW, Bennett LE, Breen TJ: Effect of donor age on outcome of kidney transplantation. A two-year analysis of transplants reported to the United Network for Organ Sharing Registry. *Transplantation* 57: 871–876, 1994
 27. Basadonna GP, Matas AJ, Gillingham K J, Payne WD, Dunn DL, Sutherland DE, Gores PF, Gruessner RW, Najarian JS: Early versus late acute renal allograft rejection: Impact on chronic rejection. *Transplantation* 55: 993–995, 1993
 28. Matas AJ, Gillingham KJ, Payne WD, Najarian JS: The impact of an acute rejection episode on long-term renal allograft survival ($t_{1/2}$): *Transplantation* 57: 857–859, 1994
 29. Rubin RH, Tolkoff-Rubin NE, Oliver D, Rota TR, Hamilton J, Betts RF, Pass RF, Hillis W, Szmuness W, Farrell ML: Multi-center seroepidemiologic study of the impact of cytomegalovirus infection on renal transplantation. *Transplantation* 40: 243–249, 1985
 30. Najarian JS, Gillingham KJ, Sutherland DE, Reinsmoen NL, Payne WD, Matas AJ: The impact of the quality of initial graft function on cadaver kidney transplants. *Transplantation* 57: 812–816, 1994
 31. Yokoyama I, Uchida K, Kobayashi T, Tominaga Y, Orihara A, Takagi H: Effect of prolonged delayed graft function on long-term graft outcome in cadaveric kidney transplantation. *Clin Transplant* 8: 101–106, 1994

Access to UpToDate on-line is available for additional clinical information
at <http://www.jasn.org/>