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Increase in proteinuria >200 mg/g after late rejection is associated with poor graft survival

Arjang Djamali1,2, Millie Samaniego3, Jose Torrealba4, John Pirsch1,2 and Brenda L Muth1

1University of Wisconsin Madison SMPH, Department of Medicine, 2University of Wisconsin Madison SMPH, Department of Surgery, 3University of Wisconsin Madison SMPH, Department of Pathology and 4University of Michigan Ann Arbor

Correspondence and offprint requests to: Arjang Djamali; E-mail: axd@medicine.wisc.edu

Abstract

Background. There is no information on the effects of proteinuria on outcomes following rejection.

Methods. We addressed this question in a retrospective study of 925 kidney transplant recipients between January 2003 and December 2007. Selection criteria were based on (i) biopsy proven diagnosis of a first episode of acute rejection, and (ii) available data on urine protein to creatinine (UPC) ratios at baseline (lowest serum creatinine before biopsy), time of biopsy and 1 month after biopsy. We examined the effects of a change in UPC (ΔUPC = UPC 1 month after biopsy—baseline UPC) on outcomes.

Results. We identified 82 patients with both acute rejection and available data on proteinuria. Mean time (±SE) to acute rejection was 19 ± 2.3 months, and patients were followed up for 38.7 ± 2.6 months after transplant. Median ΔUPC was 200 mg/g (95% confidence interval 0.00 to 0.300). Forty-two patients had a ΔUPC ≥200 (high pro-
teinuria group). Baseline characteristics were similar between high and low proteinuria groups except for more induction therapy with interleukin-2 receptor blockade in the former (71 vs. 47%, P = 0.04). Patient with ΔUPC ≥200 had higher rates of graft loss (26 vs. 15%, P = 0.01) or combined graft loss or death (38 vs. 20%, P = 0.002 by log–rank). In univariate and multivariate Cox regression analyses, ΔUPC ≥200 mg/g, sirolimus therapy 1 month after rejection and re-transplant status were significant factors associated with death-censored graft loss (hazard ratio (HR) 4.4, 14.9 and 6.2, P ≤ 0.008) or combined graft loss or patient death (HR 3.8, 6.5 and 3.9, P ≤ 0.03).

**Conclusions.** An increase in proteinuria ≥200 mg/g after late acute rejection is associated with poor graft survival and patient outcomes. Clinical trials are needed to determine whether post-rejection anti-proteinuric strategies improve outcomes.

**Keywords:** kidney transplantation; outcomes; proteinuria; rejection

### Introduction

Recovery of renal function following acute rejection is more indicative of long-term graft survival than acute rejection per se [1]. Patients who had an acute rejection episode and whose serum creatinine levels returned to within 95% of baseline levels at 1 year post-transplantation demonstrated similar long-term graft survival as those who never experienced acute rejection [1]. In contrast, an incrementally greater risk of graft loss was associated with failure to restore 95% of baseline renal function in the same time frame. These data indicate that the subgroups of patients with acute rejection who do not regain adequate renal function are most vulnerable to graft loss.

We questioned if this was true for proteinuria. Post-transplant proteinuria may arise from the native kidneys in the early post-transplant period; transplant glomerulopathy, recurrent or de novo glomerular disease, chronic allograft tubulointerstitial injury, obesity and drugs including sirolimus [2,3]. In a study of kidney transplant recipients undergoing protocol biopsies at 1 year post-transplant, 45% had proteinuria greater than 150 mg/d [4]. In this study, proteinuria was associated with reduced graft survival independent of other risk factors including glomerular disease, graft function, recipient age and acute rejection [4]. These results have been confirmed by others showing that post-transplant proteinuria is associated with poor graft and patient outcomes [5–9]. However, there is limited data on the relationship between acute rejection, proteinuria and outcomes. We hypothesized that an increase in proteinuria after acute rejection is associated with poor patient and allograft survival.

### Subjects and methods

**Patients**

We tested our hypothesis in a retrospective study of 925 kidney transplant recipients between January 2003 and December 2007. Selection criteria were based on two elements [1], biopsy proven first acute rejection [2] and available data on urine protein to creatinine (UPC) ratios at baseline (time of lowest serum creatinine before biopsy), time of biopsy and 1 month after biopsy. We examined the effects of a change in UPC (ΔUPC = UPC 1 month after biopsy—baseline UPC) on outcomes. To achieve this goal, we first defined the range of ΔUPC after rejection. We then divided patients in two groups (high and low proteinuria) based on the median ΔUPC and compared baseline characteristics and outcomes between the high (ΔUPC ≥ median value) and low (ΔUPC < median value) proteinuria groups. Early morning urine samples prior to the clinic visit were used to measure UPC levels.

We collected clinical information and laboratory values at baseline, rejection and 1-month follow-up. These included age, gender, race, body mass index (BMI), cause of end-stage renal disease (ESRD) including diabetes mellitus (DM) type 1, DM type 2, hypertension (HTN), glomerular disease (GD), polycystic kidney disease (PKD) or other, transplant type: kidney–pancreas (KP), living donor (LDKT) or deceased donor (DDKT), number of rejection episodes after the first biopsy proven rejection, times re-transplanted, length of pre-transplant dialysis, serum creatinine (Scr), MDRD-based estimated GFR (eGFR = 175 × standardized Scr) and diabetes mellitus (DM) type 1, DM type 2, hypertension (HTN), glomerular disease or BK nephropathy. We reviewed information on induction and maintenance immunosuppression before and after rejection as well as treatment for rejection. We collected data on the use of statins and angiotensin-converting enzyme inhibitors (ACE-I) or angiotensin receptor blockers (ARB) at rejection and at 1 month after rejection, mean arterial pressure (MAP), HbA1c, LDL cholesterol and albumin at 1 month after rejection. All analyses were performed with the approval of the UW Hospital and Clinics Institutional Review Board.

**Histopathology and treatment modalities**

All kidney biopsies were indicated based on a rise in serum creatinine >20% from baseline. Acute rejection was diagnosed according to the Banff criteria [10–12]. Acute cellular rejection was reported as suspicious, IA, IB and IIA. The diagnosis of acute antibody-mediated rejection was primarily based on C4d scoring and peritubular capillaritis. In addition, we used donor-specific antibody titers using single antigen bead Lumines assays after 2006. Transplant glomerulopathy (TG) was defined by the characteristic duplication of glomerular basement membrane (GBM) observed by light microscopy, as recommended by the Banff working group [10–12]. Interstitial (I), tubular (T), vascular (V) and glomerular (G) scores were reported according to Banff classification of acute and chronic kidney allograft injury [10–12].

All patients with acute rejection received high-dose steroids. Rabbit anti-thymocyte globulin was used in patients with steroid-resistant cellular rejection. Rituximab, IV Ig and plasmapheresis were used in patients with antibody-mediated rejection based on the preference of the provider. The overall dose of CNI was reduced in the CNI-induced nephrotoxicity group, in general by at least 25%.

**Statistical analysis**

We first defined the range of ΔUPC after rejection. We then divided patients in two groups (high and low proteinuria) based on the median ΔUPC and compared baseline characteristics and outcomes between the high (ΔUPC ≥ median value) and low (ΔUPC < median value) proteinuria groups. Univariate regression analyses were performed to determine independent covariates affecting death-censored graft loss and patient death. These covariates included age, gender, race, BMI, cause of ESRD, type of transplant, type of rejection, times re-transplanted, length of pre-transplant dialysis, biochemical characteristics such as Scr, UPC, HbA1c, LDL cholesterol, serum albumin, hepatitis C antibodies, immunosuppressive regimens (induction and maintenance), treatment for rejection, use of statins at rejection and 1 month, and use of ACE-I or ARB at rejection and 1 month. Those variables that were statistically significant (P ≤ 0.05) were then entered into stepwise multivariable Cox regression analyses. Variables were retained if P ≤ 0.05 and dismissed if P > 0.05. Next, graft and patient survival rates were modelled using Kaplan–Meier survival curves. The time interval was time of transplant to outcome. Data analysis was performed using Microsoft Excel 2003, version 11.8012.6568 and MedCalc, version 10.
Results

Median change in proteinuria after rejection was 200 mg/g

Figure 1 displays the non-parametric distribution of ΔUPC after rejection. The median ΔUPC was 200 mg/g with a 95% confidence interval (CI) of 0 to 300 mg/g. The majority (60%) of patients had zero to 1000 mg/g proteinuria. We defined two groups based on the median ΔUPC. There were 42 patients with ΔUPC ≥200 (high proteinuria group) while 40 patients had a ΔUPC <200 (low proteinuria group).

Baseline characteristics and immunosuppression

Table 1 represents the baseline characteristics of patients with high versus low proteinuria. A total of 82 patients fulfilled our selection criteria between 2003 and 2007. The mean follow-up after transplant was 38.7 ± 2.6 months. The mean time to biopsy was 19 ± 2.3 months. The two groups were comparable except for induction therapy. Alemtuzumab (Campath) and Basiliximab (Simulect) induction were used in 14 and 71% of patients with high proteinuria (P = 0.03 and 0.04, respectively). There were no differences in immunosuppression at baseline and 1 month after rejection as there were no significant differences between blood pressure (MAP), ACE or ARB use, LDL and HbA1c levels. Donor age was similar between the two groups (42 ± 2.3 vs. 44 ± 3.3 years, P = not significant (ns)).

Histopathological findings and treatment

There were 13 (31%), 15 (36%) and 16 (38%) patients with antibody-mediated, cellular and mixed rejection in the high proteinuria group (Table 2). Banff scores includ-
To convert Scr to micromole per liter multiply by 88.4.

Table 2. Kidney function and outcomes

<table>
<thead>
<tr>
<th></th>
<th>ΔUPC ≥ 200</th>
<th>ΔUPC &lt; 200</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Scr at baseline (mg/dL)</td>
<td>1.6 ± 0.05</td>
<td>1.7 ± 0.12</td>
<td>0.8</td>
</tr>
<tr>
<td>Scr at biopsy (mg/dL)</td>
<td>2.9 ± 0.3</td>
<td>2.5 ± 0.1</td>
<td>0.3</td>
</tr>
<tr>
<td>Scr 1 month after biopsy (mg/dL)</td>
<td>1.9 ± 0.08</td>
<td>1.8 ± 0.08</td>
<td>0.2</td>
</tr>
<tr>
<td>ΔScr</td>
<td>0.32 ± 0.07</td>
<td>0.05 ± 0.01</td>
<td>0.06</td>
</tr>
<tr>
<td>Baseline eGFR (ml/min/1.73 BSA)</td>
<td>48.6 ± 2</td>
<td>47.6 ± 3</td>
<td>0.7</td>
</tr>
<tr>
<td>eGFR at biopsy (ml/min/1.73 BSA)</td>
<td>30 ± 2</td>
<td>33 ± 2</td>
<td>0.4</td>
</tr>
<tr>
<td>eGFR 1 month after biopsy (ml/min/1.73 BSA)</td>
<td>40 ± 2</td>
<td>43 ± 2</td>
<td>0.3</td>
</tr>
<tr>
<td>ΔeGFR</td>
<td>8 ± 2</td>
<td>-4.3 ± 2</td>
<td>0.2</td>
</tr>
<tr>
<td>UPC baseline (mg/g)</td>
<td>0.5 ± 0.07</td>
<td>0.8 ± 0.2</td>
<td>0.3</td>
</tr>
<tr>
<td>UPC at biopsy (mg/g)</td>
<td>1.5 ± 0.2</td>
<td>1.2 ± 0.3</td>
<td>0.01</td>
</tr>
<tr>
<td>UPC 1 month after biopsy (mg/g)</td>
<td>1.6 ± 0.2</td>
<td>0.5 ± 0.1</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>ΔUPC</td>
<td>1.15 ± 0.2</td>
<td>-0.3 ± 0.14</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Graft loss</td>
<td>5 (12)</td>
<td>2 (5)</td>
<td>0.07</td>
</tr>
<tr>
<td>Death</td>
<td>11 (26)</td>
<td>6 (15)</td>
<td>0.01</td>
</tr>
<tr>
<td>Graft loss or death</td>
<td>16 (38)</td>
<td>8 (20)</td>
<td>0.002</td>
</tr>
</tbody>
</table>

To convert Scr to micromole per liter multiply by 88.4.

*aDifference between 1 M post-biopsy and baseline.*

Table 3. Biopsy characteristics

<table>
<thead>
<tr>
<th></th>
<th>ΔUPC ≥200</th>
<th>ΔUPC &lt;200</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Antibody mediated rejection (%)</td>
<td>13 (31)</td>
<td>8 (20)</td>
<td>0.3</td>
</tr>
<tr>
<td>Cellular rejection (%)</td>
<td>15 (36)</td>
<td>18 (45)</td>
<td>0.4</td>
</tr>
<tr>
<td>Mixed rejection (%)</td>
<td>16 (38)</td>
<td>15 (37.5)</td>
<td>1</td>
</tr>
<tr>
<td>C4d+ (%)</td>
<td>29 (69)</td>
<td>22 (55)</td>
<td>0.2</td>
</tr>
<tr>
<td>Diffuse C4d+ (%)</td>
<td>24 (57)</td>
<td>18 (45)</td>
<td>0.3</td>
</tr>
<tr>
<td>Focal C4d+ (%)</td>
<td>5 (12)</td>
<td>4 (10)</td>
<td>1</td>
</tr>
<tr>
<td>Banff IA (%)</td>
<td>13 (31)</td>
<td>13 (32.5)</td>
<td>1</td>
</tr>
<tr>
<td>Banff IB (%)</td>
<td>11 (26)</td>
<td>12 (30)</td>
<td>0.8</td>
</tr>
<tr>
<td>Banff IIA (%)</td>
<td>2 (4.3)</td>
<td>3 (7.5)</td>
<td>0.6</td>
</tr>
<tr>
<td>Banff suspicious (%)</td>
<td>4 (9.5)</td>
<td>5 (12.5)</td>
<td>0.7</td>
</tr>
<tr>
<td>i (Mean ± SE)</td>
<td>0.6 ± 0.1</td>
<td>0.4 ± 0.1</td>
<td>0.4</td>
</tr>
<tr>
<td>t (Mean ± SE)</td>
<td>1 ± 0.1</td>
<td>0.6 ± 0.1</td>
<td>0.02</td>
</tr>
<tr>
<td>g (Mean ± SE)</td>
<td>0.2 ± 0.06</td>
<td>0.2 ± 0.06</td>
<td>0.7</td>
</tr>
<tr>
<td>v (Mean ± SE)</td>
<td>0.1 ± 0.06</td>
<td>0.05 ± 0.03</td>
<td>0.3</td>
</tr>
<tr>
<td>cg (Mean ± SE)</td>
<td>0.8 ± 0.1</td>
<td>0.7 ± 0.1</td>
<td>0.3</td>
</tr>
<tr>
<td>ci (Mean ± SE)</td>
<td>0.8 ± 0.1</td>
<td>0.7 ± 0.1</td>
<td>0.6</td>
</tr>
<tr>
<td>ct (Mean ± SE)</td>
<td>0.7 ± 0.1</td>
<td>0.6 ± 0.1</td>
<td>0.7</td>
</tr>
<tr>
<td>cv (Mean ± SE)</td>
<td>0.7 ± 0.1</td>
<td>0.7 ± 0.1</td>
<td>0.9</td>
</tr>
<tr>
<td>TG (%)</td>
<td>10</td>
<td>3</td>
<td>0.07</td>
</tr>
</tbody>
</table>

Kidney function, ΔUPC and outcomes

There were 5 deaths and 11 graft losses in the high proteinuria group compared to two deaths and six graft losses in the low proteinuria group (P = 0.002 for the combined endpoint by log-rank test) (Table 3). There was no significant difference in death rates (5 vs. 2, P = 0.07). Likewise, there were no significant differences in Scr, eGFR and baseline UPC levels between the two groups. However, proteinuria at the time of biopsy and ΔUPC were significantly greater in the high proteinuria group.

We performed univariate regression analyses to determine factors associated with death-censored graft loss in the entire cohort (Table 4a). All variables from Tables 1 and 2 including TG were included in these analyses. In addition, we used ΔUPC > 200 mg/g (the median ΔUPC) as a categorical variable. The following variables were associated with a significant increase in death-censored allograft loss: ΔUPC > 200, Scr at baseline, cg score, deceased donor type, sirolimus use 1 month after rejection, plasmapheresis and re-transplant status (Table 4a). Of all these, stepwise Cox regression analyses retained ΔUPC >200, sirolimus use and re-transplant status as independent factors associated with poor allograft outcomes (we omitted ΔUPC as a continuous variable from the multivariable analyses). Similar analyses were performed for the combined endpoint of death or graft loss and showed that ΔUPC >200, deceased donor, sirolimus use 1 month after rejection, plasmapheresis and re-transplant status were significantly associated with poor outcomes (Table 4b).

We then examined outcomes in patients with high vs. low proteinuria by combining Kaplan–Meier and log–rank analyses (survival time from transplantation) (Figure 2). These studies confirmed that death-censored graft loss was significantly greater in patients with ΔUPC >200 mg/g (Hazard ratio (HR) 3.6, 95% CI 1.6 to 10.0, P = 0.01). Similarly, the combined end point of graft loss or death was more prevalent in patients with high proteinuria (HR 2.4, 95% CI 1.05 to 5.40, P = 0.03). Differences in patient survival alone did not reach statistical significance (data not shown).
Discussion

We demonstrate that an increase in proteinuria greater than 200 mg/g 1 month after late rejection is associated with a 3-fold greater risk of graft failure. The composite end point of graft loss or patient death was also significantly worse in patients with ΔUPC ≥200 mg/g. Differences in patient survival alone did not reach statistical significance. These findings are in agreement with observations by Meier-Kriesche et al. who showed that patients whose Scr levels did not return to baseline after rejection had worse allograft outcomes [1]. In our study, ΔUPC ≥200 mg/g was
a stronger predictor of outcomes than baseline Scr or Scr 1 month after biopsy. It seems therefore that partial recovery of kidney function after injury, assessed by either proteinuria (our study) or Scr levels [1], has a negative effect of allograft outcomes.

Negative effects of proteinuria on graft and patient outcomes have been reported elsewhere [5, 7, 8]. For example, 1 and 3-month proteinuria were powerful, independent predictors of graft loss in 484 renal transplant recipients from France [5]. In another retrospective study, proteinuria at 1 year after transplantation (both as a categorical and continuous variable) was an important and independent variable influencing patient and graft outcomes [7]. Likewise, the Spanish Chronic Allograft Nephropathy Study showed that proteinuria after the first year of transplantation was a marker of poor long-term allograft prognosis and an independent risk factor for total and cardiovascular mortality [8]. Our study adds further light to these data as we show that mild increases in proteinuria after rejection are associated with poor allograft outcomes. The type of rejection was not a significant determinant of long-term outcomes similar to the study by Amer et al. [4].

We also showed that re-transplant status and sirolimus use 1 month after rejection were associated with poor graft survival. These findings confirm previous reports in which sirolimus use was associated with higher prevalence of proteinuria and higher protein excretion [4, 13, 14]. The exact mechanisms of sirolimus-induced proteinuria are unknown yet both conversion and de novo use of sirolimus can cause proteinuria [4, 13, 14]. In addition, sirolimus may be a less potent immunosuppressant compared to calcineurin inhibitors. Results from the ELITE-SYMPhONY trial support this contention [15]. The regimen of daclizumab, mycophenolate and corticosteroids with low-dose tacrolimus resulted in better renal function, allograft survival and acute rejection rates, while low-dose sirolimus was associated worst graft outcomes and highest adverse event rates [15].

Mean time to rejection was 19 months in our study while most acute rejection episodes occur in the first 3 months after transplant. This difference results most likely from two factors related to selection criteria. Firstly, the design of the study was such that each patient had to have a biopsy proven diagnosis of rejection or CNI toxicity with measured UPC at baseline, time of biopsy and 1 month after biopsy. Unfortunately, UPC levels were not routinely monitored early post-transplant, which prevented the selection of those patients with acute rejection who had no data on proteinuria. Secondly, a significant proportion of our patients received Alemtuzumab induction. These patients may present with late antibody-mediated rejection. Nevertheless, we demonstrate that small increases in proteinuria after late rejection have negative prognostic implications for allograft survival. The causal relationship between proteinuria and poor outcomes in kidney transplant recipients remains unknown. There is conflicting evidence regarding the effects of ACE-I and angiotensin II blockade on long-term graft and patient outcomes [16-18]. The ongoing Canadian ACE-I trial (ISRCTN-78129473) and Angiotensin II Blockade for the Prevention of Crotal Interstitial Expansion and Graft Loss in Kidney Transplant Recipients (NCT00067990) studies will provide clinically meaningful evidence regarding these questions. Meanwhile, if tolerated, it may be justifiable to use RAS blockade strategies after acute rejection in patients with proteinuria.

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Author contribution to the work:
* Participated in research design: BLM, MS, AD
* Participated in the writing of the paper: BLM, AD
* Participated in the performance of the research: BLM, MS, JT, JP, AD
* Participated in data analysis: BLM, MS, JT, JP, AD

Conflicts of interest statement. None declared.

References
13. van den Akker JM, Wetzels JF, Hoitsma AJ. Proteinuria following transplantation affects not only graft survival but also patient survival. Transplantation 2001; 72: 438–444


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**The histological development of acute antibody-mediated rejection in HLA antibody-incompatible renal transplantation**

Rob Higgins¹, Daniel Zehnder¹,², Klaus Chen¹, David Lowe³, Joanna McKinnell¹, For T. Lam¹, Habib Kashi¹, Lam Chin Tan¹, Chris Imray¹, Simon Fletcher¹, Nithya Krishnan¹, Rizwan Hamer¹,² and David Briggs³

¹Transplant Unit, University Hospitals Coventry and Warwickshire, Coventry, West Midlands, UK, ²Clinical Sciences Research Institute, Warwick Medical School, University of Warwick, Coventry, West Midlands, UK and ³Histocompatibility Laboratory, National Blood and Transplant, Birmingham, West Midlands, UK

*Correspondence and offprint requests to: Rob Higgins; E-mail: Robert.Higgins@uhcw.nhs.uk*

**Abstract**

**Background.** The aim of this study was to examine the development of acute antibody-mediated rejection in HLA antibody-incompatible renal transplantation in relation to the Banff 07 histological classification.

**Methods.** Renal biopsies were scored using the Banff 07 diagnostic criteria, and paraffin-embedded sections were stained with the pan-leucocyte marker CD45.

**Results.** Thirty-six patients had 72 renal biopsies. In biopsies performed 30 min after graft reperfusion, the mean number of CD45+ cells per glomerulus was higher than in control grafts (*P*<0.04) and was associated with the donor-specific antibody (DSA) level at transplantation measured by microbeads (*P*<0.01), and eight out of nine patients with greater than five CD45+ cells per glomerulus had early post-transplant rejection or oliguria, compared to 11 out of 20 with less than five cells per glomerulus (*P*<0.01). In the first 10 days post- transplant, although peritubular capillary (PTC) leucocyte margination grade 3 and C4d deposition were specific for rejection, their sensitivities were low. PTC C4d staining was only seen in two out of 11 biopsies taken in the first 5 days after transplant, even in the presence of rejection, but was present in the majority of later biopsies with rejection. In biopsies stained for CD3, CD68 and CD20, it was notable that CD20+ cells were not seen during acute rejection, the infiltrates comprising CD3+ and CD68+ leucocytes.

**Conclusions.** Glomerular margination of leucocytes occurred early after transplantation and was associated with DSA level and early graft dysfunction. The Banff 07 PTC margination scoring system was easy to apply, especially when CD45 staining was used, and PTC margination grade 3 was always associated with clinical rejection.

**Keywords:** antibody-incompatible transplantation; CD20; complement C4d; HLA antibodies; renal allograft histology

**Introduction**

Previous reports of the histological appearances of acute antibody-mediated renal allograft rejection (AMR) have been reviewed in detail [1,2]. The 1997 (updated 2003) and 2007 Banff classifications of renal allograft pathology define criteria for histological diagnosis [3,4]. The key feature of acute AMR is margination of neutrophils and mononuclear leucocytes into glomeruli and peritubular capillaries (PTC). There is also diffuse and circumferential staining for the complement component C4d on PTC. These appearances contrast with those seen in acute T cell-mediated rejection where there is tubulitis associated with a cellular infiltrate largely made up of cells staining for CD68 or CD3 [5].

Grafts that have failed from hyperacute rejection have cortical necrosis and widespread thromboses in the microvascularity. A 1968 study found that the earliest feature predictive of hyperacute rejection was neutrophil margination into glomeruli, four or more neutrophils per sectioned glomerulus in a biopsy taken after reperfusion [6].