Protocol Biopsy in Kidney Transplant Recipients

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Has no real or apparent conflicts of interest to report.
Long term graft survival has not improved despite falling acute rejection rates

Acute Rejection (AR) %

Relative Risk of Graft Loss

Chronic allograft nephropathy (CAN) is second only to “death with functioning graft” in causes of late graft loss. (Pascual; NEJM 2003, vol 346: 580-90).

Nankivell, N Engl J Med 2003;349:2326-33
Interstitial Fibrosis/Tubular Atrophy (IF/TA) (CAN) is a histological result of immune and non-immune processes.

- **Immunologic**
  - Acute Rejection
  - Subclinical Rejection
  - Chronic Rejection

- **Non-immunologic**
  - Poor Donor Graft Quality
  - Hypertension
  - DGF
  - Chronic CNI Toxicity
A quick review of the Banff classification of renal allograft biopsies

- **T-cell mediated rejection - ACUTE:**
  - IA: >25% interstitial infiltration, moderate tubulitis (i2, t2)
  - IB: >25% interstitial infiltration, severe tubulitis
  - IIA and IIB: mild – severe intimal arteritis

- **Chronic Allograft Nephropathy (CAN or IF/TA):**
  - I: mild <25% cortical area affected
  - II: moderate 26-50% cortical area affected
  - III: severe >50% cortical area affected

- **Borderline Changes:** “Suspicious” for acute T-cell mediated rejection (10-24% interstitial infiltrate).

- **Subclinical Rejection:** Tubulointerstitial mononuclear infiltration identified from a biopsy specimen, without concurrent functional deterioration (Creat within 10-20% of baseline).

Solez et al, AJT 2008; 8: 753-60
To look for subclinical rejection (SCR) or not: important questions

- Does SCR detected by protocol biopsy impact graft outcome?
- Can treating SCR detected by protocol biopsy impact graft outcome?
- How often is SCR found on protocol biopsy?
Nankivell’s study shed light on the significance of SCR in protocol biopsies

- 120 SPK recipients from 1987 to 2001
- Large variation in immunosuppression depending on era (CsA, AzA, Tac, MMF, pred)
- Protocol biopsies done 1 and 2 wk, 1, 3, 6, and 12 mo, then yearly x 10 yrs
- SCR present in 34% of all (959) biopsy specimens

Nankivell et al; N Engl J Med 2003;349:2326-33
Nankivell et al; Transplantation 2004;78: 242–249
SCR leads to progression of IF/TA and reduced GFR

Nankivell et al; *Transplantation* 2004;78: 242–249

Protocol biopsy performed at day 14 on those with stable graft function.

Immunosuppression included pred + CsA (n=274) or Tac (n=30 since 1998), MMF (n=50 since 2000).

Clinical AR in 33%. SCR or borderline rejection diagnosed in 50%, SCR in 13%
SCR diagnosed 14 days after transplant is associated with worse graft survival

(SCR)

Choi et al. AJT 2005; 5: 1354–1360
Graft survival has been worse in patients with combined IF/TA and SCR

Moreso et al. AJT 2006; 6: 747-752
Cosio et al. AJT 2005; 5: 2464–2472
Does treating SCR seen in protocol biopsies effect outcome?

- 72 renal transplant recipients (11 LRD and 61 cadaveric) 1992 – 1995 randomized to protocol biopsy group (1, 2, 3, 6, 12 mo) or control group (biopsy at 6 and 12 mo only).
- Immunosuppression = CsA, Aza, pred. OKT3 as induction for 12 ptns.
- All rejection (SCR or clinical) treated with pulse steroids.
- Primary endpoint: 50% reduction in acute or chronic pathology at 6 month biopsy

Less acute rejection and IF/TA in the protocol biopsy group

<table>
<thead>
<tr>
<th>Clinical AR</th>
<th>Biopsy (n=36)</th>
<th>Control (n=36)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mo 2-3</td>
<td>41%</td>
<td>69%</td>
<td>.02</td>
</tr>
<tr>
<td>Mo 7-12</td>
<td>11%</td>
<td>33%</td>
<td>.03</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th></th>
<th>Biopsy</th>
<th>Control</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>SCR 1 mo</td>
<td>43%</td>
<td>15%</td>
<td>.09</td>
</tr>
<tr>
<td>2 mo</td>
<td>32%</td>
<td>26%</td>
<td></td>
</tr>
<tr>
<td>3 mo</td>
<td>27%</td>
<td></td>
<td></td>
</tr>
<tr>
<td>6 mo</td>
<td></td>
<td>26%</td>
<td></td>
</tr>
<tr>
<td>IF/TA 1 mo</td>
<td>0%</td>
<td>6%</td>
<td>.05</td>
</tr>
<tr>
<td>2 mo</td>
<td>0%</td>
<td>24%</td>
<td></td>
</tr>
<tr>
<td>3 mo</td>
<td>3%</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Creatinine at 1 year lower in protocol biopsy group

Protocol biopsies tested in a prospectively in living-related transplants

- 102 low-risk recipients of living-related donor kidneys from 2004-2005 randomized to biopsy group 1 (3 and 6 mo) or no protocol biopsies (group 2).
- Primary endpoint 6 and 12 month creatinine
- All rejection (SCR and clinical) treated with pulse steroids
- SCR seen in 17% at 1 mo, 12% at 3 mo.

Kurtkoti et al. AJT 2008; 8: 317–323
# Majority of patients on CsA-based immunosuppression

## Baseline Immunosuppression

<table>
<thead>
<tr>
<th></th>
<th>Biopsy (n=52)</th>
<th>Control (n=50)</th>
</tr>
</thead>
<tbody>
<tr>
<td>CsA</td>
<td>50</td>
<td>46</td>
</tr>
<tr>
<td>Tac</td>
<td>2</td>
<td>4</td>
</tr>
<tr>
<td>MMF</td>
<td>33</td>
<td>22</td>
</tr>
<tr>
<td>Aza</td>
<td>19</td>
<td>28</td>
</tr>
<tr>
<td>Clinical AR (1yr)</td>
<td>8</td>
<td>9</td>
</tr>
</tbody>
</table>

Kurktoti et al. *AJT* 2008; 8: 317–323
Improved creatinine and GFR in biopsy group

Creatinine (mg/dl)

<table>
<thead>
<tr>
<th>Time</th>
<th>Group I</th>
<th>Group II</th>
<th>p-Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 month</td>
<td>1.18 ± 0.21</td>
<td>1.19 ± 0.24</td>
<td>0.82</td>
</tr>
<tr>
<td>6 months</td>
<td>1.28 ± 0.33</td>
<td>1.55 ± 0.39</td>
<td>0.0003</td>
</tr>
<tr>
<td>12 months</td>
<td>1.20 ± 0.33</td>
<td>1.52 ± 0.41</td>
<td>&lt;0.0001</td>
</tr>
</tbody>
</table>
Prospective study of protocol biopsies with Tac/MMF/pred maintenance

- Biopsy arm: 1, 2, 3, 6 months. Control arm: 6 mo only.
- All patients received Tac/MMF/pred maintenance immunosuppression, no data on induction.
- All rejection (SCR and clinical) treated with 2 week tapering course of oral pred (200mg).

Rush et al. AJT 2007; 7: 2538–2545
No difference between groups at 6 mo with Tac/MMF regimens – very low rejection %

- IF/TA (ci + ct score) same in both groups (prim endpoint).

<table>
<thead>
<tr>
<th></th>
<th>Biopsy Arm</th>
<th>Control Arm</th>
<th>P</th>
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<tbody>
<tr>
<td>Creatinine (mmol/L)</td>
<td>119</td>
<td>124</td>
<td>.84</td>
</tr>
<tr>
<td>GFR (ml/min)</td>
<td>73</td>
<td>69</td>
<td>.18</td>
</tr>
<tr>
<td>SCR (at 6 mo)</td>
<td>9%</td>
<td>6%</td>
<td>.48</td>
</tr>
<tr>
<td>Clinical AR (first 6 mo)</td>
<td>9%</td>
<td>7.5%</td>
<td>.44</td>
</tr>
</tbody>
</table>

Rush et al. AJT 2007; 7: 2538–2545
## Prevalence of SCR found on protocol biopsy is variable

<table>
<thead>
<tr>
<th>Study</th>
<th>Induction</th>
<th>Immunosuppression</th>
<th>Bx schedule</th>
<th>Prev of SCR (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nankivell ’87-‘00</td>
<td>Rare</td>
<td>Various (CsA, Tac, MMF, AZA, Pred)</td>
<td>3mo, 6-12mo, 2-5yrs, 6-10yrs</td>
<td>42, 37, 20, 12</td>
</tr>
<tr>
<td>Gloor ’98-’00</td>
<td>About 1/2</td>
<td>Tac + MMF + Pred</td>
<td>3mo</td>
<td>2.6</td>
</tr>
<tr>
<td>Rush ‘92–’95</td>
<td>Rare</td>
<td>CsA (sandimmune) + Aza + Pred</td>
<td>1, 2, 3, 6mo</td>
<td>43, 32, 27, 15</td>
</tr>
<tr>
<td>Nickerson ‘96-’98</td>
<td>Rare</td>
<td>CsA (Neoral) + MMF + Pred</td>
<td>1, 2, 3, 6mo</td>
<td>38, 25, 31, 25</td>
</tr>
<tr>
<td>Cosio ’98–’01</td>
<td>Thymo 67%</td>
<td>Tac + MMF + Pred (72%). CsA (10%), Rapa (18%).</td>
<td>12mo</td>
<td>5</td>
</tr>
<tr>
<td>Choi ‘93-’03</td>
<td>None</td>
<td>CsA + Pred (most)</td>
<td>2 weeks</td>
<td>13</td>
</tr>
<tr>
<td>Moreso ‘88-’03</td>
<td>About 1/3</td>
<td>Various (CsA, Tac, MMF, AZA, Pred, Rapa)</td>
<td>1-6mo</td>
<td>Tac: 16%; CsA: 32%; CNI free: 56%</td>
</tr>
<tr>
<td>Rush ‘01-’04</td>
<td>None</td>
<td>Tac + MMF + Pred</td>
<td>1, 2, 3, 6mo</td>
<td>4.6 overall</td>
</tr>
<tr>
<td>Kurtkoti ‘04-’05</td>
<td>Rare</td>
<td>CsA&gt;Tac, + MMF or Aza, + pred</td>
<td>1, 3mo</td>
<td>17, 12</td>
</tr>
<tr>
<td>Heilman ‘03-’08</td>
<td>All (mixed)</td>
<td>Tac + MMF (pred-free)</td>
<td>1, 4mo</td>
<td>27 (total)</td>
</tr>
<tr>
<td>Yango ‘04-’05</td>
<td>All (Thymo)</td>
<td>Tac + MMF + Pred</td>
<td>6mo</td>
<td>0</td>
</tr>
</tbody>
</table>
### Baseline immunosuppression effects histology seen in protocol biopsies in Nankivell’s SPK study

<table>
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<tr>
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</thead>
<tbody>
<tr>
<td>Patients (n)</td>
<td>61</td>
<td>13</td>
<td>25</td>
<td>21</td>
</tr>
<tr>
<td>Immunosuppression</td>
<td>CsA(s), Aza, pred</td>
<td>CsA(me), Aza, pred</td>
<td>CsA(me), MMF, Pred</td>
<td>Tac, MMF, Pred</td>
</tr>
<tr>
<td>SCR @ 3mo</td>
<td>63%</td>
<td>33%</td>
<td>24%</td>
<td>4.7%</td>
</tr>
</tbody>
</table>

Nankivell et al; *Transplantation* 2004;78: 242–249
Can we identify patients at higher risk for SCR?

- **LNRD vs. LRD transplant, 2 HLA-DR mismatch, previous AR** (Cosio et al. AJT 2005; 5: 2464–2472)
- **Donor age >40, PRA >10%, CNI free regimen** (Moreso et al. AJT 2006; 6: 747-52)
- **Re-transplantation** (Seron et al. KI 2007; 72: 690-97)
- **Steroid free** (Heilman et al. AJT 2010)
- **DGF** (Qureshi et al. Transplantation 2002; 74:1400-1404)
Limitations to protocol biopsies

- **Complications:**
  - Furness et al: 2127 protocol biopsies in 5 centers: no death, 1 graft loss (.04%), 6 hemorrhage requiring procedure or transfusion (.28%) (Transplantation 2003 Vol. 76, 969–973).
  - Schwarz et al: 1171 protocol biopsies at single center: hematuria 3.5%, hematoma 2.5%, requiring procedure or transfusion 1%, no graft loss (AJT 2005; 5: 1992–1996).

- **“Sample effect”:** rejection and fibrosis are patchy processes. Sorof et al. reported on 70 transplant biopsy sample pairs examined independently by pathologists: AR grade differed by ≥1 Banff grade in 30% of cases unblinded, and 50% of cases blinded (Transplantation 1995; 60: 1215).

- Cost and availability of clinical resources.
- Inconvenience to patients, especially those that commute long distances to the transplant center.
A quick word about chronic lesions in protocol biopsies

- Chronic histologic damage is seen frequently in allografts, even early after transplant (25-30% at 3mo).
- Degree of IF/TA is predictive of graft function and survival, and does not appear to vary with CNI (Cosio et al. AJT 2005; 5: 2464-2472).
- CNI-free protocols have seen less chronic damage:
  - CNI avoidance: Flechner et al: 61 patients randomized to SRL vs. CsA, + MMF and steroids, after KTx. 48 had protocol biopsy at 2yrs:
  - CNI withdrawal: RMR trial: 547 KTx recipients on CsA, SRL, pred randomized to either stop or continue CsA at 3mo. 1yr protocol biopsy in a subset of 96 showed significantly less chronic damage in the withdrawal arm.
Summary

- SCR found on protocol biopsy can lead to fibrosis and atrophy, worse graft outcomes.
- Prevalence of SCR has been widely variable in the literature and depends on immunosuppression, timing of biopsy, histologic definition.
- In the pre-”tacrolimus/MMF/pred” era, treatment of SCR found on protocol biopsy led to improved outcomes compared to a control group, but may not be as beneficial for all patients in the modern era where SCR rates are low.
- Protocol biopsy may be useful for patients with risk factors for SCR, and for identifying those at risk for graft loss due to IF/TA.
Thank You