Pathology and Management of Chronic Allograft Dysfunction

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Mission Impossible?
PLAN

• To review the description of chronic allograft dysfunction (NOT “CAN” or “chronic rejection”)
• To review the “Banff 2007 Update”
• To review the causes of chronic allograft dysfunction
• To review the potential treatment modalities of chronic allograft dysfunction
Case Discussion

- 21 year old African American woman
- ESRD due to lupus nephritis (class V)
- Living donor transplant
- Microscopic hematuria, proteinuria (2.7 g/day) and elevated serum creatinine (2.5 mg/dl) six years after transplantation
- **Kidney biopsy**: Focal proliferative GN with crescents and immune complexes (WHO class III, consistent with SLE)
Case Discussion

- Steroid pulses (250 mg x 3), prednisone 60 mg/d for one month with taper over the next 3 months
- Severe herpes esophagitis and CMV infection
- ACE inhibitor, good blood pressure control, on 1000 mg twice a day of mycophenolate mofetil
- Returned to dialysis 8 years after transplantation-2 years after the biopsy
Kidney Transplantation

- The number of patients on the waiting list for a deceased donor (DD) kidney transplant: 73,555-competing for approximately 11,200 kidneys

  *USRDS 2009 Annual Data Report*

- Shortage of kidneys suitable for transplantation
- Waiting times are quite long (4-7 years)
- **Need to maximize graft survival**
Chronic Allograft Dysfunction

- Progressive graft failure with slowly rising serum creatinine and decreasing GFR
- End-stage kidney disease from a variety of insults to the graft
- Independent of acute rejection
- Variable degrees of hypertension and proteinuria
- Features of chronic allograft nephropathy: vascular intimal hyperplasia, interstitial fibrosis and tubular atrophy
Causes of Allograft Injury

- Immunologic (Antigen-dependent)
  - Cellular immunity
    - Inadequate immunosuppression/noncompliance
  - Humoral immunity
  - Acute rejection
  - HLA-matching
  - Donor-specific antibodies (DSA)

- Infections
  - Cytomegalovirus (CMV)
  - BK virus
Causes of Allograft Injury

- Nonimmunologic (Antigen-independent)
  - Organ viability
    - Living vs deceased
    - Donor age
    - Brain death
    - **Prolonged cold ischemia time**
    - Ischemia-reperfusion injuries
    - **Delayed graft function** / acute tubular necrosis

- Recipient-related factors
  - **Hypertension**
  - Hyperlipidemia
  - Compliance
  - Obstruction
  - **Recurrent disease**

- Treatment- **nephrotoxicity due to CNIs**
Banff 2007 Update

1. Normal
2. Antibody-mediated changes
   - C4d deposition without morphologic evidence of active rejection
   - Acute antibody-mediated rejection
   - Chronic active antibody-mediated rejection
3. Borderline changes: “suspicious” for acute T-cell-mediated rejection
4. T-cell-mediated rejection
   - Acute T-cell-mediated rejection
   - Chronic active T-cell-mediated rejection
5. Interstitial fibrosis and tubular atrophy (IF/TA)
6. Other: Changes not considered to be due to rejection

Solez K, et al. AJT 2008
Mechanisms of Injury

• Acute cellular or acute humoral rejection: might leave residual injury predisposing to chronic injury
• Chronic humoral rejection with the presence of C4d as well as reduction in regulatory T-cell (Treg) numbers, and inhibition of their function
Table 1: Morphology of specific chronic diseases

<table>
<thead>
<tr>
<th>Etiology</th>
<th>Causes of IF/TA (non-rejection)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chronic hypertension</td>
<td>Arterial/fibrointimal thickening with reduplication of elastica, usually with small artery and arteriolar hyaline changes.</td>
</tr>
<tr>
<td>CNI&lt;sup&gt;1&lt;/sup&gt; toxicity</td>
<td>Arteriolar hyalinosis with peripheral hyaline nodules and/or progressive increase in the absence of hypertension or diabetes. Tubular cell injury with isometric vacuolization.</td>
</tr>
<tr>
<td>Chronic obstruction</td>
<td>Marked tubular dilation. Large Tamm–Horsfall protein casts with extravasation into interstitium, and/or lymphatics.</td>
</tr>
<tr>
<td>Bacterial pyelonephritis</td>
<td>Intratubular and peritubular neutrophils, lymphoid follicle formation.</td>
</tr>
<tr>
<td>Viral infection</td>
<td>Viral inclusions on histology and immunohistology and/or electron microscopy.</td>
</tr>
</tbody>
</table>

Glomerular Abnormalities
- Transplant Glomerulopathy
- Recurrent Disease
- Glomerulosclerosis

Tubular Atrophy
- Ischaemic-Reperfusion injury
- Acute severe rejection
- Subclinical rejection
- Calcineurin toxicity
- Polyoma virus

Interstitial Fibrosis
- Ischaemic-Reperfusion injury
- Subclinical rejection
- Calcineurin toxicity

Fibro intimal Hyperplasia
- True Chronic Rejection
- Donor Disease
- Hypertension

Arteriolar Hyalinosis
- Calcineurin toxicity
  (diabetes mellitus)

Peritubular Capillary
- C4d deposition and multilamination – chronic immune mediated

Transplant Vasculopathy (TV)

Artery from a vehicle-treated allograft

Artery with TV; NI: neointima; arrowhead: elastin

• Renal allograft biopsy (*silver staining*)

• Evidence of “double contours” in capillary loops

• Mesangial proliferation and matrix expansion and basement membrane thickening

• Renal allograft biopsy with C4d deposition (*in brown*) in peritubular capillaries consistent with antibody-mediated rejection

The Natural History of Chronic Allograft Nephropathy

- A prospective study of 120 recipients with type 1 diabetes, all but 1 of whom had received kidney–pancreas transplants (1987-2000)
- 961 kidney-transplant–biopsy specimens taken regularly from the time of transplantation to 10 years thereafter

## Table 1. Characteristics of the Allograft at and after Transplantation.*

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>At Transplantation† (N=135)</th>
<th>3 Mo (N=138)</th>
<th>6–12 Mo (N=188)</th>
<th>2–5 Yr (N=223)</th>
<th>6–10 Yr (N=81)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Banff score‡</td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>Chronic interstitial fibrosis</td>
<td>0.09±0.24</td>
<td>0.70±0.53</td>
<td>1.07±0.56</td>
<td>1.34±0.67</td>
<td>1.64±0.74</td>
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<tr>
<td>Tubular atrophy</td>
<td>0.06±0.20</td>
<td>0.56±0.51</td>
<td>0.99±0.52</td>
<td>1.26±0.67</td>
<td>1.57±0.76</td>
</tr>
<tr>
<td>Fibrointimal thickening</td>
<td>0.03±0.16</td>
<td>0.11±0.30</td>
<td>0.17±0.38</td>
<td>0.31±0.51</td>
<td>0.33±0.51</td>
</tr>
<tr>
<td>Chronic glomerulopathy</td>
<td>0.0±0.04</td>
<td>0.08±0.10</td>
<td>0.08±0.26</td>
<td>0.12±0.30</td>
<td>0.24±0.48</td>
</tr>
<tr>
<td>Mesangial matrix (mm)</td>
<td>0.09±0.29</td>
<td>0.18±0.41</td>
<td>0.31±0.41</td>
<td>0.44±0.48</td>
<td>0.62±0.57</td>
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<tr>
<td>Arteriolar hyalinosis</td>
<td>0.16±0.35</td>
<td>0.29±0.48</td>
<td>0.39±0.54</td>
<td>0.72±0.71</td>
<td>1.22±0.83</td>
</tr>
<tr>
<td>Sclerosed glomeruli (%)</td>
<td>1.7±4.3</td>
<td>2.3±6.2</td>
<td>2.1±4.9</td>
<td>14.1±18.1</td>
<td>37.2±21.9</td>
</tr>
<tr>
<td>Subclinical rejection (%)</td>
<td>NA</td>
<td>41.8</td>
<td>36.8</td>
<td>19.5</td>
<td>12.3</td>
</tr>
<tr>
<td>Isotopic glomerular filtration rate (ml/min)</td>
<td>NA</td>
<td>59.3±16.8</td>
<td>60.7±17.0</td>
<td>54.7±19.8</td>
<td>50.2±27.2</td>
</tr>
<tr>
<td>Serum creatinine (mg/dl)</td>
<td>NA</td>
<td>1.48±0.61</td>
<td>1.45±0.33</td>
<td>1.56±0.55</td>
<td>1.62±0.48</td>
</tr>
</tbody>
</table>

* Plus–minus values are means ± SD. The numbers are the numbers of biopsy specimens. To convert values for serum creatinine to micromoles per liter, multiply by 88.4. NA denotes not applicable.
† Samples were obtained up to one week after transplantation.
‡ Banff scores range from 0 to 3, with higher scores indicating more severe abnormalities.
Relevant Donor Abnormalities

- Advanced donor age
- Pre-existing disease or injury to the donor: glomerulosclerosis (>20%), microvascular disease
- HLA-mismatch
- Prolonged cold ischemia time
- Living vs deceased donors: ischemia-reperfusion injury
- Using time-zero biopsies: might be very helpful to assess subsequent biopsies
*Delayed graft function (DGF): major predictor of graft failure overall with cold ischemia time (CIT) as an important independent factor

*Prolonged CIT, directly and independently of DGF and AR, compromises the long-term graft survival

HLA-Specific Antibodies Developed in the First Year Posttransplant are Predictive of Chronic Rejection and Renal Graft Loss

Lee, Po-Chang; Zhu, Lan; Terasaki, Paul I.; Everly, Matthew J.

Transplantation 2009

• Retrospective case-controlled study from Taiwan
• 278 patients, transplanted between 1991-2004
• 25 patients with failed graft (230 serum samples) and 25 patients with a functioning graft (305 serum samples)
• HLA antibody development within 1-year posttransplant markedly lowers allograft survival

Lee PC, et al. Transplantation 2009
Hypertension after Kidney Transplantation

• Very common
• Not well controlled-despite multiple antihypertensive medications
• Independent risk factor for graft failure and mortality
• ACE inhibitor use (from 1993 to 2002): 25% of patients were treated in the first year after transplantation

Improved Long-Term Outcomes with Blood Pressure Control

• 24,404 patients transplanted between 1987 and 2000-Collaborative Study Database

• Patients whose SBP was $>140$ mmHg at 1 year posttransplant but controlled to $\leq 140$ mmHg by 3 years had significantly improved long-term graft outcome compared with patients with sustained high SBP to 3 years

• At 5 years: SBP lowering after year 3 was associated with improved 10-year graft survival

Figure 2: Long-term kidney graft survival calculated from 3 to 10 years in relation to systolic blood pressure of <140 or \( \geq 140 \) mmHg at 1 and 3 years posttransplant. The respective 1- and 3-year pressures are indicated to the right of each curve, together with numbers of patients studied.

Figure 4: Relationship of blood pressure profiles examined over 5 years on kidney graft survival from 5–10 years. Systolic blood pressures \( >140 \) or \( \leq 140 \) mmHg at 1, 3 and 5 years posttransplant are indicated for each patient group studied. All patients in this analysis had a SBP of \( >140 \) mmHg 1 year posttransplant.

Antihypertensives for Kidney Transplant Recipients

- Meta-analysis of randomized controlled trials
- 60 trials, enrolling 3802 recipients
- Twenty-nine trials (2262 patients) compared calcium channel blockers (CCB) with placebo or no treatment
- 10 trials (445 patients) compared ACEi with placebo or no treatment
- 7 studies (405 patients) compared CCB with ACEi
- In direct comparison with CCB, ACEi decreased GFR, proteinuria, hemoglobin, and increased hyperkalemia
- Graft loss data were inconclusive

Cross AN, et al. Transplantation 2009
**A**

<table>
<thead>
<tr>
<th>Study name</th>
<th>ACE/ARB</th>
<th>Control</th>
<th>Mean Difference (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Curtis</td>
<td>18</td>
<td>18</td>
<td>-5.0 (-16.0, 6.0)</td>
</tr>
<tr>
<td>Mourad</td>
<td>16</td>
<td>15</td>
<td>-9.0 (-22.5, 4.5)</td>
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<tr>
<td>Hernandez E</td>
<td>10</td>
<td>11</td>
<td>-9.0 (-30.1, 12.1)</td>
</tr>
<tr>
<td>Beckingham</td>
<td>15</td>
<td>10</td>
<td>-5.8 (-23.0, 11.4)</td>
</tr>
<tr>
<td>Sonnoesaal</td>
<td>10</td>
<td>10</td>
<td>-6.7 (-18.0, 5.5)</td>
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<td>Van der Schaft</td>
<td>20</td>
<td>20</td>
<td>-8.0 (-19.6, 3.6)</td>
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<tr>
<td>Cleciura</td>
<td>43</td>
<td>32</td>
<td>-1.7 (-10.1, 6.7)</td>
</tr>
<tr>
<td>Suwelack</td>
<td>48</td>
<td>48</td>
<td>1.1 (-11.2, 13.4)</td>
</tr>
<tr>
<td>Inigo</td>
<td>17</td>
<td>17</td>
<td>-10.0 (-30.4, 10.4)</td>
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<tr>
<td>Midvedt</td>
<td>76</td>
<td>78</td>
<td>-9.0 (-14.0, -4.0)</td>
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<tr>
<td>Martinez-Castelac</td>
<td>42</td>
<td>42</td>
<td>-2.6 (-18.8, 13.7)</td>
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<tr>
<td>Tylcki</td>
<td>14</td>
<td>14</td>
<td>-1.2 (-9.5, 7.1)</td>
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<tr>
<td>Overall fixed</td>
<td>329</td>
<td>315</td>
<td>-5.7 (-8.7, -2.8)</td>
</tr>
<tr>
<td>Overall random</td>
<td>329</td>
<td>315</td>
<td>-5.7 (-8.7, -2.8)</td>
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**B**

<table>
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<tr>
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<td>-9.0 (-22.5, 4.5)</td>
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<td>43</td>
<td>32</td>
<td>1.7 (-10.1, 6.7)</td>
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<tr>
<td>Suwelack</td>
<td>48</td>
<td>48</td>
<td>1.1 (-11.2, 13.4)</td>
</tr>
<tr>
<td>Midvedt</td>
<td>76</td>
<td>78</td>
<td>-9.0 (-14.0, -4.0)</td>
</tr>
<tr>
<td>Overall fixed</td>
<td>183</td>
<td>173</td>
<td>-6.4 (-10.3, -2.5)</td>
</tr>
<tr>
<td>Overall random</td>
<td>183</td>
<td>173</td>
<td>-5.8 (-10.5, -0.99)</td>
</tr>
</tbody>
</table>

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*Change in GFR*  
**Change in proteinuria**  

The use of ACEI/ARB therapy was associated with longer patient and graft survival after renal transplantation (2,031 patients, transplanted 1990-2003)

Risk of Long-Term Graft Loss

- 1505 patients with biopsy-proven GN from Australia (1988-1997)
- Most frequent causes of allograft loss at 10 years: 1. Chronic rejection, 2. Death with a functioning graft, 3. Recurrence
- The incidence of allograft loss due to recurrence at 10 years was 8.4% and increased overtime
- Recurrence is more frequent than acute rejection as a cause of allograft loss during first 10 years after transplant

Briganti EM, et al NEJM 2002
Briganti EM, et al NEJM 2002
Recurrent Disease (True Recurrence): Diagnosis

Biopsy proven disease on native kidney

↓↓

Posttransplant proteinuria or hematuria or elevated creatinine

↓↓

Same biopsy proven disease on kidney transplant
Recurrent Glomerular Diseases (GN)

- **Recurrence of primary GN**: FSGS, MPGN, IgA nephropathy
- **Recurrence of secondary GN**: SLE, Henoch-Schönlein, HUS/TTP, anti-GBM disease
- **Recurrence of metabolic or systemic disease**: diabetic nephropathy, amyloidosis, scleroderma, oxalosis, Fabry disease
Recurrent GN in the Transplant

• The prevalence of GN as the cause of ESRD: 10-25%, higher prevalence in children and white patients
• The prevalence of recurrent GN: 1.9%-31% in different series
• True prevalence of recurrent GN: patients who lost their grafts due to recurrence + patients who have recurrence with a functioning graft
• Cause of graft loss: 1-8.4% of all graft failures
Potential Problems for Identifying Recurrent GN in the Transplant

- Primary disease-native kidney disease-is unknown for many patients
- No unified approach for patients with urinary abnormalities and increased serum creatinine after transplantation (histological vs clinical diagnosis)
- Transplant biopsy is not routinely submitted for IF and EM examination
Potential Problems for Identifying Recurrent GN in the Transplant

- Interpretation of the biopsy: DIFFICULT, de novo vs recurrent-MPGN vs transplant glomerulopathy
- Most of the studies are small and retrospective with variable follow-up periods
- No randomized, prospective studies for different treatment regimens
Protocol Biopsies

- Processes that lead to late graft loss begin early and can be detected by protocol biopsies (1–3 months).
- Chronic tubulo-interstitial and vascular changes can be seen in one third of transplants after 1 year and at later times become nearly universal.
- Detection of abnormalities in early protocol biopsies (the presence of IF/TA) is predictive of subsequent graft function and loss.
- Biopsies at 3 months scored as Banff ci0 and cv0 have a significantly better graft survival at 5 years.
- Early treatment may have a dramatic effect on the outcome of the graft—No clear treatment options.
Treatment of Subclinical Rejection

- 72 patients: randomized to biopsies at 1, 2, 3, 6, and 12 mo (Biopsy group), or to 6- and 12-mo biopsies only (Control group)
- SCR: treated during the first 3 months with methylprednisolone boluses
- Treatment of SCR was associated with a reduced progression of IF/TA at 6 months and better graft function at 2 years
- The prevalence of SCR: 30%-patients were on cyclosporine, azathioprine, and steroids

Protocol Biopsies

- Role is not clear on managing transplant patients
- Can we identify patients who are at risk of developing graft dysfunction?
- Benefit of this approach has yet to be evaluated in large, multicenter, and prospective trials (efficacy variable in clinical trials)
- **Complications** - all within 4 hours: gross hematuria 3.5%, perirenal hematoma 2.5%, and A-V fistula with mostly spontaneous resolution 7.5%

Minimizing the Impact of CNI-induced Nephrotoxicity

• CNI avoidance: not very successful in the past
• Conversion: CNI withdrawal at 3-mo or 6-mo; conversion to MMF or sirolimus
• Minimization of CNIs/additional agents: low dose CNI with MMF/MPA ± steroids or mTOR inhibitors
Minimizing the Impact of CNI-induced Nephrotoxicity

- CNI minimization:
  - The Symphony trial: 1 year follow-up
  - 4 arm study (1589 patients):
    - Daclizumab induction, 2 g MMF, low-dose tacrolimus (target level 3-7) and steroids resulted in better renal function and lower acute rejection and graft loss rates compared with three other regimens: two with low-doses of cyclosporine or sirolimus and one with no induction and standard cyclosporine
  - Similar results at additional 2-year follow-up (958 patients)

Ekberg H, et al. AJT 2009
<table>
<thead>
<tr>
<th>End Point</th>
<th>Standard-Dose Cyclosporine (N=390)</th>
<th>Low-Dose Cyclosporine (N=399)</th>
<th>Low-Dose Tacrolimus (N=401)</th>
<th>Low-Dose Sirolimus (N=399)</th>
<th>P Value†</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Primary end point</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean calculated GFR — ml/min‡</td>
<td>57.1±25.1</td>
<td>59.4±25.1</td>
<td>65.4±27.0</td>
<td>56.7±26.9</td>
<td>&lt;0.001</td>
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<tr>
<td>P value for comparison with tacrolimus</td>
<td>&lt;0.001</td>
<td>0.001</td>
<td>Reference</td>
<td>&lt;0.001</td>
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<td><strong>Secondary end points</strong></td>
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<td></td>
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<tr>
<td>Mean measured GFR — ml/min§</td>
<td>63.5±25.4</td>
<td>65.3±26.6</td>
<td>69.6±27.9</td>
<td>64.4±28.5</td>
<td>0.04</td>
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<td>0.02</td>
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<tr>
<td>Mean calculated GFR — ml/min‡</td>
<td>46.2±23.1</td>
<td>50.2±23.1</td>
<td>54.3±23.9</td>
<td>47.5±26.1</td>
<td>&lt;0.001</td>
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<td>P value for comparison with tacrolimus</td>
<td>&lt;0.001</td>
<td>0.007</td>
<td>Reference</td>
<td>&lt;0.001</td>
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<tr>
<td><strong>Acute rejection</strong></td>
<td></td>
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<tr>
<td><strong>At 6 mo</strong></td>
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<td></td>
</tr>
<tr>
<td>Biopsy-proven (excluding borderline values) — %</td>
<td>24.0</td>
<td>21.9</td>
<td>11.3</td>
<td>35.3</td>
<td>&lt;0.001</td>
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<td>P value for comparison with tacrolimus</td>
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<td>Reference</td>
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<tr>
<td><strong>At 12 mo</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Suspected and treated — %</td>
<td>32.8</td>
<td>29.5</td>
<td>17.2</td>
<td>43.5</td>
<td>&lt;0.001</td>
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<tr>
<td>Biopsy-proven (including borderline values) — %</td>
<td>30.1</td>
<td>27.2</td>
<td>15.4</td>
<td>40.2</td>
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<td>&lt;0.001</td>
<td></td>
</tr>
</tbody>
</table>

*Ekberg H, et al. NEJM 2007*
Minimizing the Impact of CNI-induced Nephrotoxicity

- New agents: biologics (belatacept, alefacept, efalizumab) and small molecules (Janus Kinase inhibitors)
  - 218 patients: randomized to intensive or a less intensive regimen of belatacept or cyclosporine
  - All patients received induction with basiliximab, MMF, and corticosteroids
  - At six months, the incidence of acute rejection was similar among the groups: 7% for intensive belatacept, 6% for less-intensive belatacept, and 8% for cyclosporine
  - At 12 months, the GFR was significantly higher with belatacept and chronic allograft nephropathy was less common with belatacept than with cyclosporine

Belatacept Potently and Selectively Blocks T-cell Activation

- No cell division
- No cytokine production
- Anergy
- Apoptosis
Belatacept Studies

- Less diabetes, better BP control, better lipids; very few patients with DSA
- More acute rejection (up tp 22%); but better GFR at 12 months
- PTLD: 8 in MI (6 CNS), 6 in LI (3 CNS), 2 in CsA arm; most of them EBV negative
- Despite a favorable vote from FDA Advisory Committee (3/2010), FDA did not approve the use of belatacept and requested longer-term clinical data for the product
Conversion Studies

- Conversion from cyclosporine to tacrolimus-5 year data (CRAF study): significant improvement in renal function but no impact on patient or graft survival
  

- **Convert** trial: 830 patients, 6-120 mo posttx and receiving cyclosporine or tacrolimus, were randomly assigned to continue CNI (n=275) or convert to sirolimus (n=555)

- At 2 years, SRL conversion among patients with baseline GFR> 40 mL/min was associated with excellent patient and graft survival, no difference in BCAR, increased urinary protein excretion, and a lower incidence of malignancy compared with CNI continuation

  Schena FP, et al. Transplantation 2009
Mycophenolate Mofetil (MMF) versus Azathioprine (AZA)

- Systematic review of the literature and meta-analysis (1985-2007)
- Randomized controlled studies
- Direct comparison of MMF vs AZA
- 27 publications from 19 trials included

Knight SR, et al. Transplantation 2009
Mycophenolate Mofetil (MMF) versus Azathioprine (AZA)

- 3143 patients (1775 on MMF vs 1368 on AZA)
- The use of MMF significantly reduced the risk of acute rejection compared with AZA over all
- The hazard of graft loss was lower in the MMF group

Knight SR, et al. Transplantation 2009
BK Nephropathy

- Intranuclear inclusion bodies in epithelial cells and severe tubular injury
- Interstitial fibrosis
- Positive immunohistochemical staining
- Electron microscopy demonstrating viral particles
BK/JC Virus Infection

• 400 healthy blood donors aged 20-59 years were tested for BKV- and JCV-specific antibodies against virus-like particles

• IgG seroprevalence was 82% (328 of 400 donors) for BKV and 58% (231 of 400) for JCV

• Asymptomatic urinary shedding of BKV and JCV was observed in 28 (7%) and 75 (19%) of 400 subjects, respectively

BK Nephropathy

- The prevalence rate varies from 1% to 10% due to different local immunosuppression protocols and diagnostic approaches
- Pathophysiology:
  - Replicates best in uroepithelial cells, but also found in lymphoid, other tissues
  - Asymptomatic viruria/viremia
  - Ureteral ulceration, stricture & stenosis, hemorrhagic cystitis
  - Progressive loss of renal allograft function
  - Urothelial malignancy & vasculopathy
### Table 1. Diagnostic Testing for BK Virus Nephropathy

<table>
<thead>
<tr>
<th>Test</th>
<th>Threshold Value</th>
<th>Correlation With PVAN on Biopsy</th>
</tr>
</thead>
<tbody>
<tr>
<td>Decoy cells (&gt;10/cytospin)</td>
<td>&gt;10 cells/cytospin</td>
<td>+</td>
</tr>
<tr>
<td>Urine BK virus DNA quantitative PCR</td>
<td>&gt;$1 \times 10^7$ copies/mL</td>
<td>++</td>
</tr>
<tr>
<td>Blood/plasma BK virus DNA quantitative PCR</td>
<td>&gt;$1 \times 10^4$ copies/mL</td>
<td>+++</td>
</tr>
</tbody>
</table>

Abbreviations: PCR, polymerase chain reaction; PVAN, polyomavirus-associated nephropathy.

<table>
<thead>
<tr>
<th>Test</th>
<th>Sensitivity</th>
<th>Specificity</th>
<th>PPV</th>
</tr>
</thead>
<tbody>
<tr>
<td>Decoy Cells (&gt;10/cytospin)</td>
<td>25%</td>
<td>84%</td>
<td>5-20%</td>
</tr>
<tr>
<td>Viruria (&gt;10⁷ copies/ml)</td>
<td>100%</td>
<td>92%</td>
<td>31%</td>
</tr>
<tr>
<td>Viremia (&gt;10⁴ copies/ml)</td>
<td>100%</td>
<td>96%</td>
<td>50-60%</td>
</tr>
</tbody>
</table>

Wiseman AC. Am J Kid Dis 2009
Treatment of BK Nephropathy

• No specific antiviral drug treatment
• Reduction/adjustment in immunosuppression remains the cornerstone
• Cidofovir, leflunomide, quinolones, and intravenous immunoglobulin: no randomized prospective clinical trial
Impact of Preemptive Reduction of Immunosuppression

- 200 adult renal transplant recipients: randomized to tacrolimus (n = 134) or cyclosporine (n = 66)
- Urine and blood were collected weekly for 16 weeks and at months 5, 6, 9 and 12 and analyzed for BK viral load
- By 1 year, 70 patients (35%) developed viruria and 23 (11.5%) viremia; neither were affected independently by immunos used
- Viruria was highest with TAC-MMF (46%) and lowest with CsA-MMF (13%), p = 0.005

Impact of Preemptive Reduction of Immunosuppression

- Management of immunosuppression:
  - Identification of BK viremia triggered discontinuation of AZA or MMF
  - If viremia failed to clear within 4 weeks, the calcineurin inhibitor dose was tapered to trough CsA levels of 100–200 ng/mL or trough TAC levels of 3–5 ng/mL

- After reduction of immunosuppression, viremia resolved in 95%, without increased acute rejection, allograft dysfunction or graft loss

- No BK nephropathy was observed

BK virus and Pre-Emptive Immunosuppression Reduction 5-Year Results

- A retrospective 5-year review
- 5 year follow up in 97% of patients
- Viremia resolved in 95% of patients with reduction of immunosuppression
- 5-year patient survival 91% and graft survival 84%
- Immunosuppression and viremia did not influence graft survival
- Acute rejection: 12% by 5-years after transplant
- No BK nephropathy-no protocol biopsies

Hardinger KL, et al. AJT 2010
Screening and Diagnostic Testing for BK

- Linear progression is an opportunity: from viruria (30-40%) to viremia (10-20%) to nephropathy (1-10%) and graft dysfunction/loss
- Blood and/or urine samples every 3 months for first 2 years, then once a year and in the event of allograft dysfunction
- Careful reduction of immunosuppression and close follow-up for development of acute rejection
Evaluation of a Patient with Late Allograft Dysfunction

- Exclusion of obvious causes such as obstruction, dehydration, high CNI levels, uncontrolled hypertension, and UTI/urosepsis
- Urinalysis, spot urine protein/creatinine ratio, and 24-h urine collection
- BK viral load (blood/urine)
- Kidney biopsy: consider early before significant graft dysfunction
Evaluation of a Patient with Late Allograft Dysfunction

- Adequate biopsy sample to make a correct diagnosis (at least 10 glomeruli and two arteries; two cores of cortex preferably); comparison with time-zero biopsies, if possible
- Light microscopy to assess fibrosis and tubular atrophy, as well as specific stains (PAS and silver stain)
- IF to assess recurrent or de novo GN or C4d deposition
- EM to detect early transplant glomerulopathy or immune deposits
New Diagnostic Methods

• Gene and protein expression profiles (peripheral blood, urine and graft)
• Predictors of interstitial fibrosis in biopsies
  • Early phenotypic changes indicative of epithelial-to-mesenchymal transition (*de novo* vimentin expression and translocation of $\beta$-catenin into the cytoplasm of tubular cells)


• Validation of these tests in transplant patients—diagnosis of a certain disease and also specific treatment
Therapy for Chronic Allograft Dysfunction

- There is no single specific treatment; several therapies and approaches
- Early diagnosis/early intervention: IMPORTANT
- Changes in serum creatinine occur at a late stage
- Serum creatinine might underestimate deterioration in GFR
Therapy for Chronic Allograft Dysfunction

- Minimization of cold ischemia time
- Aggressive management of hypertension: BP goal <130/80 mmHg
- Management of diabetes/PTDM
- Treatment of hyperlipidemia-LDL target <100
- Reduction of proteinuria
- Treatment of infections-CMV/BK
- Treatment of recurrent diseases
Therapy for Chronic Allograft Dysfunction

- Manipulation of immunosuppression
  - More potent immunosuppressive therapy early after transplantation followed by minimization of immunosuppression, especially CNIs, to avoid CNI toxicity and BK nephropathy
  - Treatment of subclinical rejection
  - Monitoring and removal of HLA antibodies
  - Minimization or elimination of calcineurin inhibitors
  - Use of non-nephrotoxic immunosuppressive agents
  - Tolerance induction strategies
Mission Possible?
Recurrent FSGS

- Recurrence rate: 20-50% (difficult because of the focal nature of the distribution of lesions and possible sampling error)
  - Primary vs secondary vs de novo-rapamycin related?
  - Familial forms do not recur after transplantation (linked to genes encoding various podocyte-related proteins, such as podocin, alfa-actinin 4 and nephrin)
- In series with primary FSGS and including pediatric patients and young adults: the incidence of recurrence is as high as 50%
Treatment for Recurrent FSGS

- No prospective randomized studies
- Plasmapheresis ± dipyridamole (pre-or post transplant), ~50% relapse after stopping plasmapheresis, less effective in adults
- Pre-transplant plasmapheresis in children
- Plasma protein adsorption
- High-dose cyclosporine, cyclophosphamide
- ACE inhibitors, NSAIDs, rapamycin use (controversial)
Treatment for Recurrent FSGS

- Combination of ACE-inhibitor, an AT 1 receptor blocker and the direct renin inhibitor aliskiren  
- Combination of plasmapheresis and rituximab
- Anti-TNF alpha treatment (infliximab then etanercept)
- Galactose (oral or IV): a sugar with high *in vitro* affinity for FSPF in chromatographic studies; a trace amount of galactose blocks or reverses the increase in $P_{alb}$
- High dose of oral steroids, IV cyclosporine followed by oral treatment, PE with 5% albumin replacement until month 9, and ramipril
Screening Protocol Based on BK Viral Load

Treatment of BK Nephropathy

- Multicenter prospective studies are needed:
  - Stratifying histologic grading and renal function
  - Use of viral load for diagnosis
  - Evaluation of different treatment strategies: assessing the possibility of chronic allograft dysfunction due to systematic reduction of immunosuppression
  - Longer follow-up
Early detection and intervention before CAN occurs

Review CNI dose to avoid nephrotoxicity

Avoid acute and subclinical rejection episodes

Early intervention for CAN

Tailor patient management considering donor, recipient and graft factors
- HLA match
- Reduce ischemic injury
- Maximize graft function
- Increase surveillance for older donor kidney

Manage and monitor co-morbidities
- Hyperlipidemia
- Hypertension
- Diabetes

Death-censored allograft survival stratified by % reduction in iDSA at 14 days postbiopsy (p = 0.021; log-rank)

Kaplan–Meier survival in patients with or without DSA at rejection diagnosis (p = 0.001; log-rank)

52 patients with acute rejection; 16 (31%) with de novo DSA; median follow-up 27.0±17.4 months post acute rejection; de novo DSA significant risk factor for allograft loss but prompt DSA reduction was associated with improved allograft survival

Subclinical Rejection (SCR)

- Looking at the predictive value of SCR and/or CAN in protocol biopsies on death-censored graft survival
- Protocol biopsy was done during the first 6 months in stable grafts (n=435 transplants)
- Borderline changes and acute rejection: SCR
- Presence of interstitial fibrosis and tubular atrophy: CAN
- Mean follow-up was 91±46 months
- Biopsies were classified as normal (n=186), SCR (n=74), CAN (n=110) and SCR with CAN (n=65)

Cox regression analysis: SCR with CAN and hepatitis C virus were independent predictors of graft survival

<table>
<thead>
<tr>
<th>Trial</th>
<th>Treatment Groups</th>
<th>GFR (mL/min)</th>
<th>AR (%)</th>
<th>Notes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Larson, et al&lt;sup&gt;36&lt;/sup&gt;</td>
<td>Mod-High RAPA</td>
<td>63&lt;sup&gt;*&lt;/sup&gt;</td>
<td>19&lt;sup&gt;*&lt;/sup&gt;</td>
<td>*P=NS vs. standard TAC. Mostly living donors</td>
</tr>
<tr>
<td></td>
<td>Standard TAC</td>
<td>61</td>
<td>14</td>
<td></td>
</tr>
<tr>
<td>CAESAR&lt;sup&gt;37&lt;/sup&gt;</td>
<td>CYA Withdrawal</td>
<td>51</td>
<td>38&lt;sup&gt;*&lt;/sup&gt;</td>
<td>*P=0.04 vs. standard CYA;</td>
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<tr>
<td></td>
<td>CYA Minimization</td>
<td>51</td>
<td>25</td>
<td>*P=0.03 vs. CYA minimization.</td>
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<tr>
<td></td>
<td>Standard CYA</td>
<td>49</td>
<td>28</td>
<td></td>
</tr>
<tr>
<td>SYMPHONY&lt;sup&gt;19&lt;/sup&gt;</td>
<td>Low TAC</td>
<td>65&lt;sup&gt;*&lt;/sup&gt;</td>
<td>12&lt;sup&gt;*&lt;/sup&gt;</td>
<td>*P&lt;0.001 vs. others.</td>
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<tr>
<td></td>
<td>Low CYA</td>
<td>59</td>
<td>24</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Low RAPA</td>
<td>57</td>
<td>37</td>
<td>“Low-medium risk” mostly deceased donor transplants</td>
</tr>
<tr>
<td></td>
<td>Standard CYA</td>
<td>57</td>
<td>26</td>
<td>Serious adverse events highest in RAPA group.</td>
</tr>
<tr>
<td>Belatacept Study Group&lt;sup&gt;18&lt;/sup&gt;</td>
<td>High Belatacept</td>
<td>66&lt;sup&gt;*&lt;/sup&gt;</td>
<td>7</td>
<td>6-month results. *P=0.01;</td>
</tr>
<tr>
<td></td>
<td>Low Belatacept</td>
<td>62&lt;sup&gt;5&lt;/sup&gt;</td>
<td>6</td>
<td>*P=0.04 vs. standard CYA. Monthly IV infusions required for Belatacept.</td>
</tr>
<tr>
<td></td>
<td>Standard CYA</td>
<td>54</td>
<td>8</td>
<td></td>
</tr>
<tr>
<td>FREEDOM&lt;sup&gt;22&lt;/sup&gt;</td>
<td>CS Avoidance</td>
<td>59</td>
<td>32&lt;sup&gt;*&lt;/sup&gt;</td>
<td>*P=0.007 vs. standard CS.</td>
</tr>
<tr>
<td></td>
<td>Early CS Withdrawal</td>
<td>59</td>
<td>26</td>
<td>Blacks underrepresented. CYA-based regimens.</td>
</tr>
<tr>
<td></td>
<td>Standard CS</td>
<td>61</td>
<td>15</td>
<td></td>
</tr>
<tr>
<td>Astellas Steroid Withdrawal Group&lt;sup&gt;23&lt;/sup&gt;</td>
<td>Early CS Withdrawal</td>
<td>59</td>
<td>18&lt;sup&gt;*&lt;/sup&gt;</td>
<td>5-year results. *P=0.04 vs. low CS. More CAN (post hoc). TAC-based regimens</td>
</tr>
<tr>
<td></td>
<td>Low CS</td>
<td>60</td>
<td>11</td>
<td></td>
</tr>
<tr>
<td>CONVERT&lt;sup&gt;20&lt;/sup&gt;</td>
<td>CNI Conversion to RAPA</td>
<td>63&lt;sup&gt;*&lt;/sup&gt;</td>
<td>16</td>
<td>2-year results. *P=0.009 vs. CNI Continuation. Adverse events higher but malignancy lower with RAPA.</td>
</tr>
<tr>
<td></td>
<td>CNI Continuation</td>
<td>60</td>
<td>15</td>
<td></td>
</tr>
</tbody>
</table>
Why do Patients Reject on Belatacept?

- Low saturation of CD86/CD80
- Presence of memory cells
- T cell activation through other costimulation pathways
- Blockade of negative signaling
- Inhibition of T regulatory cells
Belatacept

• BENEFIT study-phase III: 686 patients; 3-arm, basiliximab induction
  • At Month 12, both belatacept regimens had similar patient/graft survival versus cyclosporine (MI: 95%, LI: 97% and cyclosporine: 93%)
  • Belatacept patients experienced a higher incidence (MI: 22%, LI: 17% and cyclosporine: 7%) and grade of acute rejection episodes
  • 5 PTLD in the belatacept group vs 1 in CsA group

Vincenti F, et al. AJT 2010
Belatacept

- BENEFIT-EXT study-phase III: 578 patients; 3-arm, basiliximab induction
  - Patient/graft survival with belatacept was similar to cyclosporine (86% MI, 89% LI, 85% cyclosporine) at 12 months
  - The incidence of acute rejection was similar across groups (18% MI; 18% LI; 14% cyclosporine)
  - One patient (0.5%) in the belatacept MI group and two patients (1%) in the LI group had PTLD during the 12-month period and one additional patient in each belatacept group developed PTLD after Month 12
  - Four of the five cases involved the central nervous system, and two of five (both of the post- Month 12 cases) had CMV disease
- No patients on cyclosporine developed PTLD

Durrbach A, et al. AJT 2010
# Relative Risk of Acute Rejection

Knight SR, et al. Transplantation 2009

<table>
<thead>
<tr>
<th>Study/subcategory</th>
<th>MMF (n/N)</th>
<th>AZA (n/N)</th>
<th>RR [95% CI]</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. CsA SIM</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sollinger 1995</td>
<td>70/333</td>
<td>67/166</td>
<td>0.52 [0.39,0.69]</td>
</tr>
<tr>
<td>Keown 1996</td>
<td>66/337</td>
<td>60/166</td>
<td>0.54 [0.40,0.73]</td>
</tr>
<tr>
<td><strong>Subgroup total</strong></td>
<td>670</td>
<td>332</td>
<td><strong>0.53 [0.43,0.65]</strong></td>
</tr>
<tr>
<td>2. CsA ME</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Egford 1999</td>
<td>8/25</td>
<td>11/25</td>
<td>0.73 [0.35,1.50]</td>
</tr>
<tr>
<td>Suhall 2000</td>
<td>2/20</td>
<td>7/20</td>
<td>0.29 [0.07,1.21]</td>
</tr>
<tr>
<td>Milladipour 2002</td>
<td>4/40</td>
<td>10/40</td>
<td>0.40 [0.14,1.17]</td>
</tr>
<tr>
<td>Sadek 2002</td>
<td>27/162</td>
<td>43/157</td>
<td>0.61 [0.40,0.93]</td>
</tr>
<tr>
<td>Tuncer 2002</td>
<td>7/38</td>
<td>13/38</td>
<td>0.54 [0.24,1.20]</td>
</tr>
<tr>
<td>Remuzzi 2004</td>
<td>57/124</td>
<td>65/124</td>
<td>0.88 [0.68,1.13]</td>
</tr>
<tr>
<td>Merviile 2004</td>
<td>5/37</td>
<td>7/34</td>
<td>0.66 [0.23,1.87]</td>
</tr>
<tr>
<td>Joh 2005</td>
<td>10/34</td>
<td>9/34</td>
<td>1.11 [0.52,2.39]</td>
</tr>
<tr>
<td>Weimer 2006</td>
<td>5/31</td>
<td>9/25</td>
<td>0.45 [0.17,1.17]</td>
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<tr>
<td><strong>Subgroup total</strong></td>
<td>511</td>
<td>497</td>
<td><strong>0.70 [0.58,0.85]</strong></td>
</tr>
<tr>
<td>3. Tacrolimus</td>
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<td></td>
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<tr>
<td>Mendez 1998</td>
<td>24/117</td>
<td>19/59</td>
<td>0.64 [0.38,1.07]</td>
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<tr>
<td>Johnson 2000</td>
<td>12/72</td>
<td>16/76</td>
<td>0.79 [0.40,1.56]</td>
</tr>
<tr>
<td>Wlodarczyk 2002</td>
<td>46/243</td>
<td>70/246</td>
<td>0.67 [0.48,0.92]</td>
</tr>
<tr>
<td><strong>Subgroup total</strong></td>
<td>432</td>
<td>381</td>
<td><strong>0.68 [0.52,0.87]</strong></td>
</tr>
<tr>
<td>4. CsA (unknown)</td>
<td></td>
<td></td>
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</tr>
<tr>
<td>Army Hospital Delhi 2002</td>
<td>1/17</td>
<td>3/16</td>
<td>0.31 [0.04,2.71]</td>
</tr>
<tr>
<td>Baltar 2002</td>
<td>1/14</td>
<td>5/12</td>
<td>0.17 [0.02,1.27]</td>
</tr>
<tr>
<td>Sun 2002</td>
<td>2/40</td>
<td>6/46</td>
<td>0.38 [0.08,1.79]</td>
</tr>
<tr>
<td><strong>Subgroup total</strong></td>
<td>71</td>
<td>74</td>
<td><strong>0.29 [0.10,0.82]</strong></td>
</tr>
<tr>
<td><strong>Overall total</strong></td>
<td>1584</td>
<td>1284</td>
<td><strong>0.62 [0.55,0.70]</strong></td>
</tr>
</tbody>
</table>
Hazard ratio for graft loss including death with a functioning graft

Knight SR, et al. Transplantation 2009