Novel Targets for Immunosuppression in Clinical Trial

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Total Transplants, Vanderbilt/Nashville VA Kidney/Pancreas Transplant Program

Total Number of Transplants Kidney/Pancreas Program, Vanderbilt/Nashville VA, 2004 – 2008
## Adult Kidney Transplants VUMC/VA

<table>
<thead>
<tr>
<th></th>
<th>2004</th>
<th>2005</th>
<th>2006</th>
<th>2007</th>
<th>2008</th>
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<tr>
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<td>60</td>
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<td>58</td>
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<tr>
<td>Living Donor</td>
<td>61</td>
<td>83</td>
<td>71</td>
<td>55</td>
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<td>Total</td>
<td>121</td>
<td>145</td>
<td>129</td>
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<tr>
<td>Deceased Donor</td>
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<td>2</td>
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<tr>
<td>Living Donor</td>
<td>6</td>
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<td>7</td>
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<tr>
<td>Total</td>
<td>8</td>
<td>9</td>
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## Adult Pancreas Transplants

<table>
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<tr>
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<td>SPK</td>
<td>4</td>
<td>5</td>
<td>12</td>
<td>5</td>
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<tr>
<td>PAK</td>
<td>7</td>
<td>2</td>
<td>1</td>
<td>2</td>
<td>1</td>
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<tr>
<td>Total</td>
<td>11</td>
<td>7</td>
<td>13</td>
<td>7</td>
<td>10</td>
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SPK – Simultaneous Pancreas Kidney  
PAK – Pancreas After Kidney
## VUMC/Nashville VA Patient and Graft Survival

### Vanderbilt Adult

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<tr>
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<th>1 Year</th>
<th>3 Year</th>
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<tr>
<td>Patient Survival</td>
<td>98%</td>
<td>94%</td>
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<tr>
<td>Graft Survival – Deceased Donor</td>
<td>93%</td>
<td>84%</td>
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<tr>
<td>Graft Survival – Living Donor</td>
<td>95%</td>
<td>93%</td>
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### Vanderbilt Pediatric

<table>
<thead>
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<th>1 Year</th>
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<tbody>
<tr>
<td>Patient Survival</td>
<td>94%</td>
<td>94%</td>
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<tr>
<td>Graft Survival – Deceased Donor</td>
<td>80%</td>
<td>75%</td>
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<td>Graft Survival – Living Donor</td>
<td>92%</td>
<td>85%</td>
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<tr>
<td></td>
<td>1 Year</td>
<td>3 Year</td>
</tr>
<tr>
<td>--------------------------</td>
<td>--------</td>
<td>--------</td>
</tr>
<tr>
<td>Patient Survival</td>
<td>98%</td>
<td>96%</td>
</tr>
<tr>
<td>Graft Survival – Deceased Donor</td>
<td>91%</td>
<td>88%</td>
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<tr>
<td>Graft Survival – Living Donor</td>
<td>100%</td>
<td>100%</td>
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Total Waitlist Additions, Vanderbilt/Nashville VA Kidney/Pancreas Transplant Program

Total Number of Waitlist Additions Kidney/Pancreas Program, Vanderbilt/Nashville VA, 2004 – 2008

- VUMC
- VA
- Total

Year | VUMC | VA | Total
--- | --- | --- | ---
2004 | 166 | 51 | 217
2005 | 203 | 52 | 255
2006 | 208 | 71 | 279
2007 | 278 | 52 | 330
2008 | 266 | 69 | 335
## Median Waiting Time for Deceased Donor Kidney Transplant, VUMC

<table>
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<tr>
<th>Calendar Year</th>
<th>Median (Days)</th>
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<td>2008</td>
<td>561</td>
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<td>2007</td>
<td>415</td>
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<td>2006</td>
<td>548</td>
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<td>2005</td>
<td>292</td>
</tr>
<tr>
<td>2004</td>
<td>609</td>
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Patients Waiting for a Kidney Transplant vs. Deceased Donor Kidneys Available

Kidney Transplant Waitlist vs. Deceased Donors

- Tennessee statewide Waitlist
- VUMC + Nashville VA Waitlist
- Tennessee Statewide Deceased Donors
New Immunosuppressive Transplant Medications

- ISA 247 - CNI
- Lea29Y – Co Stimulatory Blockade
- Efalizumab - Co Stimulatory Blockade
- AEB – PKC Inhibition
- CP690550 – JAK3 Inhibition
- KRP – 203 Lymphocyte Trafficking Inhibition
Naive T cell

Signal 1

IL-2R

CD28

ICOS

Immune synapse

TCR-CD3

CD40L

Signal 1

Activation

CD40

MHC-antigen

B7

B7RP-1

↓

Signal 1

↑

Signal 2

↓

Signal 2

IL-2R

↓

Signal 3

IL-2

IL-2 mRNA

APC

↓

CD40

MHC-antigen B7

B7RP-1

↓

Signal 1

↑

Signal 2

↓

Signal 2

IL-2R

↓

Signal 3
Calcineurin Inhibition

ISA 247

- Oral analogue of CS
- Modification of the functional group on AA I
- More potent CNI than CSA
- No nephrotoxicity in animals
- Prolongs organ transplants in animals
- Phase III Psoriasis trial underway

Gregory et al. Transplantation 78:2004
Structure of ISA 247 and cyclosporine A

Yabu, JM; Vincenti, F. Transplantation updates 2(1), 6. 2008
6 month phase II renal transplant study (n=120).

ISA 247 (0.4, 0.6, 0.8 mg/lg 2x/d) vs. TAC, IL2R-ab

Induction, Maintenance:
MMF + Steroids

AR 14%, 12%, 0% ISA 247 v 14% TAC
Costimulation in T cell activation

(a) APC

- CD40
- MHC-antigen
- B7
- B7RP-1

Immune synapse

Naive T cell

- TCR-CD3
- CD28
- ICOS
- IL-2R
- IL-2
- Signal 1
- Signal 2
- Activation
- IL-2 mRNA

(b) APC

- CD40
- MHC-antigen
- B7
- B7RP-1

- TCR-CD3
- CD28
- ICOS
- IL-2R
- IL-2
- Signal 1
- Signal 2
- Activation
- IL-2 mRNA

(c) APC

- CD40
- MHC-antigen
- B7RP-1
- B7
- CTLA-4

- TCR-CD3
- ICOS
- IL-2R
- IL-2
- Signal 1

- Anergy
- T cell
CD 28 – CD 80/86 Pathway

1st studied reagent CTLA4Ig
- fusion protein
- extra cellular domain of CTLA4
- Fc portion of human Ig

Excellent pre clinical results but less spectacular non-human primate data

Sayegh and Turkha NEJM 338:1998
CD28 – CD80/86

- LEA 29Y (Belatacept)
  - Leucine 104 → Glutamate
  - Alanine 29 → Tyrosine
- 2nd generation CTLA4Ig
- Substitutions increase theoretic efficacy
  - 2 fold ↑ binding to CD 80
  - 4 fold ↑ binding to CD 86
  - 10 fold ↑ T cell function inhibition

Larsen et al AJT 5:2005
CD28 – CD 80/86 Pathway

LEA 29Y Phase II Human Trial

- n = 218
- 3 Arms
  - High Dose LEA 29Y
  - Low Dose LEA 29Y
  - CSA Control
- Induction: basiliximab
- Maintenance: MMF + Seroids
- Results:
  - a) AR similar
  - b) GFR, CAN rate, metabolic complications favored

LEA 29Y

Interference with Adhesion
Molecule Directed Signal Two

- CD 154 - C40 Pathway
- CD 28 - CD80/86 Pathway
- LFAI – ICAM Pathway
CD 154 – CD 40 Pathway

- Humanized moab directed to D 154
- proof of principle in non-human primates

Kirk et al. Nat Med 5:1999
CD 154 – CD 40 Pathway

- Phase 1 human trial of Hu5C8 – humanized α CD 154 MOAB
  - Short course of steroids
    - MMF
    - CNI free
  - Halted because of thromboembolic events
  - Complication not epitope specific, other α CD 154 MOABs had same complication
CD 154 – CD 40 Pathway

MOAB targeting CD 40 have not been shown to be procoagulant and may be prepared for study
CD 28 – CD 80/86 Pathway

1\textsuperscript{st} studied reagent CTLA4Ig
- fusion protein
- extra cellular domain of CTLA4
- Fc portion of human Ig

Excellent pre clinical results but less spectacular non-human primate data

Sayegh and Turkha NEJM 338:1998
Chimeric versus humanized monoclonal antibodies

Complementarity determining regions (CDRs)

Light chain (L)

Heavy chain (H)

Mouse mAb

Chimeric Human–mouse mAb

Human mAb

Fully humanized mAb

CDRs giving desired antigen-binding site (i.e., specificity)
CD28 – CD80/86

LEA 29Y (Belatacept)

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Alanine 29 → Tyrosine

2nd generation CTLA4Ig

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CD28 – CD 80/86 Pathway

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- Results:
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  - b) GFR, CAN rate, metabolic complications favored

LEA 29Y

Phase III in ECD kidneys are in standard donor recipients.
Conversion trial (CNI)
Rapid steroid withdrawal trial (thymoglobulin + LEA 29 Y + either MMF or mTOR I)
PHASE 3 BELATACEPT TRIALS

**Benefit Extended** – trial in extended criteria donor recipients

3 arms – CSA/pred/MMF control; “high” dose Bela/MMF/pred; “moderate” dose bela/MMF/pred

**Benefit** – trial in standard criteria donor recipients of deceased and live donor kidneys

Same 3 arms

**Steriod Sparing**

3 arms – Group A: bela/mmf; Group B bela/rap; Group C tac/mmf
1. In both Benefit EXT and Benefit:
   a. More acute rejection in Bela group, easily reversed
   b. Equal graft and patient outcomes over the 4 years of report
   c. Statistically better GFR and lower creatinine in Bela groups
   d. Less hypertension and hyperlipidemia in bela groups
   e. No difference in number or degree of DGF in EXT

2. In Steroid Sparing Trial
   a. Bela permitted rapid discontinuance of steroids at 1 week
   b. Acute rejections when bela combined with tac or rap single digits
   c. Best results were in bela + rap group
CD28 – CD 80/86 Pathway

LEA 29 Y

- After periop induction LEA 29 Y administered monthly
- Parental administration reduces non-adherence to daily pill regimens
- A subcutaneous preparation would increase patient acceptance.
LFA I – ICAM Pathway

Efalizumab – Humanized moab α CD11a

- LFA I – 2 chain heterodimer β 2 integrin
  (α + β Chain)
- α chain = CD 11a
- β chain = CD 18
- Approved for use in psoriasis
Efalizumab Phase I/II human Trial

- Full dose CSA + MMF _ steroids v. half dose CSA + mTOR I + steroids
- 2 Doses of efalizumab (0.5 or 2.0 mg/kg) q week x 12 weeks
- AR at 6 mos 11% (4/38)
- Full dose CSA + high efalizumab abandoned because of 3 cases PTLD

Vincenti et al AJT 7:2007
Intracellular Signalling

- Protein Kinase C Signalling Couples T cell receptor engagement to downstream activation (Signal One)
  
  and
  
  Costimulation (CD28) to gene activation (Signal two)
<table>
<thead>
<tr>
<th>Isoform</th>
<th>Predominant Tissue Expression</th>
<th>PKC Knockout Phenotype</th>
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<tr>
<td>Conventional © PKC subfamily</td>
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<td>α</td>
<td>Ubiquitous, high in T cells</td>
<td>T-cell defect</td>
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<tr>
<td>β</td>
<td>Ubiquitous, high in B cells</td>
<td>Neutrophil, B-, and mast-cell defects</td>
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<tr>
<td>γ</td>
<td>Brain</td>
<td>Not determined</td>
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<td>Novel (n) PKC subfamily</td>
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<td>T cells, platelets, monocytes</td>
<td>T-cell signaling defect</td>
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<td>τ</td>
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Site of action of the JAK 3 inhibitors.

Cytokine
IL-2, IL-4, IL-7, IL-9, IL-15, IL-21

Cytokine Receptor

Jak 1
Jak 3
Stat5

G1 → Transcription → S

Yabu, JM; Vincenti, F. Transplantation updates 2(1), 7. 2008
Intracellular Signalling

- PKC α, β, θ important in Lymphocyte Signaling
- PKC θ T cell restricted, mediates transcription factor protein-1 & Nfkβ (on ko mice)

Intracellular Signalling

AEB
- oral low MW inhibitor of PKC isoforms
- inhibits T cell activation and IL 2 synthesis
- little effect on NFAT
- No effect on cytokine directed cell proliferation-(signal three)

Evenou et.al. AJT 6:2006
Intracellular Signaling

AEB Preclinical

- Prolonged heart and kidney grafts
  - Bruns et al. AJT 6:2006
  - Wagner et al. AJT 6:2006

- Prolonged Kidney transplantation in cynomolgus monkey
  - Bigaud et al. AJT 6:06
Intracellular Signaling

3 Phase II human AEB Trials

1. 12 mos. 3 Arms – AEB + TAC; AEB + reduced TAC; MMF + TAC- induction α IL2 R
   maintenance: steroids + MMF

2. 12 mos. 2 ARMS – AEB vs TAC; Induction α IL2 R - maintenance : steroids + MPA

3. 12 mos. 2 ARMS – AEB vs CSA- Induction - IL2 R , maintenance: mTor I + steroids
Intracellular Signaling from the Cytokine Receptor

CP690550

- JAK 3 inhibitor
- JAK 3 resistant to hematopoietic cells
- JAK 3 associates with common α chain of cytokine R for IL2, 4, 7, 9, 11, 21
- Dimerizes STAT 5 after phosphorylation, then can traverse nucleus and activate gene regulation of cell division

Saemann et al Transplant Int 17:2004
Intracellular Signaling from the Cytokine Receptor

CP690550

Preclinical Studies

- **Non-human primate studies**- good graft survival, no nephrotoxicity ↑ polyoma, anemia, GI sx (Borie et al. Transplantation 79:2005)
Intracellular Signaling from the Cytokine Receptor

CP690550

Phase II human Kidney transplant Trial
- 3 ARMS: 15mg CP690550 BID vs. 30mg. CP690550 BID vs. TAC BID
- Induction IL2R ab
- Maintenance: MMF + Steroids

Results
- Comparable AR
- 100% pt and graft survival
- No PTLD
- CMV & polyoma in high dose group

- Study into 2 yr extension
- 2nd phase II against CSA underway
Lymphocyte Trafficking

FTY 720

- 2, Phase III renal Transplant trials showed no benefit
- Important side effects
  - Bradycardia
  - Macular edema
- Studies in transplant abandoned

Tedesco – Silva et al. Transplantation 82: 2006
Salvadori et al. AJT 6:2006
Lymphocyte Trafficking

- Selective agonist at G protein coupled Sphingosine 1-phosphate receptor
- Thought to avoid FTY 720 side effects
- Prolonged rodent heart transplants

Kahan et al. AJT 7: 2007
Shimizu et al. Circulation 111:2005
Table 1: Small molecules in clinical trials

<table>
<thead>
<tr>
<th>Drug</th>
<th>Pathway</th>
<th>Phase of Study</th>
<th>Maintenance Regimen</th>
<th>Results/1° Endpoint</th>
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<tbody>
<tr>
<td>ISA247 (Isotechnika)</td>
<td>Calcineurin inhibitor (signal one)</td>
<td>Phase II</td>
<td>Three dose levels of ISA247 versus Tac. All patients are treated with MMF + CS</td>
<td>1° Endpoint: similar efficacy and renal function in all treatment groups</td>
</tr>
<tr>
<td>AEB07 (Novartis)</td>
<td>Protein kinase C (signal one and two)</td>
<td>Phase II</td>
<td>AEB + Everolimus + CS</td>
<td>1° Endpoint: acute rejection</td>
</tr>
<tr>
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<td>Phase II</td>
<td>AEB + Tac + CS with Tac withdrawal at 3 months versus Tac + MPS + CS</td>
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<tr>
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<td>Phase II</td>
<td>AEB + MPS + CS versus Tac + MPS + CS</td>
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<tr>
<td>CP 690,550 (Pfizer)</td>
<td>Janus kinase 3 (signal three)</td>
<td>Phase Ila</td>
<td>CP 690,550 15 mg or 30 mg bid + MMF + CS versus Tac + MMF + CS</td>
<td>Comparable efficacy between all treatment groups. More infections in high CP690, 550</td>
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<td>Clinical trial in progress with 2 doses CP 690,550 15 mg and 10 mg bid with MMF + CS</td>
<td>1° Endpoint: acute rejection</td>
</tr>
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</table>

CS = corticosteroids; Tac = tacrolimus; MPS = mycophenolic sodium; MMF = mycophenolate mofetil.