Pharmacogenetics to tailor Drug Exposure and Outcomes in Kidney Transplantation

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Disclosures

Consulting fees
Astellas Pharma, Glaxo Smith Kline, Novartis Pharma

Grant support
Astellas Pharma and Bristol-Myers Squibb

Lecture fees
Astellas Pharma, Chiesi Pharma, Fresenius Medical Care, MSD, Roche
“Personalized medicine”

- Choose the most appropriate drug for each individual
- Select an optimal dose
- Identify those at risk from (atypical) adverse drug reactions
Focus on tacrolimus
Tacrolimus

- Dosed by means of therapeutic drug monitoring (TDM)
- Metabolized by Cytochrome P450 3A5 (CYP3A5)
- Several single-nucleotide polymorphisms (SNP) in CYP3A5 gene
- Best characterized is the CYP3A5*3 SNP
- Causes alternative splicing and results in the absence of protein

Dose-corrected tacrolimus C0
(ng/mL per mg/kg)

Time after transplantation (days)

CYP3A5 expressers
CYP3A5 non-expressers

Hesselink, Pharmacogenet Genom 2008;18:339-48
Dose-corrected tacrolimus C0 (ng/mL per mg/kg)

CYP3A5 expressers
CYP3A5 non-expressers

Time after transplantation (days)

Hesselink, Pharmacogenet Genom 2008;18:339-48
**Tacrolimus and CYP3A5**

- *CYP3A5* expressors need a ~50% higher Tac dose to reach target concentrations after kidney transplantation.

- This has been observed among heart, lung and liver transplant recipients, both adults and children.

References:

- Thervet, Transplantation 2003;76:1233-5;
- Anglicheau, Clin Pharmacol Ther 2004;75:422-33
- Haufroid, Pharmacogenetics 2004;14:147-54;
- MacPhee, Am J Transplant 2004;4:914-9;
- Tsuchiya, Transplantation 2004;78:1182-7
- Zhao, Clin Pharmacol Ther 2009,86:609-18
- Birdwell, Pharmacogenet Genom 2011;22:32-42
CYP3A5*3 is the top SNPP
Residual variability

Genomewide Association Study of Tacrolimus Concentrations in African American Kidney Transplant Recipients Identifies Multiple CYP3A5 Alleles

W. S. Oetting1,*, D. P. Schladt2, W. Guan3, M. B. Miller3, R. P. Remmel1, C. Dorr2, K. Sanghavi1, R. B. Mann4, B. Herrera5, A. J. Matas6, D. R. Salomon7, P.-Y. Kwok5, B. J. Keating8 and A. K. Israni2,9,10; and P. A. Jacobson1,7 for the DeKAF Investigators

The Pharmacogenomics Journal 15, 288-292 (June 2015) | doi:10.1038/tpj.2014.67

High frequency and founder effect of the CYP3A4*20 loss-of-function allele in the Spanish population classifies CYP3A4 as a polymorphic enzyme

M Apellániz-Ruiz, L Inglada-Pérez, M E G Naranjo, L Sánchez, V Mancikova, M Currás-Freixes, A A de Cubas, I Comino-Méndez, S Triki, A Rebai, M Rasool, G Moya, M Grazina, G Opocher, A Cascón, P Taboada-Echalaz, M Ingelman-Sundberg, A Carracedo, M Robledo, A Lierena and C Rodríguez-Antona

The Pharmacogenomics Journal 15, 288-292 (June 2015) | doi:10.1038/tpj.2014.67
Novel *CYP3A4* intron 6 polymorphism

- *CYP3A4* intron 6 SNP (rs35599367 C>T), *CYP3A4*\(^{*}22\) allele

- T variant associated with decreased hepatic mRNA expression and decreased *CYP3A4* enzymatic activity

- T variant associated with lower statin doses required for lipid control

- Minor allele frequency ~5%

Wang, Pharmacogenomics J 2010;11:274-86
**CYP3A5 + CYP3A4 genotype affects Tac dose requirement**

- Poor metabolizers (group 1)
- Intermediate metabolizers (groups 2+3)
- Extensive metabolizers (group 4)

**CYP3A5 non-expressers + CYP3A4*22 T variant allele carriers**

- CYP3A5 non-expressers + CYP3A4*22 CC wildtype or
- CYP3A5 expressers + CYP3A4*22 T variant allele

- CYP3A5 expressers + CYP3A4*22 CC wildtype

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Elens, Clin Chem 2011;57:1574-83
Poor metabolizers are at risk of early Tac overexposure

Fig. 1. Percentage of patients within each CYP3A metabolizer cluster stratified by day 3 values of $C_0$ below or above the 15-μg/L supratherapeutic threshold.
**CYP3A5 + CYP3A4 + POR genotype and tacrolimus**

Kuypers, Pharmacogenetics Genom 2014;24:597-606
CYP3A5 + CYP3A4 + POR genotype and tacrolimus

Kuypers, Pharmacogenetics Genom 2014;24:597-606
DEMAND EVIDENCE AND THINK CRITICALLY
Hypothesis

- A CYP3A5-based tacrolimus (starting)dose is more effective and safe compared to conventional bodyweight-based Tac dosing
TACTIC trial - Study design

Transplantation
TACTIC trial - Study design

Transplantation

Genotyping → Basiliximab / ATG

MMF + CS
TACTIC trial - Study design

Transplantation

Genotyping → Basiliximab / ATG

MMF + CS

Randomization

Day 7

Start tacrolimus (n = 280)
TACTIC trial - Study design

Transplantation

Genotyping

Randomization
Day 7
Start tacrolimus
(n = 280)

Basiliximab / ATG
MMF + CS

Usual daily dose
All: 0.20 mg/kg/d

Daily dose according to CYP3A5
CYP3A5 expressors: 0.30 mg/kg/d
CYP3A5 non-expressors: 0.15 mg/kg/d

Courtesy of dr. Eric Thervet
TACTIC trial - Study design

Transplantation

Genotyping

Randomization
Start tacrolimus (n = 280)

Day 7

Basiliximab / ATG
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Day 10
Proportion of patients within therapeutic target (10-15 ng/ml)

Courtesy of dr. Eric Thervet
Higher proportion within target range through genotyping

<table>
<thead>
<tr>
<th>End point</th>
<th>Control group (n=120)</th>
<th>Adapted-dose group (n=116)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Primary end point</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Proportion of patients with TAC C₀ in target range after six oral doses, % (95% CI)</td>
<td>29.1 % (22.8-35.5)</td>
<td>43.2% (36.0-51.2)</td>
<td>0.030</td>
</tr>
</tbody>
</table>
Higher proportion within target range through genotyping

**Underexposure**
- $P = 0.035$
- $n = 6$ ; $n = 4$

**Overshoot**
- $P < 0.0001$
- $n = 96$ ; $n = 90$

Thervet, Clin Pharmacol Ther 2010;87:721-6
TACTIC Clinical endpoints

- Comparable patient and graft survival
- Comparable incidence and severity of delayed graft function
- Comparable incidence of acute rejection
- Comparable renal function
- No differences in other adverse events
Extrapolation of TACTIC?

- Delayed introduction of Tac (after day 6)
- Heavy immunosuppression (rATG induction; high dose MMF)
- Immunologically low-risk population
Relevance of TACTIC?

- More relevant when Tac introduced on day of transplant?
- More relevant for immunologically high-risk population?
- Or when giving no rATG induction therapy or “normal” dose MMF?

➢ “Mozaiek study” (NTR 2226; www.trialregister.nl)
Hypothesis of the trial (NTR2226)

Pharmacogenetic *(CYP3A5)* adaptation of the Tac starting dose

- is more effective in reaching the Tac target concentration range

and

- leads to superior clinical outcomes

compared with conventional, bodyweight-based Tac dosing
Trial Design

Work-up for living kidney transplantation

Genotyping
Trial Design

Work-up for living kidney transplantation

Genotyping

TRANSPLANTATION
Randomization
Start tacrolimus
**Trial Design**

**Work-up for living kidney transplantation**

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Genotyping
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**TRANSPLANTATION**

**Randomization**

**Start tacrolimus**

- **Usual daily dose**
  - 0.20 mg/kg/d

- **Daily dose according to CYP3A5**
  - CYP3A5 expressors: 0.30 mg/kg/d
  - CYP3A5 non-expressors: 0.15 mg/kg/d
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Start tacrolimus

Usual daily dose
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CYP3A5 expressors: 0.30 mg/kg/d
CYP3A5 non-expressors: 0.15 mg/kg/d

Proportion of patients within therapeutic Tac range (day 3)
Trial Design

Work-up for living kidney transplantation

Genotyping

TRANSPLANTATION
Randomization
Start tacrolimus

Usual daily dose
0.20 mg/kg/d

Daily dose according to CYP3A5
CYP3A5 expressors: 0.30 mg/kg/d
CYP3A5 non-expressors: 0.15 mg/kg/d

Proportion of patients within therapeutic Tac range (day 3)

MONTH 3 Clinical endpoints: acute rejection, nephrotoxicity
571 Screened

Not included (n = 317)
- Deceased donor kidney transplantation (n = 179)
- AB0-incompatible kidney transplantation (n = 25)
- HLA-identical kidney transplantation (no basiliximab) (n = 28)
- Use of immunosuppressive drugs other than prednisolone (n = 34)
- No DNA available (n = 23)
- Use of interacting drugs (n = 17)
- Participation in other clinical trial (n = 2)
- Non-renal transplantation (n = 5)
- Re-transplanted and already participated in trial (n = 4)

254 Eligible patients

No informed consent (n = 14)

240 Randomized
## Primary endpoint

<table>
<thead>
<tr>
<th>Concentration Type</th>
<th>Standard-dose group</th>
<th>Genotype-based group</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>Supra-therapeutic concentration</td>
<td>39 (39.4%)</td>
<td>31 (29.8%)</td>
<td>0.20</td>
</tr>
<tr>
<td>Therapeutic concentration</td>
<td>37 (37.4%)</td>
<td>37 (35.6%)</td>
<td>0.79</td>
</tr>
<tr>
<td>Sub-therapeutic concentration</td>
<td>23 (23.2%)</td>
<td>36 (34.6%)</td>
<td>0.10</td>
</tr>
</tbody>
</table>

Shuker et al., Am J Transplant 2016;16:2085-96
Comparable time-to-target Tac concentration range

\[ P = 0.72 \]

Shuker et al., Am J Transplant 2016;16:2085-96
Safety

- AEs: 728 (SDG) versus 750 (GBG); \( p = 0.56 \)
- SAEs: 148 (SDG) versus 167 (GBG); \( p = 0.40 \)
- 1x death from bacterial peritonitis (SDG)
- Graft survival 97.5% versus 99.2% (3-month)
- 4 graft losses (all vascular complications)
No difference in BPAR

<table>
<thead>
<tr>
<th>Rejection type</th>
<th>Whole group (n = 237)</th>
<th>Standard-dose group (n = 119)</th>
<th>Genotype-based group (n = 118)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Borderline</td>
<td>5 (2.1%)</td>
<td>3 (2.5%)</td>
<td>2 (1.7%)</td>
</tr>
<tr>
<td>Type I</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1A</td>
<td>0 (0.0%)</td>
<td>0 (0.0%)</td>
<td>0 (0.0%)</td>
</tr>
<tr>
<td>1B</td>
<td>1 (0.4%)</td>
<td>1 (0.8%)</td>
<td>0 (0.0%)</td>
</tr>
<tr>
<td>Type 2</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2A</td>
<td>9 (3.8%)</td>
<td>4 (3.4%)</td>
<td>5 (4.2%)</td>
</tr>
<tr>
<td>2B</td>
<td>7 (3.0%)</td>
<td>3 (2.5%)</td>
<td>4 (3.4%)</td>
</tr>
<tr>
<td>Type 3</td>
<td>1 (0.4%)</td>
<td>1 (0.8%)</td>
<td>0 (0.0%)</td>
</tr>
<tr>
<td>ABMR</td>
<td>0 (0.0%)</td>
<td>0 (0.0%)</td>
<td>0 (0.0%)</td>
</tr>
<tr>
<td>Mixed ACR and AMBR</td>
<td>7 (3.0%)</td>
<td>3 (2.5%)</td>
<td>4 (3.4%)</td>
</tr>
<tr>
<td><strong>Total BPAR</strong></td>
<td><strong>25 (10.5%)</strong></td>
<td><strong>12 (10.1%)</strong></td>
<td><strong>13 (11.0%)</strong></td>
</tr>
</tbody>
</table>

Shuker et al., Am J Transplant 2016;16:2085-96
Conclusions

• *CYP3A5* genotype-based Tac dosing does not lead to:
  
  ✓ an earlier achievement of the tacrolimus target concentration

or

✓ an improvement of clinical outcomes

as compared with standard, bodyweight-based dosing

• Routine genotyping for *CYP3A5* cannot be recommended
Conclusions (2)

• Only ~35% “on target” at first steady state

• Considerable & unexplained residual variability in Tac exposure

• Rapid achievement of target exposure in both groups with TDM
General conclusions *CYP3A5* genotyping

- Pharmacogenetics-assisted Tac dosing
  - ... may get patients on target faster
  - ... may limit Tac over -and underexposure
- No demonstrated clinical benefit
- Other SNPs may explain residual variability in Tac PK
Algorithms to aid tacrolimus dosing

A New CYP3A5*3 and CYP3A4*22 Cluster Influencing Tacrolimus Target Concentrations: A Population Approach

Franc Andreu1,2 · Helena Colom2 · Laure Elens3,6 · Teun van Gelder4,5,6 · Ronald H. N. van Schaik5,6 · Dennis A. Hesselink4,6 · Oriol Bestard1 · Joan Torras1 · Josep M. Cruzado1 · Josep M. Grinyó1 · Nuria Lloberas1

Algorithms to aid tacrolimus dosing

Key Points

This is the first population PK study combining CYP3A5 and CYP3A4 genotype, age, and hematocrit that influence tacrolimus concentrations in renal transplant recipients.

This a externally validated prediction model to propose new clear dosage guidelines for each genotype.

Algorithms to aid tacrolimus dosing

Størset, Transplantation 2015;99:2158-66
Algorithms to aid tacrolimus dosing

**Standard risk**

<table>
<thead>
<tr>
<th>Week</th>
<th>Computer Group</th>
<th>Control Group</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>82</td>
<td>81</td>
</tr>
<tr>
<td>2</td>
<td>91</td>
<td>87</td>
</tr>
<tr>
<td>3</td>
<td>90</td>
<td>78</td>
</tr>
<tr>
<td>4</td>
<td>91</td>
<td>66</td>
</tr>
<tr>
<td>5</td>
<td>84</td>
<td>72</td>
</tr>
<tr>
<td>6</td>
<td>94</td>
<td>67</td>
</tr>
<tr>
<td>7</td>
<td>91</td>
<td>89</td>
</tr>
<tr>
<td>8</td>
<td>89</td>
<td>80</td>
</tr>
</tbody>
</table>

Computer Group vs Control Group

*Størset, Transplantation 2015;99:2158-66*
### Algorithms to aid tacrolimus dosing

#### Table 3.
Overview of clinical outcome at 8 weeks

<table>
<thead>
<tr>
<th></th>
<th>Computer group</th>
<th></th>
<th>Control group</th>
<th></th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Number/median (95% CI)</td>
<td>Range</td>
<td>Number/median (95% CI)</td>
<td>Range</td>
<td></td>
</tr>
<tr>
<td>Biopsy-proven acute rejections</td>
<td>3</td>
<td>5</td>
<td></td>
<td></td>
<td>0.455</td>
</tr>
<tr>
<td>Recorded infections</td>
<td>6</td>
<td>3</td>
<td></td>
<td></td>
<td>0.289</td>
</tr>
<tr>
<td>Glomerular filtration rate (mL/min per 1.73 m²)</td>
<td>59 (55-64)</td>
<td>30-87</td>
<td>53 (48-57)</td>
<td>31-80</td>
<td>0.046</td>
</tr>
<tr>
<td>Fasting plasma glucose, mmol/L</td>
<td>5.3 (5.1-5.5)</td>
<td>4.3-6.7</td>
<td>5.5 (5.4-5.7)</td>
<td>4.6-8.5</td>
<td>0.058</td>
</tr>
<tr>
<td>2-h plasma glucose, mmol/L</td>
<td>5.9 (5.6-6.6)</td>
<td>2.9-9.3</td>
<td>6.8 (6.1-8.1)</td>
<td>4.2-13.5</td>
<td>0.008</td>
</tr>
</tbody>
</table>
Future

- Further development of algorithm-based tacrolimus dosing
- Implementation of such algorithms into clinical practice
- Need for end-point studies
- Unmet need for prediction of pharmacodynamics / adverse events
BACK UP
Example - Azathioprine

- Treatment of:
  - Neoplasia (ALL)
  - Autoimmune disease (rheumatoid arthritis)
  - Inflammatory bowel disease (Crohn’s disease)
  - Prevention of rejection after transplantation

- Metabolized to 6-thioguanine nucleotides:
  - Immunosuppressive effect
  - Myelotoxicity, hepatotoxicity, pancreatitis, gastro-intestinal disturbances
Azathioprine (2)

- Thiopurine S-methyl transferase (TPMT) inactivates azathioprine
- TPMT deficiency leads to accumulation of active 6-thioguanine metabolites, resulting in severe hematologic toxicity
Thiopurine S-methyltransferase (TPMT) phenotype

TPMT activity shows a trimodal distribution

## Clinical Pharmacogenetics Implementation Consortium guideline

<table>
<thead>
<tr>
<th>Phenotype</th>
<th>Dosing recommendations for Aza</th>
<th>Classification of recommendations</th>
</tr>
</thead>
<tbody>
<tr>
<td>Homozygous wildtype or normal, high activity</td>
<td>Start with normal starting dose (e.g. 2-3 mg/kg/d). Allow 2 weeks to reach steady-state after each dose adjustment.</td>
<td>Strong</td>
</tr>
<tr>
<td>Heterozygote or intermediate activity</td>
<td>Start at 30-70% of “target-dose” (e.g. 1-1.5 mg/kg/d) and titrate based on tolerance. Allow 2-4 weeks to reach steady state after each adjustment.</td>
<td>Strong</td>
</tr>
<tr>
<td>Homozygous variant, deficient activity</td>
<td>Consider alternative agents. If necessary start at 10% of “target dose” and dose thrice weekly instead of daily. Allow 4-6 weeks to reach steady-state after each dose adjustment.</td>
<td>Strong</td>
</tr>
</tbody>
</table>
... Exome sequencing revealed a novel SNP (c.802C>T) resulting in a premature stop codon in *CYP3A4* exon 5. ... This is, to our knowledge the first case of a complete failure of CYP3A4 in humans.