IMI project - SAFE-T
An European consortium approach to renal safety biomarkers

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On behalf of DIKI subgroup
Introduction to SAFE-T

• **Safer And Faster Evidence-based Translation**
• **Innovative Medicines Initiative - Qualification of Translational Safety Biomarkers**
• Partnership of pharmaceutical companies, academic centres, small business enterprises having open dialogue with regulatory authorities
• 5 year project started in June 2009
• 36M € ($44M) research budget
  – Funding from European Commission with in-kind contributions from Pharma
The SAFE-T Project Objectives

• To evaluate utility of safety biomarkers for monitoring organ safety in humans.
• To develop assays and devices for clinical application of safety biomarkers.
• To compile evidence to qualify safety biomarkers for regulatory decision-making in clinical drug development.
• To gain evidence for how safety markers may be used in disease diagnosis and in clinical practice (e.g. intensive care units).
Three areas of focus for safety markers

• **Drug-Induced Kidney Injury**
  – Serum Creatinine + BUN are significantly increased only when 50% of kidney function is lost.

• **Drug-Induced Liver Injury**
  – Transaminases are not specific and or predictive of who will recover vs. develop liver failure.

• **Drug-Induced Vascular Injury**
  – There are currently no clinical biomarkers to monitor vascular injury.

• **Overall objective of SAFE-T programs**
DIKI Biomarker Qualification Strategy

Biomarker step 1 list
- Literature
- Databases
- SAFE-T sources

Select

Biomarker step 2 list
- Evaluation

Select

Biomarker step 3 list
- Regulatory advice
- Assay availability / development
- Biomarker step 3 list
- Assay / stat analysis / select specific + sensitive BMs

Samples

Biomarker step 4 list
- Background variability
- Thresholds (ROCs)
- Regulatory advice
- Assay / stat analysis / select specific + sensitive BMs
- Biomarker step 4 list
- Assay / stat analysis / select specific + sensitive BMs

Current status

Confirmatory phase
- Regulatory advice
- Assay / stat analysis / select specific + sensitive BMs
- Biomarker final list

Qualification

Submit to health authorities

Regulatory approval

Q2 2009
- Q1 2010
- Q4 2012
- Q4 2014

SAFE-T Renal Safety Biomarkers ProjectERA-EDTA Meeting, Paris 2012 6
Overall DIKI Project Timelines

<table>
<thead>
<tr>
<th>2009</th>
<th>2010</th>
<th>2011</th>
<th>2012</th>
<th>2013</th>
<th>2014</th>
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- ♦ SAFE-T kick-off
- Biomarker selection process & Initial clinical plans
- Biomarker assay development/ validation
- Exploratory studies protocol preparation

Regulatory meetings ♦

- Acute GN study
- Contrast study
- Cisplatin study

Regulatory meetings ♦

- Confirmatory study

Regulatory submissions ♦
1. Selection process

Candidate biomarker selection

- Literature evidence
- Previous experience in rat studies
- Pharma company databases

Renal injury populations

- Review of drugs that cause renal injury
- Prevalence/feasibility/region of kidney injury
- Shortlist based on kidney region & feasibility
<table>
<thead>
<tr>
<th>Type of biomarker</th>
<th>Biomarker name</th>
<th>Main significance</th>
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<tbody>
<tr>
<td><strong>Functional biomarkers</strong></td>
<td>Microalbumin</td>
<td>Marker of impaired proximal tubular re-absorption</td>
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<td></td>
<td>α-1 microglobulin</td>
<td>Marker of impaired proximal tubular re-absorption (and indirectly glomerular injury)</td>
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<td></td>
<td>Cystatin C</td>
<td>Evaluation of glomerular filtration rate (serum)</td>
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<td>Retinol Binding Protein-4 (RBP-4)</td>
<td>Marker of impaired proximal tubular re-absorption</td>
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<td><strong>Tissue injury</strong></td>
<td>N-acetyl-β-D-glucosaminidase (NAG)</td>
<td>Marker of proximal tubular injury</td>
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<tr>
<td>leakage markers</td>
<td>Glutathione-S-transferase-α (GST-α)</td>
<td>Marker of proximal tubular injury</td>
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<tr>
<td></td>
<td>Glutathione-S-transferase-π (GST-π)</td>
<td>Marker of distal tubular injury</td>
</tr>
<tr>
<td></td>
<td>Liver-type fatty acid binding protein (L-FABP)</td>
<td>Marker of proximal tubular injury</td>
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<td></td>
<td>Collagen IV</td>
<td>Marker of glomerular injury</td>
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<tr>
<td></td>
<td>Podocin</td>
<td>Marker of glomerular injury</td>
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<tr>
<td></td>
<td>Nephrin</td>
<td>Marker of glomerular injury</td>
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<td>Aquaporin-2</td>
<td>Marker of collecting duct injury</td>
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<tr>
<td></td>
<td>Calbindin D28</td>
<td>Marker of injury to distal regions of nephron and collecting ducts</td>
</tr>
<tr>
<td><strong>Tissue injury</strong></td>
<td>Kidney injury molecule-1 (KIM-1)</td>
<td>Marker of proximal tubular injury/regeneration</td>
</tr>
<tr>
<td>response markers</td>
<td>Clusterin</td>
<td>Marker of tubular injury/regeneration (no apparent specific nephronal localization)</td>
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<td>Neutrophil gelatinase associated lipocalin (NGAL)</td>
<td>Marker of tubular (mainly proximal) injury</td>
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<td>Trefoil Factor 3 (TFF3)</td>
<td>Marker of proximal tubular injury</td>
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<tr>
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<td>Osteopontin</td>
<td>Marker of injury to distal regions of nephron</td>
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<td></td>
<td>Tissue inhibitor of metalloproteinase-1 (TIMP-1)</td>
<td>Marker of interstitial fibrosis and tubular injury</td>
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<tr>
<td></td>
<td>Connective Tissue Growth Factor (CTGF)</td>
<td>Marker of interstitial fibrosis</td>
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<td></td>
<td>Interleukin-18 (IL-18)</td>
<td>Marker of inflammation</td>
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<tr>
<td></td>
<td>Monocyte chemoattractant protein-1 (MCP-1)</td>
<td>Marker of inflammation</td>
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</table>
2. Exploratory studies

• Preparation for study conduct
  – Assay development
  – Setting up of biobank facility for clinical samples
  – Academic sites selected
  – eCRF design and database set-up

• Design of clinical studies
  – Renal injury studies
  – Control population studies
2. Exploratory phase: main studies

- **Baseline studies**
  - Healthy volunteer study
  - Chronic kidney disease study
  - Non-renal disease patient samples

- **Renal injury studies**
  - Proximal tubular damage studies
    - Cisplatin in cancer patients study
    - Contrast induced nephropathy study
  - Glomerular damage studies
    - Acute glomerulonephritis patient study

   - Establish normative range and variability of each marker
   - Longitudinal case control studies
   - Cross-sectional case control study

**NOTE:** Nephrotoxicity studies will be in patients receiving Standard of Care treatment
2. End of exploratory phase

• Results interpretation
  – Selection of biomarkers with good sensitivity & specificity
  – Setting of appropriate thresholds for injury

• Planning for confirmatory phase studies
  – Identifying appropriate populations
  – Optimising study designs (endpoints, sampling timepoints, sample size calculation, etc.)

• Interactions with Regulatory Agencies
  – Presenting data from exploratory studies
  – Sharing plans for confirmatory studies to gain buy-in
3. Confirmatory studies

- Confirmatory phase 2013-14
  - Intent is to conduct 1-2 confirmatory studies
  - Choice of populations and studies TBD
    - proximal conv. tubular ± glomerular injury study(ies)
    - co-ordinate with PSTC to avoid duplication of effort
  - Study design(s) based on exploratory study results
3. Confirmatory phase: other studies

- **Baseline studies**
  - Additional healthy volunteer samples
  - Non-renal disease patients studies

- **Specificity studies**
  - Organ injury studies done as part of liver and vascular injury SAFE-T projects

- **Supportive studies**
  - Renal biopsy study in transplant patients
  - *Study in patients in ICU setting?*
  - *Other supportive studies?*

Studies started in exploratory phase but main body of work will be conducted in confirmatory phase.
Exploratory Phase Studies
Healthy Volunteer Study

• Single centre, non-drug study: completed
• Design:
  – 25 healthy subjects
    • 12 male, 13 female subjects:
      – 6+7 subjects 18-45 years old
      – 6+6 subjects 46-65 years old
  – 3 study periods
    • Day 0, Day 7, Day 28
    • In each period:
      – 6 blood samples collected over 24H
      – 1 spot urine plus urine collections over 24H (0-4, 4-12, 12-24h)
    • Blood analysed for serum creatinine, BUN, serum cystatin C
    • Urine samples analysed for all urinary biomarkers
• Assay work ongoing: results expected 3Q2012
Renal Injury Studies: Objectives

– To collect blood and urine samples in target population and control subjects.
– To characterise between-and within-subject variability of novel biomarkers vs. BUN/ serum creatinine.
– To compare patterns of novel biomarker changes relative to BUN/ serum creatinine to:
  • select candidate biomarkers to progress to confirmatory stage and establish cut-off values for these biomarkers.
  • characterise the time course of biomarker changes to optimise the study design of confirmatory studies.
Cisplatin Study

Populations

• **Group A**: patients with various cancers who are scheduled to start high dose cisplatin therapy.
  \[N=100\]
  [20 subjects enrolled to date]

• **Group B**: control patients with similar cancers treated with local radiotherapy or non-nephrotoxic drugs.
  \[N=20\]
  [18 subjects enrolled to date]

• **Group C**: non-treatment healthy volunteers.
  \[N=20\]
  [25 subjects enrolled]

• **Ongoing study: anticipated completion 1H2013**
Cisplatin Study Design

Patients with cancer due to receive cisplatin chemotherapy as Standard of Care

Pre-Tx  Cis.  Post-1st cycle of cisplatin

Urine & blood samples
(BUN/ s creatinine, serum and spot urine samples for novel markers)

Control subjects: two samples taken 4 days apart
Populations

• **Group A**: patients with symptoms of acute GN and renal biopsy-confirmed diagnosis.  
  \( N=100 \) patients with confirmed acute GN  
  [71 subjects enrolled to date]

• **Group B**: control patients with chronic renal impairment due to polycystic kidney disease.  
  \( N=20-50 \)  
  [32 subjects enrolled to date]

• **Group C**: healthy volunteers.  
  \( N=20 \)  
  [25 subjects enrolled]

• **Ongoing study**: anticipated completion 3Q2012
Patients presenting with symptoms suggestive of acute GN

Renal biopsy

Symptom onset

Note: no baseline sample

Urine & blood samples within 3-6 month period (BUN/ s creatinine, serum and spot urine samples for novel markers)

Control subjects will have 2 samples taken over 2-4 week period
Contrast-Induced Nephropathy Study

Populations

- **Group A**: High-risk subjects: patients with chronic renal impairment and 1 other factor predisposing to CIN and scheduled for coronary angiography.  
  \[N=200 \text{ patients}\]  
  [86 subjects enrolled]

- **Group B**: Low-risk subjects: patients scheduled for contrast radiology study at low risk of developing CIN.  
  \[N=20 \text{ patients}\]

- **Group C**: non-treatment healthy volunteers.  
  \[N=20\]  
  [25 subjects enrolled]

*Ongoing study: anticipated completion 4Q2012*
Patients scheduled to undergo contrast injection as part of planned radiological investigation.

Contrast administration

B/L Post-contrast administration

- Urine & blood samples
  - (BUN/ s creatinine, serum and spot urine samples for novel markers)

Control subjects: 2 samples taken 4 days apart
Chronic Kidney Disease Study

- Supportive study – will continue into confirmatory phase
- Main objective
  - Collect blood and urine samples in CKD patients.
- Study population
  - $N = 200$ patients with diabetic nephropathy.
- Study design
  - Subjects are participating in a Phase 2 Pharma drug study.
  - $1^{st}$ sample taken at baseline before start of randomised treatment
  - $2^{nd}$ sample taken 2 weeks post-cessation of randomised treatment (drug will have washed out by this time)
- Samples will be analysed for novel biomarkers
- *Ongoing study: anticipated completion 4Q2012*
  [130 subjects enrolled to date]
Renal Transplant Biopsy Study

• Supportive study – will continue into confirmatory phase
• Main objective
  – correlate DIKI biomarkers and renal histopathological findings.
• Study population
  – \( N = 400 \) post-renal renal transplant patients.
    • patients scheduled to have a renal biopsy
      – Routine biopsy
      – Biopsy to determine cause of potential graft failure
• Study design
  – eligible patients have blood and urine samples taken prior to biopsy on day of planned procedure
• Endpoints
  – DIKI biomarker patterns correlated to renal biopsy findings
• **Ongoing study: anticipated completion 2014**
  [50 subjects enrolled to date]
Summary

• Consortium-based approach to safety biomarker qualification working with Regulatory Agencies and academic community
• Novel kidney biomarkers of interest chosen with new assays developed as necessary
• First healthy volunteer study completed with additional samples collected in other studies
• Three exploratory phase studies are ongoing to assess renal markers of glomerular damage and renal tubular injury
Next Steps

- Completion of exploratory studies
- Analysis of novel biomarker data and determine which are appropriate to test in confirmatory phase
  - Interactions with PSTC to align strategies
- Design of confirmatory studies with Regulatory Agency advice