Improving the prediction of disease progression: The search for improved markers of CKD

Division of Nephrology
30th Arnold O. Beckman Conference
2013 Kidney Week, Atlanta, GA
November 5-6, 2013
Chi-yuan Hsu, MD, MSc
Professor and Chief
UCSF Division of Nephrology

Disclosures
• Urine biomarker assays (NGAL) donated Abbott (in CKD Biomarker Consortium)

Topics
• Examining the role of “AKI biomarkers” in CKD progression
• Lessons learnt
• Future opportunities
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• Lessons learnt
• Future opportunities

Recent data: AKI accelerates progression/development of CKD

Amdur KI 2009
AKI-CKD connection

• “the AKI-CKD connection is of far reaching clinical significance....Identification of the AKI-CKD nexus represents the single most important advance in understanding of the mechanisms of progression since hyperfiltration was shown to occur following renal ablation.”

Venkatachalam Am J Physiol Renal Physiol 2010

Old paradigm: The trajectory of CKD is relatively smooth

New paradigm: The course of CKD is punctuated by episodes of AKI
Motivation for this line of investigation

- If clinically evident cases of AKI accelerate the progressiondevelopment of CKD, are more subtle episodes of injury—detected by "AKI biomarkers"—also important?
- Candidate injury biomarkers include neutrophil gelatinase-associated lipocalin (NGAL) and kidney injury molecule-1 (KIM-1)

Neutrophil gelatinase-associated lipocalin (NGAL)

"AKI biomarkers" are detectable in CKD

- Urine NGAL measured in baseline urine samples among 3386 Chronic Renal Insufficiency Cohort (CRIC) study participants (mean eGFR 42 ml/min/1.73m²; 1.1 gm proteinuria)
- Median concentration 17.2 ng/mL (IQR 8.1-39.2 ng/mL)(5% between 178.9-3069.6 ng/mL)
- Higher NGAL concentration among those who are female, non-white, with lower eGFR, higher proteinuria, DM, higher BP
**Higher urine NGAL an independent risk factor for ESRD/eGFR halving**

<table>
<thead>
<tr>
<th>Quartiles of baseline urine NGAL concentration (ng/ml)</th>
<th>Events</th>
<th>Rate (per 100 person-years)</th>
<th>Unadjusted HR (95% confidence interval)</th>
<th>Additionally adjusted for other baseline covariates</th>
</tr>
</thead>
<tbody>
<tr>
<td>≤ 0.9</td>
<td>47</td>
<td>1.9</td>
<td>Ref</td>
<td>Ref</td>
</tr>
<tr>
<td>&gt; 0.9 to &lt; 12.9</td>
<td>77</td>
<td>3.2</td>
<td>1.75 (1.22-2.51)</td>
<td>1.37 (1.04-1.80)</td>
</tr>
<tr>
<td>&gt; 12.9 to &lt; 22.6</td>
<td>105</td>
<td>4.8</td>
<td>2.52 (1.79-3.56)</td>
<td>1.24 (0.88-1.70)</td>
</tr>
<tr>
<td>&gt; 22.6 to &lt; 49.3</td>
<td>173</td>
<td>8.1</td>
<td>4.29 (3.11-5.09)</td>
<td>1.39 (0.97-2.00)</td>
</tr>
<tr>
<td>&gt; 49.5</td>
<td>267</td>
<td>10.9</td>
<td>5.23 (3.58-7.27)</td>
<td>1.70 (1.18-2.40)</td>
</tr>
<tr>
<td>HR per 1 unit increase in log (urine NGAL)</td>
<td>1.75 (1.66-1.85)</td>
<td>1.11 (1.01-1.22)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

* age, sex, race/ethnicity, eGFR, 24-hr urine protein, DM, SBP, BMI, use of ACE-I/ARB, education attainment, hx of CVD

Liu KI 2013

**Effect strongest in the first 2 years of follow-up**

![Figure 2](image)

Category-free NRI 24.7% (0.4 to 38.5%)

ARIC study

[FR-PO275] Urinary Biomarkers as Risk Factors of End-Stage Renal Disease in the General Population: CKD Biomarkers Consortium

Meredith C. Foster, Josef Coresh, MD, PhD, FASN, Joseph V. Bonventre, MD, PhD, FASN, Chi-yuan Hsu, MD, Paul L. Kimmel, MD, FASN, Theodore E. Mifflin, PhD, Robert G. Nelson, MD, PhD, Vasan S. Ramachandran, Venkata Sabbisetti, PhD, Sushrut S. Waikar, MD, Kathleen D. Liu, MD.
Median time between biomarker measurement and ESRD event = 6 years

KIM-1/Cr
| Tertile 1 | Reference | 0.87 (0.44-1.72) |
| Tertile 2 | 0.96 (0.48-1.94) |
| Tertile 3 | 1.06 (0.77-1.45) |

Per SD

NAG/Cr
| Tertile 1 | Reference | 0.69 (0.28-1.70) |
| Tertile 2 | 1.83 (0.75-4.44) |
| Tertile 3 | 1.72 (1.11-2.68) |

Per SD

NGAL/Cr
| Tertile 1 | Reference | 3.62 (1.48-8.85) |
| Tertile 2 | 7.10 (2.88-17.51) |
| Tertile 3 | 1.85 (1.38-2.50) |

Per SD

LFABP/Cr
| Detected vs. undetectable | 0.79 (0.39-1.57) |

ESRD Hazard ratios adjusted for DM duration, demographics, HTN, HbA1c, GFR, ACR

Median time between biomarker measurement and ESRD event = 9 years

Pima Indians

[FR-PO278] Association of Urinary LFABP, KIM-1, and NAG with Incident End-Stage Renal Disease and Mortality in Type 2 Diabetes

Gudeta D. Fufaa, PhD, E. Jennifer Weil, MD, Robert G. Nelson, MD, PhD, Joseph V. Bonventre, MD, PhD, FASN, Venkata Sabbisetti, PhD, Sushrut S. Waikar, MD, Theodore E. Mifflin, PhD, Xiaoming Zhang, Dawei Xie, Harold I. Feldman, MD, FASN, Josef Coresh, MD, PhD, FASN, Vasan S. Ramachandran, Paul L. Kimmel, MD, FASN, Chi-yuan Hsu, MD, Kathleen D. Liu, MD.
Summary

• Consist with our hypothesis, some “AKI biomarkers” are independent risk factors of more rapid progression of CKD
• Little improvement in prediction of outcome events/reclassification
• Insight: effect is most notable with a 1-2 yr time horizon?
• This may make sense patho-physiologically and would need to be considered in future studies

• Examining the role of “AKI biomarkers” in CKD progression

• Lessons learnt
  • Future opportunities
1. Importance of urine handling

[FR-PO023] Urine Stability Studies for Novel Biomarkers of Acute Kidney Injury

Chirag R. Parikh, MD, PhD, FASN, Isabel Butrymowicz, MD, Angela Yu, Vernon M. Chinchilli, PhD, Meyeon Park, MD, Chi-yuan Hsu, MD, William Brian Reeves, MD, FASN, Prasad Devarajan, MD, Paul L. Kimmel, MD, FASN, Edward D. Siew, MD, Kathleen D. Liu, MD.

In press AJKD; Haase-Fielitz AJKD 2009

2. Importance of analytical chemistry lab QC

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<table>
<thead>
<tr>
<th>Step</th>
<th>Random error</th>
<th>Systematic error</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.50</td>
<td>Reject</td>
<td>Accept</td>
</tr>
<tr>
<td>1.20</td>
<td>Yes</td>
<td>No</td>
</tr>
<tr>
<td>1.00</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>0.80</td>
<td>Yes</td>
<td>Rejct</td>
</tr>
<tr>
<td>0.60</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>0.40</td>
<td>Yes</td>
<td>Rejct</td>
</tr>
<tr>
<td>0.20</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>0.00</td>
<td>Yes</td>
<td>Yes</td>
</tr>
</tbody>
</table>
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"The Westgard multirule system"
“Informatics” problems

- Misunderstanding: data transmitted incorrectly in part because the working sample manifest sent to performance lab was INVERTED relative to the sample packing order.
4. Importance of constant vigilance

![Graph showing correlation between Old Phoenix UALB (mg/L) and New UPENN UALB (mg/L).](image)

*Pearson r = 0.6357*

*Courtesy: Robert Nelson, MD, NIDDK*

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**water loss = old UCr/new UCr**

![Graph showing water loss calculations.](image)

*Courtesy: Robert Nelson, MD, NIDDK*
Corrected for Water Loss

\[
\text{Corrected for Water Loss}
\]

\[ r = 0.9362 \]

Courtesy: Robert Nelson, MD, NIDDK

Diagnosis: desiccation

- Problem with urine stored for ~20 years at -80°C when not using cryovials with gaskets
- Outcome: express urine biomarkers as per gm urine creatinine
- Measure urine biomarkers after dilution (1:4) (*sticky* samples with bead aggregation on Luminex platform)
5. Importance of blind replicates

Blind replicates biomarker/Cr ratios were better correlated.
Diagnosis: incomplete mixing

- After more investigation, including measuring other daughter tubes, we concluded that the problem was due to incomplete mixing of the large 9 cc mother tube
- Outcome: about one-quarter of the samples require re-aliquoting in order to have daughter tubes suitable for biomarker studies

- Examining the role of “AKI biomarkers” in CKD progression
- Lessons learnt
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Much remains to be learned

<table>
<thead>
<tr>
<th></th>
<th>ARIC controls (N=192)</th>
<th>Pima normo-albminuria group (N=138)</th>
</tr>
</thead>
<tbody>
<tr>
<td>DM (51%)</td>
<td>100%</td>
<td></td>
</tr>
<tr>
<td>GFR (70 ml/min/1.73m²)</td>
<td>153 ml/min (measured)</td>
<td></td>
</tr>
<tr>
<td>Urinary Alb/Cr</td>
<td>13 [3-108]</td>
<td>8 [5-14]</td>
</tr>
<tr>
<td>(median [IQR] [mg/gm])</td>
<td></td>
<td></td>
</tr>
<tr>
<td>KIM-1/Cr (median [IQR] [ng/gm])</td>
<td>659 [361-1047]</td>
<td>449 [169-997]</td>
</tr>
<tr>
<td>NGAL/Cr (median [IQR] [μg/gm])</td>
<td>15 [9-40]</td>
<td>44 [18-80]</td>
</tr>
</tbody>
</table>

48,242 aliquots made (34,954 in CKD BioCon central lab)

4cc 4cc 4cc 4cc 4cc 4cc
Within a prospective cohort study, a nested case-cohort design is an efficient way to validate novel biomarkers longitudinally.

Summary

- Some signal that “AKI biomarkers” are important with regard to the progression of CKD (especially within a 1-2 yr time horizon)
- There are many factors that need to be considered in order to optimally test the performance of novel biomarkers in large scale human studies
- Much remains to be learned but future studies may be made easier as a result of the infrastructure built by groups such as the CKD Biomarker Consortium

Acknowledgement

- NIH-NIDDK
- Kathleen Liu