Cardiovascular Implications of Proteinuria

B. S. Kasinath, MD
University of Texas Health Science Center
South Texas Veterans Health Care System
San Antonio

Objectives

• Mechanisms of albumin handling by the kidney

• Discuss the CV implications of proteinuria in non-Chronic Kidney Disease patients and possible interventions

No disclosures
Glomerular ultrafiltration

**Glomerulus**
- Unique capillary interposed between 2 arterioles
- Podocytes
- Endothelial cells
- Mesangial cells
- Role of resistance vessels

Proteinuria – mechanisms

**Glomerulus:** a size and charge selective barrier

**Role of podocyte**
- Slit diaphragm
- Diabetic nephropathy, Gene mutations, e.g., NPHS1 (nephrin), NPHS2 (podocin)

**Role of glomerular endothelial cells**
- Anionic glycocalyx: charge barrier
- Loss of fenestrae, impaired glycocalyx
- Diabetic nephropathy, Pre-eclampsia, TTP, HUS

Human glomerular proteome contains 144 matrix proteins: contribution to proteinuria?

**Proteinuria - mechanisms**

**Proximal Tubule Reabsorbs Filtered Albumin**
- Megalin, Cubilin system – brush border
- Endocytosis and delivery of cargo to lysosomes for degradation
- Recycling of Megalin and Cubilin
- Reabsorbed Proteins: Albumin, Carrier proteins and others
- Estimate in humans: ~0.5-1 g albumin filtered & absorbed
- PT-specific Meg or Cub KO: albuminuria, low mol. wt. proteins
- Other albumin transporters: Neonatal Fc receptor

**Toxic effects of proteinuria on Proximal Tubular Cells**
- Complement activation, Inflammation, TGFβ
- Activation of RAAS, fibroblast stimulation
- Renal interstitial fibrosis: CKD progression

---

**Proteinuria**

- Urinary proteins: Albumin, Uromodulin (T H protein), traces of other proteins
- Pathologic proteinuria: albumin, Igs, myoglobin, light chains
- Dipstick + protein: loss of renal function >5%/year from baseline (Clark)
- Albuminuria: Urinary Albumin Creatinine Ratio (ACR)
- ACR in CKD: >30 mg/g
- Staging of CKD now coupled to ACR (A1 0-30, A2 30-300, A3 >300 mg/g)

**Focus: Albuminuria in Cardiovascular Disease (CVD)**

Clark WF, J Am Soc Nephrol 22: 1729, 2011,
**Albuminuria – Implications for CVD**

What is the rationale for looking at albuminuria in CVD?

1. Accurate **risk assessment of CVD** in general population is critical for optimal preventive and therapeutic strategies.

2. Even after identifying and treating ‘traditional’ risk factors there is significant residual morbidity and mortality due to CVD.

3. This suggests there must non-traditional risk factors.

4. We need for new biomarkers: fit for large scale screening accessible.

5. **ACR** measurement is widely available and a good candidate for examination.

---

**Albuminuria**

**Urinary albumin excretion in the General Population**

<table>
<thead>
<tr>
<th>7265 subjects from Japan</th>
<th>11,392 subjects, ARIC study from US</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Urinary ACR mg/g</strong></td>
<td><strong>Percent</strong></td>
</tr>
<tr>
<td>&lt;5</td>
<td>53.2%</td>
</tr>
<tr>
<td>5-10</td>
<td>21.8%</td>
</tr>
<tr>
<td>11-20</td>
<td>8.7%</td>
</tr>
<tr>
<td>21-30</td>
<td>2.6%</td>
</tr>
<tr>
<td><strong>CKD</strong></td>
<td></td>
</tr>
<tr>
<td>30-300</td>
<td>4.6%</td>
</tr>
<tr>
<td>&gt;300</td>
<td>0.5%</td>
</tr>
</tbody>
</table>

Undetectable urinary albumin in 8.8%

80% of population: ACR is <10 mg/g

CKD range ACR in 5.1%

---


---

This presentation is the intellectual property of the author. Contact them for permission to reprint and/or distribute.
Albuminuria and CVD

Studies in General Population

- ACR >30 mg/g: independent risk factor for CVA, MI, death and predicts future CHF
- Is non-CKD level of ACR associated with diseases?

- HOPE study: 3498 diabetic and 5545 non-diabetic, Follow up 4.5 years, Microalbuminuria (ACR >18mg/g): 30% diabetics, 15% non-diabetics

- Analysis of ACR in deciles: An Independent Continuous Risk Factor (well below CKD definition) for CVD, All Cause Mortality and CHF admission after adjusting for other risk factors

- ? Confirmation by other studies

Gerstein HC, JAMA 286: 421, 2001

Albuminuria and Mortality

Studies in General Population

- Meta-analysis of 21 large studies (international)
- 1.2 million subjects, F/U 8 years, age 61 years
- Baseline ACR Or dipstick for protein vs. All Cause or CV mortality (hazard ratio, HR)
- Baseline eGFR vs. All Cause or CV Mortality
- 68% of subjects had ACR <10 mg/g

- After adjusting for age, sex, ethnicity, h/o CVD, BP, diabetes, smoking, total cholesterol
  - CV and AC mortality progressively rose beyond ACR of 5 mg/g
  - Note increased death WITHIN “normal” range of ACR (5 vs. 10 vs. 30)

Matsushita K, Lancet 375: 2073, 2010

This presentation is the intellectual property of the author. Contact them for permission to reprint and/or distribute.
Albuminuria and Mortality

Studies in General Population

- Adjusted for age, sex, ethnicity, h/o CVD, BP, diabetes, smoking, total cholesterol
  - Greater ACR is associated with greater mortality at each eGFR range including in the 90-120 range
- Dipstick + Proteinuria
  - Even trace proteinuria increases HR for All Cause and CV mortality across eGFR categories

Conclusions of Matsushita study

1. ACR/dipstick + proteinuria are independent risk factors for All Cause & Cardiovascular mortality
2. eGFR is also an independent risk factor
3. Is ACR of 5 mg/g the threshold for predicting Cardiovascular and All Cause mortality?

Issues:
- No uniform measurement of albumin, creatinine and protein among studies employed in meta-analysis
- Reliance on dipstick

?Evidence from a study that did not have these issues?

Matsushita K, Lancet 2010; 375: 2073, 2010
Albuminuria and CVD in General Population

Studies in General Population (South Korea) - Single Center study
- 37091 people, 53% men, mean age 46, F/u 5 yrs, All had ACR below 30 mg/g
- Analysis done after adjusting for smoking, obesity, education, eGFR
- Albuminuria in the normal range (<30 mg/g) divided into quartiles

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>Overall</th>
<th>ACR Quartiles</th>
<th>P for Trend</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Q1 (&lt;3.4 mg/g)</td>
<td>Q2 (3.4-4.7 mg/g)</td>
</tr>
<tr>
<td>N=7091</td>
<td></td>
<td>9274</td>
<td>9274</td>
</tr>
<tr>
<td>Age, y</td>
<td>45.8 (11.9)</td>
<td>42.6 (10.7)</td>
<td>44.5 (11.3)</td>
</tr>
<tr>
<td>BMI, kg/m²</td>
<td>23.6 (3.2)</td>
<td>23.7 (2.9)</td>
<td>23.9 (3.6)</td>
</tr>
<tr>
<td>Systolic BP, mm Hg</td>
<td>115.9 (14.9)</td>
<td>113.9 (12.5)</td>
<td>113.7 (13.7)</td>
</tr>
<tr>
<td>Diastolic BP, mm Hg</td>
<td>74.5 (9.9)</td>
<td>73.8 (8.9)</td>
<td>73.7 (9.5)</td>
</tr>
</tbody>
</table>

- Note: 75% of people had ACR less than 7.4 mg/ g creatinine
- BMI and higher BP associated with higher quartiles within this ‘normal’ range of ACR

Albuminuria in normal range and mortality
- Adjusted for age, sex, smoking, obesity, education, alcohol, exercise, BMI, HT, DM, history of CVD, HDL/ LDL cholesterol, eGFR
- Highest quartile (ACR >7 mg/g): 3.4-fold higher risk for CV mortality
- **CVD mortality: Note rising risk from Q1 through Q4 even in this normal range - ?a dose effect**
- No threshold: continuous risk factor
- No such risk for All Cause mortality

Sung K-C, J Am Heart Assoc 5: e003245, 2016
**Albuminuria and CVD in General Population**

Risk for Hypertension (9102 subjects followed for 3 years)
- Adjusted for age, sex, smoking, obesity, education, alcohol, exercise, BMI, HT, HDL/ LDL chol, eGFR
- Highest quartile (ACR 7 ug/g):
  - 1.97-fold higher risk for HT
- *Note rising risk across the quartiles suggests a dose effect*

No Risk for Diabetes (10930 subjects followed for 3 years)

---

**Albuminuria, CVD in General Population**

Problems with general population studies
- Urinary ACR commonly measured once – not confirmed on repeat test
  - Does not account for diurnal variation, factors such as exercise, fever
  - Variability of albuminuria is high; false + 30-50%
  - However, large numbers of subjects mitigate variability
- BP data based on one time measurement
- eGFR estimates are not reliable in the range seen
- Is ACR is risk factor for CVD and mortality? Still open

---

Sung K-C, J Am Heart Assoc 5: e003245, 2016

NephSAP 2016
**Albumin – does it cause microvascular injury?**

*Why is albuminuria associated with CVD?  Possible scenarios*
- Just a marker for associated CVD risk factors HT, diabetes, CKD, dyslipidemia
- Steno hypothesis: Albuminuria associates with global dysfunction of endothelium
- Albumin is directly injurious to blood vessels

*Potential causative role of Albumin*
- Normally, Albumin scavenges ROS
- Oxidation of albumin occurs on Cys-34
- Circulating plasma oxidized albumin correlates with CVD (Kaneda, Lim)
- Atheroma proteomics: albumin is abundant in the plaque (Lepedda 2009)
- Oxidative environment in Atheroma:
  - Plaque albumin: Cys-34 oxidation 3-fold higher vs. plasma albumin

*These data show:*
- Albumin enters into the atherosclerotic plaque and it is highly oxidized

Kaneda H, Atherosclerosis 162: 221, 2002; Lim P-S, PLOS ONE 8: e70822, 2013

---

**Albuminuria – does it cause microvascular injury?**

*Toxic effects of albumin*

*Endothelial cells exposed to Oxidized Alb*
- Increased VCAM-1, ICAM-1 expression
- Entry of monocytes into vessel wall
- Increased ROS generation; apoptosis
- Oxidative stress in endothelial cells

*Prox Tub Epithelial cells – albumin overload*
- mTORC1 activn, NFkB, infl. chemokines

- Albuminuria may be associated with albumin entry into the vessel wall
- Circulating oxidized albumin may injure endothelial cells & promote monocyte entry
- Oxidized Albumin in the vessel wall may contribute to atherosclerosis


---

This presentation is the intellectual property of the author. Contact them for permission to reprint and/or distribute.
Conclusion 1

• Albuminuria – could be a continuous independent risk factor for cardiovascular disease and mortality

• Classifying albuminuria as ‘normal’ under 30 mg/g may be debatable with regards to Cardiovascular Disease

• Methodological issues should be settled before future studies are done, e.g., assay procedure, collection time, repeat test for confirmation – timing of repeat ACR

Conclusion 2

Should we measure albuminuria as risk factor for CVD?

• Need prospective studies to show improved risk stratification for CVD

• Management:
  • Need data from prospective trials that show improved CVD outcomes when albuminuria is treated (ACE inhibitors, ARBs)
  • Otherwise, large segments of population with low albuminuria will be exposed to the risks of toxicity and costs

The value of albuminuria in CVD risk prediction and management needs further examination
Thank You

Albuminuria and CVD

*Albuminuria and Cardiovascular Disease in Hispanic Americans*

- Echo-Sol Study - 1815 subjects, 4 sites
- Normal (42%), High normal (<17mg/g) (43%), microalbuminuria (13%), macroalbuminuria (>250 mg/g) (2%)
- Micro- and macro-albuminuria associated with LVH on echo
- High normal and above: diastolic dysfunction

Hanna DB, Am J Cardiol, March 2017, [http://dx.doi.org/10.1016/j.amjcard.2017.03.039](http://dx.doi.org/10.1016/j.amjcard.2017.03.039)

This presentation is the intellectual property of the author. Contact them for permission to reprint and/or distribute.
Mechanisms of Endothelial Injury in Diabetes

ACR and CVD

- Since ACR is a continuous risk factor, it will not discriminate between people at risk for CVD vs. those who do not.
- Not so; the HR increases as ACR increases. Thus, one can identify those above the reference range (e.g., <3 mg/g) and test for CVD association.
- >3 mg/g range will cover millions of people.
- Proof needed to show reducing albuminuria ameliorates injury in non-renal blood vessels that develop atherosclerosis.

Mechanisms of Endothelial Injury in Diabetes

- Loss of fenestrae and glycocalyx
- Macrophage secretion of cathepsin-S, activation of protease-activated receptor -2, results in albuminuria
- VEGF: variable role in early vs. late stages
- NO deficiency: High glucose scavenges NO leading to inflammation, platelet aggregation, reduced angiogenesis, TGFβ activation, coagulation
- Uncoupling of eNOS: formation of peroxinitrite
- AGE induced senescence, rescued by SIRT-1

Goligorsky, M, Am J Physiol Renal Physiol 312: F466, 2017
Unused slides