ROLE OF NOVEL BIOMARKERS IN DISEASE PREVENTION: CRP, CT CALCIUM, AND CYSTATIN C

Michael G. Shlipak, MD, MPH
Professor of Medicine, University of California, San Francisco
Chief, Division of General Internal Medicine, San Francisco VA Medical Center

October 27th, 2010

Introduction

- Biomarker - new term to represent a measureable biological process
- Biomarkers most often are blood tests, but can be from DNA, imaging, ECGs, etc.
- Biomarkers have major scientific roles other than clinical practice.
- Many primarily used in epidemiology to understand etiology of disease onset and progression

Hlatky, Circulation 2009

- Useful diagnostic tests - change our clinical management by:
  - Distinguishing the presence/absence or severity of disease
  - Changing our perception of disease risk, with clinical implications
- Modifiable Risk Factors - information is useful, as provides targets for intervention to reduce disease risk
Non-Modifiable Risk Factors- information changes individual's likelihood of disease, but does not provide specific target for therapy.

Value of prognostic biomarkers is controversial—many new tests have unclear action plans.

Question 1: Which of the following best describes your attitude toward new diagnostic tests?

1. I am usually excited to use new tests to detect disease or disease risk.
2. I actively learn about new tests because my patients or peers expect me to be knowledgeable.
3. I generally don’t like learning about new tests because they rarely improve clinical care.
4. If a test was not part of my clinical training, then I probably won’t order it.

Outline

Update on Key Biomarkers:
- C-Reactive Protein (CRP)
- Coronary calcification by CT and other subclinical CVD measures
- Cystatin C
Question 2: My practice regarding CRP is best described as:

1. I measure CRP routinely to help with CVD risk stratification.
2. I measure CRP occasionally for challenging decisions for CVD prevention.
3. CRP is often measured as part of a lipid panel, but I rarely use the information.
4. I never think about CRP.

C-Reactive Protein (CRP)

- Blood test that captures systemic inflammation
- Probably not in disease pathway; triggered by IL-6
- CRP is very stable and easy to measure in blood
- Measuring inflammation has never been standard screening test

CRP as a Risk Factor

- Emerging Risk Factors Collaboration Meta-Analysis:
  - 38 studies
  - 160,309 individuals
  - 1.31 M person-years
  - 27,769 outcomes

  Koptoge S et al. Lancet, 2009
CRP and Heart Disease Risk

Koptoge S et al. Lancet 2009

RR for 3-fold higher CRP-Mortality

Koptoge S et al. Lancet 2009

Is the CRP-CVD Association Strong Enough?

- Although CRP is an “independent” risk factor, it does not add much beyond Framingham Risk Equation
- Explained partly by CRP’s correlation with other cardiovascular risk factors
- CRP’s relative risks only about 2-fold; to improve Framingham Risk Estimates, new risk factor needs $RR \geq 3.5$

Lloyd-Jones DM et al., Ann Intern Med 2006
CRP for Risk Classification

- CVD risk is classified as:
  - Low: < 10
  - Medium: 10-20
  - High: > 20 risk equivalent
  - 0-1 risk factors: ≥ 2 risk factors

- Can CRP help classify correctly?
- Some have advocated CRP testing to help separate persons with medium risk
  
  Ridker PM et al., NEJM 2002 and Ann Intern Med 2009

Adding CRP to Framingham Risk Assessment in Women

- CRP Level < 1.0 mg/L
- CRP Level 1.0-3.0 mg/L
- CRP Level >3.0 mg/L

- Multivariate Relative Risk for CVD:
  - 0%-1%
  - 2%-4%
  - 5%-9%
  - ≥10%

Lloyd-Jones DM et al., Ann Intern Med 2006

Should CRP Levels be Used to Identify Statin Candidates?

- JUPITER Trial
  - Design: 17,802 adults without CAD
  - Rosuvastatin 20mg vs placebo
  - Inclusion criteria:
    - LDL < 130 (average 108)
    - CRP > 2 (average 4.2)
  - Outcomes: CVD, death
JUPITER Rosuvastatin Decreases CVD Risk

Key Points on JUPITER

- Statin trial, not a CRP study
- Demonstrated that statins are beneficial even in low-risk adults with normal LDL
- Statins likely also beneficial for persons with CRP < 2.0
- Therefore, no clear justification to measure CRP

USPTS Summary on CRP

- CRP - the most rigorously studied of novel CVD biomarkers
- CRP may classify some medium risk persons as high risk, but unclear:
  - the yield of CRP testing
  - why not aggressively treat all med. risk?
- "Current evidence does not support the routine use of CRP for risk stratification"

Helfand M et al., Ann Intern Med 2009
Update on Key Biomarkers:
- C-Reactive Protein (CRP)
- Coronary calcification by CT and other subclinical CVD measures
- Cystatin C

Question 3: The use of CT scans for CAD should be used:
1. As an alternative to coronary angiography (cath) for low risk patients.
2. To identify patients with CAD.
3. To decide who to give statins to.
4. There are no compelling indications to measure CT calcification.

Coronary Calcification: Introduction
- CT scans have ability to detect calcium deposits in coronary (and other) arteries
- Presence of calcification is an indication of atherosclerotic plaque
- Therefore, detectable calcification in coronary arteries is an indicator of subclinical cardiovascular disease
Subclinical Cardiovascular Disease

- Defined as procedures (often radiological imaging) that detect early cardiovascular disease before clinically apparent
- Not developed as screening tests, but rather to study the development of CVD processes

The Role of Subclinical CVD Measures

- Used to isolate risk factors, such as diabetes, for specific pathological processes that lead to CVD
- Often used as clinical trial outcomes for Phase 2 studies
- All valid subclinical CVD measures are associated with clinical CVD events.

Current Subclinical CVD Measures

- Coronary artery calcification (CT)- MI risk
- Left ventricular hypertrophy (echo, MRI)- heart failure
- Carotid wall thickness (IMT by u/s)- stroke
- Ankle brachial index (SBP ratio)- peripheral arterial disease

Processes overlap, so most subclinical CVD measures predict multiple CVD outcomes
CT Coronary Artery Calcification Score
and CAD Events - the MESA Study

![Graph showing coronary artery calcification score and CAD events over time.]

Detrano R et al., NEJM 2008

CAC Score and CAD Events by Race

<table>
<thead>
<tr>
<th>Racial or Ethnic Group</th>
<th>Major Coronary Event</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Hazard Ratio (95% CI)</td>
</tr>
<tr>
<td>White</td>
<td>1.17 (0.96-1.23)</td>
</tr>
<tr>
<td>Chinese</td>
<td>1.22 (0.95-1.58)</td>
</tr>
<tr>
<td>Black</td>
<td>1.35 (1.16-1.57)</td>
</tr>
<tr>
<td>Hispanic</td>
<td>1.13 (0.92-1.39)</td>
</tr>
</tbody>
</table>

* CAC score is the number of calcium scores and CI indicates confidence interval. Risk factors were calculated with the use of the regression for coronary heart disease major event as any event for cardiovascular disease (CVD) death, fatal and non-fatal stroke, fatal and non-fatal myocardial infarction, and CVD hospitalization. Hazard ratios are calculated on the basis of modeling of CAC.

Detrano R et al., NEJM 2008

The Pros and Cons of CT Screening
(or other testing for subclinical CVD)

- **Pros:**
  - Stronger risk associations than CRP or other blood tests
  - Specific for the presence of atherosclerosis
  - Could potentially be useful for medium risk individuals needing motivation (e.g. my brother-in-law)
  - ABI is inexpensive, relatively
The Pros and Cons of CT Screening (or other testing for subclinical CVD)

- **Cons:**
  - In older populations and those with diabetes or kidney disease—nearly universal
  - Not reproducible enough for repeat testing
  - Not directly modifiable
  - Radiation exposure
  - $$$

Conclusion on CT-Calcium Score (or other testing for subclinical CVD)

- USPTF: current evidence does not support routine use of CT
  - Also, does not recommend routine use of ankle brachial index or carotid wall thickness
    
    Helfand M et al., Ann Intern Med 2009

- My opinion: if patient brings you a report documenting coronary calcification (or LVH, ABI < 0.9), then consider them “high risk”

Outline

- Update on Key Biomarkers:
  - C-Reactive Protein (CRP)
  - Coronary calcification by CT and other subclinical CVD measures
  - Cystatin C
**Question 4: Regarding cystatin C...**

1. I first heard of it when I read the syllabus.
2. I saw paper(s) about it but have not read them.
3. I know it's a kidney test, but I don't use it.
4. I have measured cystatin C.

---

**What is Cystatin C?**

- Cystatin C is a blood test of kidney function.
- FDA approved as an alternate measure of kidney function.
- Marketed by several companies, and is rapidly becoming commercially available.

---

**Filtration Marker Principles**

- Creatinine and cystatin C are filtration markers
- Filtration marker-refers to something measurable in the blood that gives indirect information about the glomerular filtration rate (GFR)
- Production and elimination
- Factors affecting creatinine production:
  - Muscle, activity, diet, health status
- Factors we use to estimate creatinine production:
  - Age, sex, race, weight
Properties of Cystatin C

- Cystatin C – produced in all nucleated cells
- Constant release into blood, perhaps via cell turnover
- Freely filtered at glomerulus
- No renal tubular secretion
- Cystatin C association with GFR relatively independent of age, sex, race and muscle mass

How can we compare tests of Kidney Function?

- GFR – the "virtual" Gold Standard
  - Measured by clearance of injected tracer
  - Rarely clinically available
  - Never available for population based cohort studies
- Our strategy- use clinical outcomes associated with kidney disease
- If cystatin C is better predictor of death or CVD, then probably a better marker of kidney function

Comparison with Measured GFR

- 20 years of studies show cystatin C has closer approximation to measured GFR than creatinine
  - DharnidharkaVR et al. AJKD 2002
- Nearly all studies were in patients with kidney disease
- No GFR studies in the general population
Cardiovascular Health Study (CHS)

- Design: NIH sponsored cohort
- Subjects:
  - Population based elderly cohort from 4 US communities
  - N = 4,444
  - Mean age: 75 years
  - Cystatin C measured at 1992-93 (Yr 3) visit
- Mean follow up: 9 years
- Outcomes:
  - All cause mortality
  - Cardiovascular mortality
  - Heart failure

Creatinine Quintiles and Mortality Risk

Cystatin C Quintiles and Mortality Risk

*Significant adjusted hazard ratio

Creatinine and Cardiovascular Mortality

Cystatin C and Cardiovascular Mortality

Kidney Function and Heart Failure Incidence
**Confirmation Studies**

<table>
<thead>
<tr>
<th>Cohort/Study</th>
<th>Population</th>
<th>Outcome(s)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Health ABC</td>
<td>Elderly</td>
<td>Death, heart failure</td>
</tr>
<tr>
<td>Shlipak et al., JASN 2006</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Kidney-Derived, Arch Intern Med 2006</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Heart and Soul</td>
<td>CAD</td>
<td>Recurrent CVD</td>
</tr>
<tr>
<td>Ix, Circulation 2007</td>
<td></td>
<td></td>
</tr>
<tr>
<td>NHANES III</td>
<td>General population</td>
<td>Death</td>
</tr>
<tr>
<td>Ante, JASN 2009</td>
<td></td>
<td></td>
</tr>
<tr>
<td>MDRD Study</td>
<td>Advanced CKD</td>
<td>Death</td>
</tr>
<tr>
<td>Meox, Arch Intern Med 2007</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**What Clinical Need could Cystatin C Fill?**

- Predicting risk: probably inadequate to justify ordering a new test (see CRP)
- However, 10s of millions of creatinine tests ordered each year
- Potential roles for cystatin C:
  - Better classification of CKD
  - Identifying preclinical impairment of kidney function
  - Targeting specific populations with unpredictable muscle mass
  - Pre-op risk stratification before surgery or contrast

**CKD Definition by Creatinine and Cystatin C**

- In early work, we considered creatinine insensitive, but specific.
- However, from the nephrologist standpoint, specificity is a problem.
- Among 16 million in the U.S. with eGFRcreat <60, who has “real CKD”?  
- We compared prognostic value of CKD defined by creatinine and/or cystatin C

Peralta CA et al., J Am Soc Nephrol, in press 2010
**CKD Definitions in the Elderly**

*Cardiovascular Health Study- mean age 72*

<table>
<thead>
<tr>
<th>CKD definition</th>
<th>Rate (1000 person years)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Neither</td>
<td>20</td>
</tr>
<tr>
<td>Cystatin C</td>
<td>40</td>
</tr>
<tr>
<td>Creatinine</td>
<td>60</td>
</tr>
<tr>
<td>Both</td>
<td>80</td>
</tr>
</tbody>
</table>

**CHS: CKD and Mortality**

**All Cause Mortality**

<table>
<thead>
<tr>
<th></th>
<th>Multivariate Adjusted Hazard Ratios with 95% Confidence Intervals</th>
</tr>
</thead>
<tbody>
<tr>
<td>No Decreased eGFR</td>
<td>1.00 (ref)</td>
</tr>
<tr>
<td>Decreased eGFRcr only</td>
<td>1.09 (0.98, 1.21)</td>
</tr>
<tr>
<td>Decreased eGFRcys only</td>
<td>1.78 (1.53, 2.08)</td>
</tr>
<tr>
<td>Decreased eGFR both</td>
<td>1.74 (1.58, 1.93)</td>
</tr>
</tbody>
</table>

**CHS: CKD and Cardiovascular Disease**

<table>
<thead>
<tr>
<th>Cardiovascular Disease</th>
<th>Multivariate Adjusted Hazard Ratios with 95% Confidence Intervals</th>
</tr>
</thead>
<tbody>
<tr>
<td>No Decreased eGFR</td>
<td>1.00 (ref)</td>
</tr>
<tr>
<td>Decreased eGFRcr only</td>
<td>1.05 (0.92, 1.20)</td>
</tr>
<tr>
<td>Decreased eGFRcys only</td>
<td>1.52 (1.26, 1.84)</td>
</tr>
<tr>
<td>Decreased eGFR both</td>
<td>1.46 (1.29, 1.65)</td>
</tr>
</tbody>
</table>
**CHS: CKD and Heart Failure**

<table>
<thead>
<tr>
<th>Heart Failure</th>
<th>Multivariate Adjusted Hazard Ratios with 95% Confidence Intervals</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1407 events</td>
</tr>
<tr>
<td>No Decreased eGFR</td>
<td>1.00 (ref)</td>
</tr>
<tr>
<td>Decreased eGFRcr only</td>
<td>0.99 (0.84, 1.18)</td>
</tr>
<tr>
<td>Decreased eGFRcys only</td>
<td>1.69 (1.33, 2.13)</td>
</tr>
<tr>
<td>Decreased eGFR both</td>
<td>1.43 (1.22, 1.67)</td>
</tr>
</tbody>
</table>

**CHS: CKD and ESRD**

<table>
<thead>
<tr>
<th>Kidney Failure</th>
<th>Multivariate Adjusted Hazard Ratios with 95% Confidence Intervals</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>84 events</td>
</tr>
<tr>
<td>No Decreased eGFR</td>
<td>1.00 (ref)</td>
</tr>
<tr>
<td>Decreased eGFRcr only</td>
<td>2.60 (1.00, 6.75)</td>
</tr>
<tr>
<td>Decreased eGFRcys only</td>
<td>6.14 (2.18, 17.29)</td>
</tr>
<tr>
<td>Decreased eGFR both</td>
<td>23.82 (12.68, 44.76)</td>
</tr>
</tbody>
</table>

**CKD Definitions in the Middle-Aged**

*Multi-Ethnic Study of Atherosclerosis- mean age ~60*

- CKD: 30% with "real CKD"
Summary on CKD Classification

- A large proportion of CKD defined by creatinine are either “false positives” or “low risk” CKD.
- Cystatin C distinguishes the high risk CKD and identifies additional CKD missed by creatinine.

Practical Issues for Cystatin C

- Upcoming CKD Guidelines will discuss the clinical role of cystatin C.
- Currently, limited availability of cystatin C because earlier method requires special machine.
- At UCSF, has been available as a “send out” test.
- New cystatin C assays -can be run on nearly all clinical chemistry analyzers.

Costs:
- Medicare reimbursement: $19
- Assay costs: $6/test
- Lab charges: variable

Thank You

Questions?